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Collaborative Development of Recommendations for Euthanasia of Stranded Cetaceans

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U.S. Department of Commerce
National Oceanic and Atmospheric Administration
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NOTICE

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Executive Summary

The safe and humane euthanasia of live stranded cetaceans is one of the most challenging of all marine mammal stranding events. The experiences of the Virginia Aquarium Stranding Response Program (VAQS) and those of our colleagues in the United States Marine Mammal Stranding Network, as well as the lack of information and guidance available, inspired this important two year collaborative John H. Prescott grant project. We believe this project has been effective in accomplishing our goal of developing recommendations for stranding networks to facilitate the euthanasia of stranded cetaceans when release or rehabilitation is not an option. In order to accomplish this goal, we completed a number of targeted objectives. We developed standardized methods of data collection for cetacean euthanasia events, collected pertinent historic data from stranding networks nationwide, and compiled a review of gray and peer-reviewed published euthanasia literature. We established a cetacean euthanasia working group of experienced veterinarians, marine biologists and stranding response personnel and convened a three day workshop during the fall of 2011. To ensure we addressed critical issues and concerns from the national stranding network during the workshop, we developed and distributed a cetacean euthanasia questionnaire, using resultant information to assist us in preparation of the workshop agenda. The working group reviewed all collected information and developed the proposed recommendations.

Overall, we received great support and participation from marine mammal stranding network members. This was demonstrated by responses to the network survey, assistance with the data collection process, and comments we received from our project presentations and other informal feedback. In general, members of the stranding network, veterinary medical staff, biologists and stranding personnel understand the need for and the complexities of the euthanasia process and appear to want more guidance and training from National Oceanic Atmospheric Administration (NOAA) and experienced colleagues. Although the data compiled from historical cetacean euthanasia events was far from complete, we were able to summarize the information into tables of commonly used drugs and doses. Perhaps the most effective part of the project was the euthanasia workshop held in Virginia Beach, Virginia, in October 2011. The Cetacean Euthanasia Working Group (CEWG) participants were extremely engaged and very open to discussing euthanasia events, successful and unsuccessful outcomes, and concerns and needs of the stranding network. All working group members provided valuable input and worked effectively to develop recommendations and identify information and/or technology needs regarding cetacean euthanasia. While this project could not directly address all of these needs, we were able to develop the following informational materials:

- General and safety recommendations for stranded cetacean euthanasia
- Euthanasia options matrices for large and small cetaceans
- Summary of effective euthanasia methods

- Species-specific length-weight equations and graphs
- Species-specific observations for stranded cetacean euthanasia
- Common hypodermic needle lengths and injection routes
- Aggregate historic drug doses for commonly encountered species
- Illustrations for ballistics, injection and exsanguinations of small cetaceans
- Cetacean euthanasia record and instructions for standardized recording of euthanasia events

The above materials are collectively included in Appendix 1. We developed and compiled this information to be used as a stand-alone document with the hope that it will aid responders during cetacean euthanasia events.

Finally, this project has only begun to address the needs and challenges faced by the marine mammal stranding community when responding to live stranded cetaceans where euthanasia has been determined to be the final outcome. While the decision to euthanize will likely always be contentious, we should continue to work to make the process itself humane, reliable and safe when it needs to be performed. We at VAQS, and the entire CEWG, hope that this work is continued by NOAA and other colleagues and that it will evolve as we learn from future experiences.

Introduction

One of the most challenging and potentially dangerous of all marine mammal stranding events is the euthanasia of a cetacean. Considerations include the welfare of the animal, personnel safety and eco-toxicological hazards as well as the availability of appropriately trained and licensed individuals. To complicate this issue, these events often attract, and are played out, under a great deal of public scrutiny and media attention. To date, no systematic survey of protocols, methods and chemicals employed in the euthanasia of stranded cetaceans has been published in the United States, and recommended guidelines (if available) are often insufficient and sometimes contradictory.

As a member of the national marine mammal stranding network, the Virginia Aquarium Stranding Response Program (VAQS) conducts work under the authority of a Marine Mammal Stranding Agreement (SA) with National Oceanic and Atmospheric Administration's National Marine Fisheries Service (NOAA/NMFS). The SA accepts the definition of "humane" as it appears in the Marine Mammal Protection Act (1972), and is defined as involving the least possible degree of pain and suffering practicable to the marine mammal involved. The SA also states that we, as network participants, shall only perform euthanasia following guidelines such as those recommended by the American Veterinary Medical Association (AVMA) Panel on Euthanasia. According to the AVMA (2007, 2013) guidelines, euthanasia is the act of inducing humane death in an animal. These guidelines also acknowledge that "there are situations involving free-ranging wildlife when euthanasia is not possible from the animal or human safety standpoint, and killing may be necessary". Conditions found in the field, although more challenging than those that are controlled, do not in any way reduce or minimize the ethical obligation of the responsible individual to reduce pain and distress to the greatest extent possible during the taking of an animal's life. It is our responsibility as veterinarians and human beings to ensure that if an animal's life is to be taken, it is done with the highest degree of respect, and with an emphasis on making the death as painless and distress free as possible. Euthanasia techniques should result in rapid loss of consciousness followed by cardiac or respiratory arrest and the ultimate loss of brain function. In addition, the techniques should minimize distress and anxiety experienced by the animals prior to loss of consciousness.

VAQS trained personnel have participated in the euthanasia of more than fifty live stranded cetaceans since 1998. These events have included 14 different species and involved single and mass stranding events, dependent calves and three species of baleen whales, as well as a sperm whale. We have encountered numerous difficulties when attempting to achieve humane euthanasia as defined by the AVMA due to the complexity and diversity of species and stranding situations. There is little published information on cetacean euthanasia and the available recommended guidelines are broad and difficult to interpret for stranding response personnel.

Intravenous administration of an acceptable chemical agent, as defined in AVMA guidelines, is considered the most rapid and reliable means of obtaining humane euthanasia in mammals (AVMA 2013). We have found that the logistics of live cetacean stranding response – for example: animal(s) in surf; animals thrashing their bodies and fluking (thrashing of flukes); availability of specialized needles required to access intramuscular or intracardiac injection sites in large cetaceans and likelihood of vessel collapse – often prevent safe intravenous access. In addition, personal communications with other stranding response organizations, combined with our own experience, has demonstrated that the sole use of intravenous barbiturates can result in animal(s) exhibiting excitatory reactions, including violent spinning and fluking, which can place responders and onlookers at risk of physical injury and present an unsatisfactory perception of the euthanasia process.

In addition to general concerns about euthanasia in cetaceans, we and our colleagues have observed species-specific sensitivity/reactions to the administration of certain drugs. Xylazine is one example. A literature review resulted in limited and conflicting data regarding the use of this drug with cetaceans. The CRC Marine Mammal Medicine Handbook, 2nd Edition (Dierauf and Gulland 2001) cautioned against the use of xylazine in baleen whales due to an excitatory reaction exhibited by a gray whale (*Eschrichtius robustus*), while Daoust and Ortenburger (2001) observed no adverse response when they administered the drug to a juvenile fin whale (*Balaenoptera physalus*). We also administered xylazine to a debilitated juvenile humpback whale (*Megaptera novaeangliae*; VAQS20061066) with no adverse reaction. In another example, NOAA personnel recommended that VAQS administer a pre-euthanasia injection of acepromazine to a stranded common dolphin (*Delphinus delphis*; VAQS20011045). The animal exhibited extreme and prolonged (>7 minutes) whole body convulsions along with retching and vocalizations after administration of the drug. Generalizing a single adverse reaction to an entire species is tenuous, but the violence of some reactions can leave such an indelible impression that these individual cases warrant consideration. This is particularly pertinent when drug options are limited, so that personnel can be prepared for the possibilities.

Many chemical agents acceptable for euthanasia are also controlled substances and can be dangerous, ultra-potent narcotics. Handling these drugs, often in very high doses and in field euthanasia events, makes safety a concern. These safety concerns, as well as cost and the need for appropriately licensed personnel and specialized equipment, can often make their use prohibitive. Specialized needles of appropriate length and gauge, and large volumes of chemical agents used by stranding responders, are not always available at small animal clinics that assist the networks with these events.

Eco-toxicological hazards resulting from residues of euthanasia agents, as well as the potential bioaccumulation of existing toxins in the animal's tissues and body fluids, are of major concern during field euthanasia events. Because of these concerns and National Environmental Policy

Act (NEPA) regulations, interest from federal officials in the use of physical euthanasia methods such as exsanguination and ballistics is understandably increasing (Tristan *pers. comm.*; Durham *pers. comm.*). In 2005, VAQS responded to a mass stranding of pilot whales involving nine live and 22 dead animals along more than 10 kilometers of beach on the outer banks of North Carolina (Hohn *et al.* 2006). NOAA personnel recommended exsanguination as the method of euthanasia to decrease the potential for environmental impact from chemical residues. Responders expressed concerns about whether exsanguination without sedation would meet the "humane euthanasia" standard; the lack of prior training; and the potential impact on responders and onlookers. NOAA ultimately permitted the use of chemical means to euthanize. This decision was contingent on the administration of only non-controlled substances and an emphasis on proper carcass disposal.

While the potential for environmental impact is an important consideration, it cannot be separated from the need for humane euthanasia of these animals. Current AVMA guidelines consider the use of several physical methods of euthanasia, including exsanguination, as inhumane unless performed on a heavily sedated, unconscious or moribund animal (AVMA 2007, 2013). While chemical sedation of managed cetacean populations is commonly practiced in controlled environments, sedation of stranded cetaceans is often difficult to achieve and can be dangerous. Well-organized and easily accessible information on cetacean euthanasia methods and outcomes is critically needed to assist stranding network personnel in making informed, ethical and environmentally responsible decisions.

The goal of this project was to compile published information and existing euthanasia data and to develop recommendations for stranding networks to facilitate the euthanasia of live stranded cetaceans once veterinary medical personnel have determined this to be the best option.

In order to achieve this goal we:

- Developed standardized methods of data collection for cetacean euthanasia events
- Performed an extensive literature search, including "gray-literature" (personal communications, medical records, stranding reports, government documents) and peer-reviewed publications and compiled pertinent information
- Compiled and analyzed data from historical euthanasia events to develop a cetacean euthanasia database
- Convened an expert advisory panel whose tasks were to:
    - critically examine euthanasia data
    - based on the literature, data and their experience, develop recommendations for safe and humane cetacean euthanasia and subsequent carcass disposal

The expert advisory panel became the basis for a Cetacean Euthanasia Working Group (CEWG), which added several interested and qualified parties to the original panel proposed in the project.

The CEWG included nationwide participants including: veterinarians experienced in stranding response and cetacean euthanasia; field response personnel from two different regions with high live cetacean stranding rates; and NOAA Marine Mammal Health and Stranding Response Program (MMHSRP) staff.

Literature Review

To date, no systematic survey of protocols employed in the euthanasia of stranded cetaceans has been published. Available published information on cetacean euthanasia and recommended guidelines are limited, insufficient, and sometimes contradictory. This Prescott project addressed this issue by compiling historic and contemporary cetacean euthanasia data through an extensive literature search and review of standing data and records. Subsequently, the assistance of experienced veterinary medical and field stranding personnel was enlisted to review this information and develop recommendations. These recommendations can be used to assist stranding personnel with the development of their own organization's protocols and determination of best practices for achieving the safest and most humane euthanasia of stranded cetaceans when this course of action is deemed necessary.

This document represents an extensive literature search, including peer-reviewed publications and "gray-literature" (personal communications, medical records, stranding reports, government documents). Taking into consideration the relatively limited amount of information available regarding cetacean euthanasia, we investigated alternative and potentially useful information that may be modified and applied to this effort. The following information is included: methods for cetacean sedation and anesthesia; whaling industry practices; and comparable means for euthanasia of companion animals, humans, wildlife, and livestock. Ultimately, less than 30 percent of the literature reviewed was deemed pertinent for inclusion in this summary.

The AVMA Panel Report on Euthanasia (AVMA 2007, and updated during the preparation of this report, AVMA 2013) is consistently referenced regarding the euthanasia of animals. The Recommendations for Euthanasia of Experimental Animals (Close et al.1996, 1997) is also commonly cited. These reports, along with other resources (Reilly and Blackshaw 2001, Longair *et al.* 1991, Carding 1977, Short and Woodnott 1969) include guidelines and contain a wealth of information regarding the humane euthanasia of animals. However, their main focus is domesticated and laboratory animals. Unfortunately, in the case of live cetaceans, these recommendations often do not adequately address the circumstances and additional problems associated with stranding events. For example, the inability to properly dispose of a large whale carcass can prohibit the use of many acceptable chemical agents due to eco-toxicological concerns. Published guidelines written specifically for cetaceans currently recommend allowing sperm and baleen whales to die a natural death rather than use substandard methods of euthanasia (Suisted 1999, Barnett et al.1998, Royal Society for the Prevention of Cruelty to Animals 1992). These animals may however, linger for days without human intervention (Hyne 1981). Taking this into consideration, it is now recognized by AVMA (2007, 2013) that, although available methods may not be "acceptable" according to current guidelines, they may be more humane than leaving an animal to suffer for extended periods of time. With the culmination of this project, we plan to provide the best possible recommendations to help facilitate the most humane and efficient death possible for all cetaceans when euthanasia has been determined to be the best option.

The most comprehensive resources we investigated regarding the euthanasia of stranded cetaceans include:

- CRC Handbook of Marine Mammal Medicine, 2nd Edition (Dierauf and Gulland 2001)
- American Association of Zoo Veterinarians (AAZV), Guidelines for the Euthanasia of Nondomestic Animals (Baer 2006)
- Marine Mammals Ashore: A Field Guide for Strandings, 2nd Edition (Geraci and Lounsbury 2005)
- Royal Society for the Prevention of Cruelty to Animals (RSPCA), Stranded Cetaceans: Guidelines for Veterinary Surgeons (RSPCA 1997)
- The British Divers Marine Life Rescue: The Marine Mammal Medic Handbook (Barnett et al.1998)
- New Zealand Department of Conservation – Marine Mammal Stranding Contingency Plans, Standard Operating Procedure (Suisted 1999)
- Australian Veterinary Association – Guidelines on humane slaughter and euthanasia (Australian Veterinary Association 1987)

Although recommendations vary and a number of different methods are used worldwide, ultimately the consistent goal in all sources is to render the animal dead as swiftly and painlessly as possible.

Methods of Euthanasia

Euthanasia methods are commonly classified into two main categories: chemical methods and physical methods. Chemical methods include: non-inhalant agents (i.e., injectable) and inhalant agents that include anesthetic gases such as halothane, methoxyflurane, and isoflurane. Physical methods of euthanasia include ballistics, explosives and exsanguination.

Chemical Methods of Euthanasia

Injectable agents used for the euthanasia of cetaceans are presently considered among the most effective and humane (AVMA 2007, 2013, RSPCA 1992). NOAA Minimum Standards for Euthanasia (Gage 2008) include the use of appropriate drugs and doses with references to three resources: 1) AVMA Panel Report on Euthanasia (2007, 2013); 2) CRC Handbook of Marine Mammal Medicine; and 3) American Association of Zoo Veterinarians Guidelines for the Euthanasia of Nondomestic Animals. AVMA (2007, 2013) refers to the use of acceptable chemical agents, primarily etorphine and pentobarbital, for the euthanasia of marine mammals (Greer and Rowles 2000). Greer *et al*. (2001) recommends the sedation of marine mammals prior to euthanasia in order to decrease the risk of serious injury to handlers.

Despite these recommendations, no drugs used for sedation or anesthesia are actually labeled for use in marine mammals. Published material for potential chemical agents lists a relatively small number of animals involving a narrow range of captive species. Even less research has been done on species-specific euthanasia in wild marine mammals. Most acceptable euthanasia agents are controlled substances, often ultra-potent narcotics, frequently requiring massive quantities to deliver effective doses for large cetaceans. Hyper-concentrating certain drugs, in order to decrease the volume of agent required, has been used in efforts to sedate North Atlantic right whales (*Eubalaena glacialis*) prior to disentanglement (Moore *et al.* 2010). However, there is little information regarding the actual bioavailability of these concentrations (Bogomolni 2006) which may pose significant risk to handlers, personnel in the immediate vicinity, and, if introduced into the environment, risks to the general public (Harrington 2010, Kotz 2010). The use of any drug in the United States in an “extra-label” manner is governed by the Animal Medical Drug Clarification Use Act of 1994 (AMDUCA). This law states that all extra-label drug use must be administered on the order of a veterinarian, within the context of a valid veterinarian-client-patient relationship, and not result in drug residues or contamination of food resources.

Other complicating factors in regard to chemical agents for cetacean euthanasia are that the cost and the requirement for appropriately licensed personnel often limit their availability and their use. Individual states and veterinarians often have different interpretations of Drug Enforcement Agency (DEA) regulations and guidelines regarding controlled drug use. Although the act of euthanizing a marine mammal does not require licensure, availability and use of the most common chemical agents to conduct the procedure requires a DEA license and often a state pharmaceutical license. Veterinarians, particularly those whose licenses are tied to a private practice, are understandably concerned about the use of large quantities of controlled drugs in the field and/or by non-licensed personnel. Many veterinarians who work with stranding response organizations are also in private practice and are not available to go into the field on short notice because of responsibilities to their clients. It is these dedicated veterinarians, and the stranding responders they work with, who must walk a fine line between humane treatment of a stranded animal and strict adherence to regulations involving controlled drugs needed to perform euthanasia.

Personnel safety issues (Mazet *et al.* 2004) and eco-toxicological effects (Otten 2001) must also be considered. In addition, drugs required for the euthanasia of large whales or of mass stranded animals may not be stocked locally in quantities and concentrations necessary for the task.

<u>Ecotoxicological Concerns of Chemical Euthanasia</u>

Many drugs used in sedation and euthanasia can be responsible for secondary toxicosis and environmental pollution (O'Rourke 2002, Otten 2001, O'Connor *et al.* 1985, Bischoff *et al.* 2011, Harms *et al*. 2014). Deaths of birds and mammals by secondary pentobarbital poisoning have been reported in over 16 states according to the United States Geologic Survey's National Wildlife Health Center in Madison, Wisconsin (www.nwhc.usgs.gov). Depending on the effect, both criminal and civil implications of exposure to these medications must be considered. Several incidents have previously assigned veterinarians with partial culpability for relay toxicosis of endangered raptors by sodium pentobarbital in carcasses of euthanized terrestrial animals (Otten 2001, O'Rourke 2002), and a nonfatal case of suspected relay toxicity to a pet dog from tissues of a euthanized whale carcass has been reported (Bischoff et al. 2011). Veterinarians and individuals from stranding response organizations are concerned about these issues and also about potential liability for any adverse effects of drugs dispensed or used on their authority. Some of these agents may be environmentally labile, but others may persist and even bioaccumulate (Eckek 1993, O'Rourke 2002, Otten 2001). Euthanasia techniques excluding use of pentobarbital or ultra-potent narcotics may be employed to reduce, but not eliminate, ecotoxicology concerns (AVMA 2013, Harms *et al.* 2014). A combination of pre-euthanasia sedatives (midazolam, acepromazine, xylazine) followed by intracardiac injection of saturated KCl solution has been employed in euthanasia of mysticetes (Harms et al. 2014). Based on tissue drug residue analysis with this protocol, trimming and safe disposal of xylazine IM injection sites was recommended as prudent to minimize risk of relay toxicity even further, should full carcass disposal not be an option.

Once an animal is euthanized, the carcass can pose a disposal dilemma. Barriers to translocation of carcasses or burial in landfills can result in carcasses remaining on the beach or becoming exposed after beach burial. Some municipalities accept euthanized cetaceans in landfills and others do not. Some rendering companies will accept euthanized carcasses, but rendering does not necessarily degrade all toxic substances and products of the rendering process have been used in pet and domestic animal food, as well as in fertilizers. Having a frank discussion with local officials and the board of health in the town where euthanasia is planned, and disposal contemplated, is highly recommended. Recently the University of New England Marine Animal Rescue Center received a grant to research composting as a method of marine mammal carcass disposal (Matassa 2009). Preliminary results indicate that composting (aerobic decomposition) is preferable and more effective than anaerobic burial. It may also be a more effective and less expensive alternative to rendering and/or incineration.

These legitimate concerns can lead to conflicting recommendations when attempting to balance priorities of animal welfare and environmental responsibility. While the RSPCA (1997) states that the use of drugs is the most practical method of humanely killing any cetacean, the New Zealand Department of Conservation does not recommend the use of drugs due to issues regarding contaminated carcass disposal (Suisted 1999). Much more work is required to assist in determining the most appropriate and safe method(s) for carcass disposal.

<u>Routes of Administration of Chemical Methods of Euthanasia</u>

Intravenous (IV)

Ideally, intravenous administration of an acceptable chemical agent is considered the most rapid and reliable means of accomplishing a humane euthanasia in mammals (AVMA 2007, 2013, Close et al.1996, National Research Council 1992). Unfortunately, intravenous access is not always an option for cetaceans due to factors such as: logistics (i.e. animal washing in the surf); behavioral challenges such as the thrashing of the animal; the limited availability of specialized needles to access intramuscular or intracardiac injection sites in large cetaceans; the potential for vasoconstriction in debilitated animals and/or shunting of blood from peripheral vessels to the specialized rete mirabile due to the vascular system of diving mammals; and lack of training and experience of personnel in accessing multiple intravenous sites.

Intravenous access can be achieved via the superficial peripheral vessels located in the flukes, pectoral flippers and/or dorsal fin. The caudal peduncle vessels are situated deeper than the others and can be accessed laterally or ventrally. Accessibility of blood vessels also varies among species and individual animals. For example, vessels in the dorsal fin of the common dolphin (*Delphinus delphis*) are generally much easier to locate and access than in many other species (Walton unpublished data). Detailed descriptions and illustrations of blood vessels along with preferred needle size and gauge are available in several resources (Geraci and Lounsbury 2005, Greer et al.2001, RSPCA 1997).

Personal communications with other stranding response organizations including the New England Aquarium, National Marine Life Center, University of North Carolina at Wilmington, and Cape Cod Stranding Network, as well as our own experience, has demonstrated that the sole use of intravenous barbiturates without sedation can result in animals exhibiting severe excitatory reactions, including spinning and fluking, which can place responders and onlookers at risk of physical injury.

Intramuscular (IM)

Intramuscular injections in cetaceans can be administered into the large epaxial muscles, just lateral to the midline, either slightly anterior or posterior to the dorsal fin. Since blubber thickness tends to increase just parallel to these areas care must be taken to ensure proper placement of the needle into muscle tissue. McBain (2001) recommends caution to avoid the thoracic cavity when injecting anterior to the dorsal fin.

The intramuscular route is not recommended for sodium pentobarbital administration because the drug's acidic nature causes pain and severe muscle necrosis. Also, slow absorption results in slow and unpredictable advancement through anesthetic stages as well as variability in the effect of the agent (Clifford 1984). Consideration must be given when choosing lengths of needles to ensure penetration through the blubber and into the muscle. Injection into the blubber can greatly impact absorption rates and may result in the failure to absorb injected agents properly (Stoskopf et al. 2001). Besides appropriate needle size (length and gauge), it is also beneficial to use a needle fitted with a stylet to decrease the potential for plugs of blubber impacting the lumen of long, large gauge needles used in large ceteaceans (Walton unpublished data).

Intraperitoneal (IP)

When intravenous administration is considered impractical or impossible, intraperitoneal administration of non-irritating agents is acceptable by AVMA standards (2007, 2013). Although sodium pentobarbital is irritating to tissues when administered perivascularly (Adams 2001), studies have indicated that the addition of lidocaine during intraperitoneal administration may result in a decrease in nociception compared to the use of sodium pentobarbital when used alone (Svendsen et al. 2007). This combination has been shown to decrease agonal gasping that can occur when euthanizing dogs and cats with sodium pentobarbital alone (Evans et al. 1993). The combination of pentobarbital and phenytoin (i.e. Euthasol®) however, may be unsuitable for intraperitoneal injection because of concerns over the different absorption rates of the two compounds. The effects of phenytoin on the heart may occur before the pentobarbital component has caused unconsciousness (Sinclair 2004).

Intraperitoneal injections are administered on the ventral midline just caudal to the umbilicus or on the right side of the abdomen. Administration into the left lateral aspect may result in injection into the stomach, which may delay and decrease uptake due to digestion. The cetacean peritoneal cavity is tightly compacted and inadvertent perforation and injection into an organ or the intestinal tract should be considered. A higher dose of barbiturate is required for intraperitoneal euthanasia (Geraci and Lounsbury 2005, Grier and Schaffer 1990).

Intrahepatic

While intrahepatic administration is not considered an acceptable method (not considered humane to use alone in conscious animal) of administration by the AVMA (2007, 2013), studies have found potential benefits to intrahepatic injections of euthanasia agents in shelter cats including less reaction to the injection when compared to intraperitoneal routes (Grier and Schaffer 1990). When correctly administered, intrahepatic injections achieve considerably faster onset in comparison to the IP route. However, accuracy is difficult and this technique may cause discomfort (Sinclair 2004). The liver is located ventrally and can be accessed ventrolaterally, caudal to the heart, at the level of the distal pectoral fins when they are folded against the body wall. More precise localization of the liver is possible by use of portable ultrasound machines.

As with intramuscular injections, the appropriate length of needle and the presence of a stylet are important to ensure effective intraperitoneal and/or intrahepatic administration. Greer et al. (2001) suggests that these routes may be most appropriate for smaller animals considering the difficulties associated with the thickness of skin, blubber and muscle in the large mysticete whales.

Intranasal/Blowhole

Preliminary studies regarding absorption of various drugs via nasal mucosa indicate this as a viable route of administration (Hirai et al.1981). Intranasal administration of several agents including midazolam, morphine, and xylazine in humans (Wermeling 2009, Ilium 2002) and game animals (Cattet et al. 2004) have all been effective. Sedation after intranasal administration of xylazine in elk often occurred in less than one minute post administration (Klossek et al. 2001). In regards to cetaceans, we identified one reference to a case that administered 60 mls of pentobarbital to a live stranded fin whale *(Balaenoptera physalus)* via the blowhole. Although upon expiration the animal expelled the 30” venous extension set used for administration, only a small amount of the pentobarbital solution was expired. Sedation occurred within 20 minutes post administration allowing the safe use of a fluke vessel for final euthanasia (Dunn 2006). These preliminary findings indicate that this is a potentially viable and advantageous alternative route that needs to be explored more thoroughly.

Intraglossal

While we found little published information on intraglossal injections, the tongue is a uniformly lean and richly vascular muscle bed (Miller and Zawistowski 2004). Sullivan et al. (1997) concluded that intraglossal injection of anesthetic and/or analgesic medications in children resulted in more rapid onset than injection at more traditional intramuscular sites. Access to the thicker areas of the tongue is difficult in small odontocetes and would require restraint. In larger odontocetes and mysticetes, it would be difficult and potentially dangerous to gain access to this area.

Sublingual injection

A sublingual vein is located on each lateral aspect of the ventral surface of the tongue. They are extremely superficial. Reports regarding laboratory animals (Waynforth 1980) and dogs (Miller and Zawistowski 2004) suggest that these vessels can be entered easily and the formation of hematomas is rare, even after repeated usage. Ordog et al. (1984) compared absorption between peripheral and sublingual vessel injections in goats. They found significantly lower blood levels after sublingual injection compared to peripheral intravenous routes. In conclusion, Ordog recommended that if a peripheral IV line is not accessible, vigorous attempts should be made to start a central line rather than wasting valuable time trying to inject sublingually. Access to these vessels may, however, be problematic in small odontocetes and dangerous in larger odontocetes and mysticetes.

Intraoral/Buccal

Oral uptake of agents primarily through the buccal mucous membranes may take 30 to 90 minutes and is highly variable (Ramsay and Wetzel 1998). While this method may be unsuitable for euthanasia, it may be more appropriate for sedation prior to euthanasia (Sinclair 2004, Ramsay and Wetzel 1998). The use of opioid medications for pain relief, such as buprenorphine, in buccal applications in cats is common, well tolerated, and effective (Robertson and Taylor 2004). Future studies of the buccal route in cetaceans may prove to be beneficial.

Retro-bulbar

Blood sampling from the retro-bulbar venous plexus is one of the most widely used sampling methods in laboratory animals (Mahl et al. 2000). It is recommended for use only in anesthetized animals (Diehl et al. 2001). This route was utilized in the case of a right whale calf where recovery and rescue was not an option, using specially designed long needles (Harms et al. 2014). The animal had been stranded for two days on a remote shoal with limited accessibility and, despite exhibiting compromised respirations and severe sunburn, continued to remain alert and active. Taking into consideration the extenuating circumstances, the high vascularity and close proximity to the brain of this route, and distance from the more hazardous working area near the flukes, made it a feasible option. Although the ultimate effectiveness of the sedation was questionable, this appears to be a potentially viable route of administration in exceptional situations and deserves further investigation.

Intracardiac (IC)

We found little published data regarding intracardiac injections and this method is soundly discouraged as a sole method of euthanasia in clinical situations. Attempting to accurately inject directly into a heart chamber is difficult (Adams 2001). Advantages with this route for cetaceans in the field are that it keeps personnel in a safer physical position to administer agents and, if the peripheral vessels are collapsed, it results in a more rapid onset of action (Baer 2006). Personal experience with small cetaceans and that of other stranding responders such as New England Aquarium (Merigo pers. comm.) and International Fund for Animal Welfare Marine Mammal Rescue (Touhey pers. comm.) has shown this method to be effective, although excitatory responses similar to IV injection are common in conscious animals. Whenever possible intracardiac administration of medication should only be performed in sedated, anesthetized, or moribund animals. The heart may be accessed from the right or left axillary space, or a parasternal approach may be used.

Intrathoracic, Intrapulmonary, Intrathecal

Throughout our research we found no specific references to intrathoracic, intrapulmonary or intrathecal methods of euthanasia administration. These methods and routes were considered unacceptable due to prolonged onset of action and potential to produce coughing, epistaxis and haemoptysis due to increased pressure and lacerations of vessels within the thoracic cavity particularly when intrathoracic and intrapulmonary methods are used (Simmons and Brick 1970, Strande 1964).

Other considerations

Most guidelines for euthanasia are developed by and refer to clinic or laboratory settings. These sources assume that the animal to be euthanized is easily accessible and/or relatively small. Although these references consider other injectable routes of euthanasia (i.e. intracardiac, intrathoracic, intrapulmonary) unacceptable in a conscious animal due to pain and unpredictability (AVMA 2007, 2013, Baer 2006, Close et al. 1997), it may be necessary to investigate and discuss these routes due to the often extraordinary circumstances associated with cetacean strandings. The alternative of letting nature take its course may result in a prolonged dying process, from hours to days for cetaceans, especially large whales.

Types of Chemical Agents for Euthanasia

When used alone, certain chemical agents (strychnine, nicotine, caffeine, magnesium sulfate, potassium chloride, cleaning agents, solvents, disinfectants and other toxins or salts, and all neuromuscular blocking agents) are considered unacceptable and consistently condemned for use on conscious animals as sole euthanasia agents due to the infliction of pain and stress to the animal (Gage 2006). Thus, consideration of appropriate sedatives for animal comfort, and tranquilizers for responder safety, must be part of any discussion on cetacean euthanasia. Below is a listing of drugs that may be used alone or in combination to euthanize a cetacean with referenced observations and recommendations when used on a variety of animals:

Pre-euthanasia drugs

The administration of pre-euthanasia drugs is recommended in multiple sources (AVMA 2007, 2013, Baer 2006, Close et al. 1997 and 1996, Harms et al. 2014, Kreeger 2007). This method is required to ensure the animal is rendered insensible to pain when conditionally acceptable euthanasia practices are used. Greer et al. (2001) recommends the sedation of marine mammals prior to euthanasia in order to decrease the risk of serious injury to handlers. Tranquilizers, sedatives and some injectable anesthetics are commonly used as pre-euthanasia drugs, but in a severely debilitated animal, they may suffice to effect euthanasia.

Several types of chemical agents are used prior to euthanasia and are described below. It is important that personnel become familiar with appropriate drugs to ensure desired effect. For example, although the use of tranquillizers will calm the animal, they may have no analgesic effects and the animal is often rousable. There is a relatively small amount of information regarding the use of anesthetic agents in marine mammals (Haulena and Heath 2001, Reidarson 2003). Ultimately the investigation and/or use of a combination of at least two to three chemical agents appears to be the most effective and consistently recommended method for humane euthanasia.

Below are some examples of pre-euthanasia combinations from the literature search:

- A sequential combination of midazolam, acepromazine, and xylazine has provided satisfactory sedation prior to administration of saturated KCl solution in mysticetes (Harms et al. 2014).
- A combination of medetomidine and ketamine has provided satisfactory sedation prior to IV administration of barbiturates in cetaceans (Greer et al. 2001).
- After the successful euthanasia of a fin whale, Daoust and Ortenburger (2001) believed that an IM method for large whale euthanasia would be optimal and include Immobilon® (no longer available, but in its place he recommended combination of α_2 agonist [detomidine, medetomidine] and synthetic opioids [carfentanil]). He also states that there is little information available regarding this combination and that cost and availability may be prohibitive.

- Tranquilizers and Sedatives
  Tranquilization reduces anxiety and induces a sense of tranquility without drowsiness. Drug-induced sedation has a more profound effect and produces drowsiness and

hypnosis. Analgesia is the reduction of pain, which depending on a drug's effect, may be more pronounced in either the viscera or the musculoskeletal system. Many drugs cannot be categorized by only one pharmacologic effect (*i.e.* as tranquilizers, sedatives, or analgesics). For example, many psychotropic drugs can either tranquilize or sedate according to the dose administered, and many sedatives are also analgesics. Also, drugs classified as tranquilizers, sedatives, and/or analgesics may have additional effects (*e.g.* behavioral modification, antiemesis (Kahn 2010).

- Benzodiazepines
  Ex: diazepam, midazolam
  - Controlled substances (Schedule IV)
  - IM, IV, SC, PO
  - Benzodiazepines are used for a variety of indications (anxiolytic, muscle relaxant, hypnotic, appetite stimulant, and anticonvulsant) in several species (Plumb 2015)
  - May be used as a sole agent to calm an animal (Fowler 1986) and as an anticonvulsant (Plumb 2015), or in combination with other agents such as ketamine (Haulena and Heath 2001) to potentiate sedation
  - Diazepam is slowly and incompletely absorbed following IM injection (Plumb 2015), but it can be used with some success by the IM route (Sepulveda et al. 1994, Fowler 1986)
  - Companion animal veterinary practices commonly use diazepam as an IV agent, administered slowly, and midazolam in situations requiring IM administration (Plumb 2015)
  - The Virginia Aquarium Stranding Response Program (VAQS) has had successful and rapid (<10 minutes) results after pre-euthanasia administration of diazepam IM in several species (*Grampus griseus*, *Delphinus delphis, Lagenorhynchus acutus*). In these cases, it facilitated an uneventful euthanasia with no adverse reaction to administration method (VAQS unpublished data)
  - Greer et al. (2001) reported IM midazolam as a pre-euthanasia sedative in an emaciated juvenile gray whale (*Eschrichtius robustus*). Initial onset was noted within 12 minutes post-administration and after 29 minutes the peduncle could be lifted with no response. The animal was euthanized 38 minutes after injection (Bogomolni et al. 2006)
  - Harms et al. (2014) used IM midazolam successfully as part of a pre-euthanasia combination (with acepromazine and xylazine) in mysticetes.

- Phenothiazines
  Ex: acepromazine, chlorpromazine, promethazine
  - Non-controlled substances
  - IM, IV, SC, PO

  - Negligible analgesic effects (Plumb 2015)
  - Used as anti-emetic (Plumb 2015)
  - Potentiate anesthetic effects; may decrease side effects of other drugs (Kreeger 2007)
  - When combined with etorphine (M99), acepromazine decreased hyper-excitability and hyperventilation during the immobilization of hoof stock (Wolfe et al. 2004, Machado et al. 1983)
  - Large animal Immobilon® is acepromazine combined with etorphine and was previously a drug of choice for cetaceans in the UK (RSPCA 1992). It is not available in the United States
  - The IM administration of acepromazine as a pre-euthanasia sedative to a common dolphin *(Delphinus delphis)* resulted in extreme and prolonged (>7 minutes) whole body convulsions along with retching and vocalizations (Walton unpublished data)
  - Responders in North Carolina have successfully used acepromazine in combination with xylazine to induce heavy sedation leading to death before barbiturates or other euthanasia agent would have been administered. The pre-euthanasia combination has resulted in acceptable euthanasia in several cetacean species, including harbor porpoise and bottlenose dolphin (Harms unpublished data)
  - Routine clinical use of acepromazine for sedation in marine mammals is discouraged but may be appropriate as a pre-euthanasia tranquilizer (Dunn pers. comm.)

- Alpha$_2$-Adrenoceptor Agonists
  Ex: xylazine, romifidine, detomidine, medetomidine, dexmedetomidine
  - Non-controlled substances
  - IM, IV, SC
  - Used for sedative and analgesic properties in a variety of species (Plumb 2015)
  - Animals may be more prone to sudden movements when immobilized with alpha$_2$-adrenoceptor agonists (Kreeger 2007)
  - May cause vomiting in canids and felids (Plumb 2015)
  - Xylazine is classified as a sedative/analgesic, which although it has muscle relaxant properties may also cause muscle tremors (Plumb 2015)
  - Retching and severe muscle tremors and fluking noted when xylazine administered IM in two Risso's dolphin (*Grampus griseus*) (Walton unpublished data)
  - Greer et al. (2001) cautioned against the use of xylazine in baleen whales due to a severe reaction exhibited by one gray whale *(Eschrichtius robustus)*, while Daoust and Ortenburger (2001) observed no adverse response when they

administered the drug to a juvenile fin whale *(Balaenoptera physalus)*. VAQS also administered xylazine to a debilitated juvenile humpback whale *(Megaptera novaeangliae)* with no adverse reaction (VAQS unpublished data). Harms et al. (2014) observed no adverse response to xylazine when preceded by midazolam and acepromazine in one right, one minke, and three humpback whales.

  - VAQS has administered pre-euthanasia xylazine IM and successfully achieved a deep sedation leading to death with pygmy sperm whales *(Kogia breviceps)* and bottlenose dolphins (*Tursiops truncatus*) (VAQS unpublished data)

- Opioids as pre-euthanasia drugs

  Ex: butorphanol, meperidine
  - Animals may be more prone to sudden movements when immobilized with opioids. Kreeger (2007) recommends the concurrent use of tranquillizers to decrease excitation response and hasten induction.
  - Controlled substances (Schedule II-IV)
  - IM, IV, SC, PO (buccal)
  - Analgesic properties

  Butorphanol
  - Controlled substance (Schedule IV)
  - IM, IV
  - Partial opioid agonist/antagonist used in a variety of species as an analgesic or premedication (Plumb 2015)
  - Preliminary findings with butorphanol in the sedation of cetaceans appear effective when midazolam is insufficient for procedures. IM = 20-30 minute onset (Chittick et al. 2006)
  - Combined with xylazine, the combination provides a deep sedation in wildlife (Kreeger et al. 1989)

  Meperidine
  - Controlled substance (Schedule II)
  - IM, slow IV, irritating SC
  - Opiate analgesic (Plumb 2015)
  - Has been used clinically in cetaceans for chemical restraint (Joseph and Cornell 1988, Moore et al. 2010)

- Injectable anesthetics as pre-euthanasia drugs or potential pre-euthanasia drugs

  Pease (2002) defines anesthesia as the loss of sensation and usually of consciousness without loss of vital functions artificially induced by the administration of one or more agents that block the passage of pain impulses along nerve pathways to the brain.

Dissociatives

Ex: ketamine, tiletamine

- Controlled substances (Schedule III)
- IM, IV, SC, IP, PO
- Not recommended to use alone due to the potential for seizures and lack of skeletal muscle relaxation (catatonia)(Plumb 2015)
- Tiletamine is not available as a single agent but is combined with zolazepam (Telazol®)
- Telazol® is only approved in dogs and cats but has been used safely and effectively on multiple wild vertebrate species (Schobert 1987)
- Ketamine is one of the most commonly used drugs for wildlife anesthesia. It is highly effective in small animals to large ungulates (Kreeger 2007)
- Plumb (2015) states that ketamine is used as a dissociative general anesthetic; and also inhibits NMDA-receptors so it may be adjunctively useful to control pain. However, Kreeger (2007) states that while it provides peripheral analgesia, visceral pain is not abolished
- Although IM administration may be painful, use is acceptable in wild species (Fowler 1986)
- Generally combined with other sedative and anesthetics due to its synergism with many other chemical agents (Kreeger 2007). However, ketamine is labeled (Package Insert; *Ketaset*®-Bristol) for use as a sole anesthetic in cats for brief procedures not requiring skeletal muscle relaxation and in non-human primates for restraint

Hypnotics

Ex. Propofol

- Non controlled substance (in review now, but currently not controlled)
- Administered only IV
- Short-acting injectable hypnotic agent (Plumb 2015)
- In combination with acepromazine, can decrease the required dose of propofol by 30-50% (Mandelker 1993, Watkins et al. 1987)
- Used successfully in cetaceans for the induction of anesthesia (Dover et al. 1999)
- Little, if any, analgesia is provided (Plumb 2015)

Adjuvants

Hyaluronidase

- An enzyme that breaks down hyaluronic acid in connective tissue
- Added to increase IM absorption rate of other drugs (Kock 1992)

<u>Euthanasia Agents</u>

Inhalant Agents

Inhalant agents are administered via the lungs (AVMA 2015). Potential application to cetacean euthanasia appears limited.

- Inhalant Anesthetics

  Inhalant anesthetics are chemical agents that are vaporized in precision equipment. They can be used either as a pre-euthanasia agent or euthanasia agent. Generally feasible only for smaller animals (<7 kg) when used alone but can be used in larger animals when used in conjunction with injectable induction agents. The AVMA (2013) lists in order of preference the following inhalant anesthetic agents acceptable for euthanasia (not necessarily the order of preference for anesthesia): isoflurane, halothane, sevoflurane, enflurane, methoxyflurane, and desflurane. Specialized equipment is generally required to administer these agents and inhalation of the vapor by breath holding species, such as all marine mammals, requires extended time (Greer et al. 2001). The rapid active phases of marine mammal respiration could easily overwhelm most delivery systems. The administration of non-vaporized isoflurane has been attempted with equivocal results (Harms et al. 2014). Although the effects may only be marginal, we consider this method as deserving of further investigation for use in extenuating circumstances.

- Carbon monoxide

  Carbon monoxide is approved for use in euthanasia by the AVMA (2007, 2013) under prescribed conditions for animals up to the size of a dog. Exposure to an environment of 5% carbon monoxide results in oxygen deprivation, unconsciousness, and death within a few minutes. If applied correctly the method is painless and quick. The only acceptable source of carbon monoxide for use in euthanasia is purified compressed cylinders. The gas is odorless so significant risks to personnel exist. There are also a host of additional problems related to human safety, respiratory characteristics of cetaceans, and delivery that must be overcome before the use of carbon monoxide can be considered for use in the euthanasia of cetaceans. However, since there is no lingering eco-toxicity, further investigation into this method may be merited.

Injectable Agents

The use of injectable euthanasia agents is the most rapid and reliable method of performing euthanasia (AVMA 2007, 2013). They include a large range of chemical substances including:

- Barbiturates
  Ex: pentobarbital, phenobarbital, thiopental
  - Controlled substances (Schedule II, III)
  - IV, IP, IC, PO (phenobarbital)

- Most common and widely accepted agents for euthanasia
- Used therapeutically as a sedative/anesthetic, and to treat intractable seizures (Plumb 2015)
- Have been used successfully in cetaceans for anesthetic induction (Ridgway and McCormick 1971)
- Cetaceans appear to require less pentobarbital to euthanize than the generally accepted dose of 1 ml /4.5 kg of pentobarbital solutions (390 mg/ml) listed for most animals (Williams pers. comm., Walton unpublished data)
- Evans et al. (1993) concluded that a combination of pentobarbital with lidocaine IV decreased time to death (TTD) compared to the use of pentobarbital alone when euthanizing dogs
- VAQS has frequently used lidocaine in combination with sodium pentobarbital when IP administration is required with no notable responses observed. However, this involved a relatively low number of animals and limited species diversity of cases (Walton unpublished data)
- In megavertebrates, the animal should be sedated or anesthetized prior to administration (Baer 2006)
- VAQS experience, as well as personal communications with other stranding networks including: Riverhead New York (Durham pers. comm.), New England Aquarium (Merigo pers. comm.), and International Fund for Animal Welfare Marine Mammal Rescue (Touhey pers. comm.) confirm that the sole use of IV and IC administration of barbiturates has resulted in cetaceans exhibiting extended excitatory phases including dangerous spinning and fluking
- RSPCA (1997) recommends that IV injection of pentobarbital should be given into the peduncle vein of cetaceans up to 60 kg (not appropriate for larger animals). If unable to access vein, it recommends administering IP
- Carcass must be disposed of properly since multiple instances of relay toxicosis have been reported (Otten 2001, O'Rourke 2002, Kreeger 2007)

- Ultrapotent opioids
  Ex: etorphine, carfentanil, thiafentanil
  - Animals may be more prone to sudden movements when immobilized with opioids. Kreeger (2007) recommends the concurrent use of tranquillizers to decrease excitation response and hasten induction.
  - Controlled substances (Schedule II)
  - IM, IV, SC, PO (buccal)
  - Analgesic properties
  - Etorphine and carfentanil are listed for megavertebrates (Atkinson 2002, Morris 2001) including marine mammals (Kreeger 2007, Greer et al. 2001)
  - Ultrapotent opioids are not typically used for cetacean euthanasia in the United States because of personnel safety concerns in challenging working environments.

  - Immobilon® (etorphine and acepromazine) (concentration of etorphine = 2.45 mg/ml) is commonly used in Europe
  - In the United States Immobilon® is not commonly available because of strict regulations
  - RSPCA (1997) recommends Large Animal Immobilon® as the drug of choice for cetacean euthanasia. They caution against IV administration because of potential danger to the responders. Instead they recommend IM injection or, failing that, injection into the blubber
  - Large Animal Immobilon® dose: IM @ 0.5 cc of Large Animal Immobilon per 1.5 meter of dolphin or, porpoise length or 4.0 cc per 1.5 meter of whale
  - Thiafentanil plus xylazine appears to provide reliable and effective immobilization of mule deer (Wolfe et al. 2004)
  - Extremely potent and risk of toxicity to humans if accidently exposed (Morkel 1993)
  - Carcass must be disposed of properly because of ecotoxicological hazards (Greer 2001)

- Miscellaneous Agent
  T-61
  - It is a combination of a local anesthetic, a hypnotic agent and a curariform drug
  - Contains embutramide, a Schedule III controlled drug in the United States
  - Considered acceptable by the AVMA (2013) only if administered slowly IV, by trained personnel
  - Not commercially available in the United States due to concern for the potential of suffocation prior to loss of consciousness, and dysphoria when administered too rapidly.
  - Hellbrekers et al. (1990) study on the evaluation of the use of T61 for euthanasia of domestic and laboratory animals shows that the loss of consciousness and loss of muscle activity occurred simultaneously
  - Can cause secondary toxicity in scavengers consuming animals euthanized by T-61 (AVMA 2013)
  - Used successfully in combination with other agents to euthanize fin whales (Dunn 2006, Daoust and Ortenburger 2001) and a southern right whale (Kolesnikovas et al. 2012)

- Paralytics *
  Ex: succinylcholine*, pancuronium*
  - Not a controlled substance
  - Administered IM, IP, IV
  - Causes generalized muscle paralysis and ultimate asphyxiation with no loss of sensation or consciousness

    - Depolarizing neuromuscular blocking agent (Kreeger 2007)
    - No analgesic or anesthetic effects (Plumb 2015)
    - Succinylcholine combined with potassium chloride decreases the amount of time to death in cetaceans according to Hyman (1990)
    - Human death can result in accidental exposure (Greer 2001)
    - In certain emergency situations, such as euthanasia of a horse with a serious injury that cannot be safely restrained and poses a risk to people, neuromuscular blocking agents may be administered if followed by appropriate techniques as soon as possible. (AVMA 2007, 2013)
    - Generally inexpensive and does not cause eco-toxicological concerns

- Electrolytes
  Potassium Chloride (KCl)*
    - Not a controlled substance
    - Administered IV, IC (IM, IP possible)
    - The use of potassium chloride injected intravenously or intracardially is only considered acceptable in an animal already under general anesthesia or unconscious (AVMA 2007, 2013, Baer 2006)
    - Muscle tissue and/or violent body movements are often seen following administration (AVMA 2007, 2013, Baer 2006)
    - Administration causes severe electrolyte alterations and painful muscle contractions (Litz et al. 2001)
    - Generally inexpensive and does not cause eco-toxicological concerns

*All paralytics and potassium chloride are explicitly listed as inhumane methods of euthanasia in a conscious animal due to pain, fear and struggling (AVMA 2007, 2013, Baer 2006, Close et al. 1997, International Whaling Commission IWC 1980). However, as noted previously, the AVMA (2007, 2013) states that although available methods may not be “acceptable” according to currently published guidelines, they may be more humane than leaving an animal to suffer for extended periods of time.

Physical Methods of Euthanasia

A physical method must rapidly induce relatively painless unconsciousness prior to death in order to be considered humane. According to Greer (2001) the brain or brain stem must be destroyed quickly and relatively painlessly. If these criteria cannot be met, the animal should be heavily sedated or unconscious or the procedure should not be used. AAZV (Baer 2006) guidelines add that, with regards to megavertebrates, unless a human life is in imminent danger; this method should not be used in a conscious animal.

Ballistics

In cases involving the use of ballistics to euthanize cetaceans, we found conflicting accounts and recommendations, with insufficient data to resolve discrepancies. In general, however, it is a conditionally acceptable form of euthanasia in odontocetes that are not sperm whales and are less than 4-8 meters in body length (AVMA 2007, 2013, Geraci and Lounsbury 2005, Øen and Knudsen 2007, Greer 2001, RSPCA 1997, Suisted 1999, Blackmore *et al.*1995, Hampton et al. 2014) in body length. Larger animals require higher caliber weapons and larger projectiles and conventional weapons may not effectively penetrate animals greater than 7 meters in length (IWC 2010, Øen and Knudsen 2007). The shooting of sperm whales and baleen whales of any length is not recommended by the majority of sources until further work can be done because of the extremely tough and thick blubber layers and unique skull anatomy (RSPCA 1997). A significant concern with ballistics is that few practitioners are well trained in the appropriate use of firearms for euthanasia (IWC 2010). Suisted (1999) and Blackmore *et al*. (1995) are the most comprehensive sources and are generally cited by others when referencing shooting cetaceans. Bullet placement must be appropriate in order to destroy the brain instantaneously (Longair et al. 1991). While the RSPCA (1997) prefers the use of drugs to euthanize animals under 3-4 meters, guidelines do not specifically recommend that the animal be unconscious prior to shooting. The use of ballistics for mass stranding events is not recommended as anxiety and fear in the conscious animals, as well as personnel and observers, may be exacerbated (National Research Council 1992).

All resources include strict regulations when involving the use of ballistics to euthanize cetaceans. In general, criteria include three main components that should be evaluated: the size and anatomy of the animal, the firearm and projectile to be used, and the skill of the marksman. If any of these components is not ideal, this procedure should be aborted (Greer 2001). Blackmore (1995) adds that another factor to be considered is the potential ricochet of projectiles on bone or stones.

The brain is the targeted area recommended in all guidelines. Anatomical species variations make it imperative that the shooter is familiar with recommendations for each species to ensure penetration without passing through and endangering other animals or people. According to Geraci and Lounsbury (2005), the best strategy for use of ballistics is to aim at the occipital condyles, halfway between the eye and the insertion of the flipper at the level of the eye, and use the projectiles to sever the spinal cord at the base of the skull. This method quickly disarticulates the spinal cord at the base of the skull resulting in immediate paralysis and followed quickly by death. Because of the very thick skull of large odontocetes and the parabola-shaped frontal bones and distance to the brain from the melon in odontocetes, aiming from the front of the head, through the melon, is not recommended due to safety concerns from the likelihood of ricochets. In Blackmore's (1995) work with common dolphin and pilot whale carcasses, while he found that the lateral approach (halfway between the eye and insertion of the flipper at the level of the eye) appears to be effective and easier for the shooter to aim in larger animals, the optimum target for smaller animals was a dorsal approach just caudal to the blowhole. Descriptions and illustrations of various species and recommended techniques can be found in RSPCA (1997) for small cetaceans, Blackmore (1995) for pilot whales and Suisted (1999) for baleen whales.

In regards to recommendations for types of firearms to be used, the RSPCA (1997) states that on no account should a shotgun or a .22 rifle be used to euthanize a cetacea**n**. Blackmore et al. (1995) concluded that a 12 gauge shotgun armed with firing slugs can be safely and humanely used to destroy cetaceans up to four meters long but further work was necessary for this to be considered ethical and legal for live animals. This study also recommended that in odontocetes up to four meters in length, a gun no less than 7.62 millimeter with only solid bullets of at least 140 grains be used. In odontocetes up to 8 meters length, Suisted (1999) recommends standard sporting rounds using .303, .30-06 or .308 with 180 grain soft or solid round-nosed projectiles. Hampton *et al*. (2014) recommend .30 caliber (7.62 mm) 180 grain hydrostatically stabilized blunt non-deforming solid bullets for cetaceans up to 6 m in length.

The distance between the end of the barrel and the head is recommended at 0.4 to < 1 meter, and the barrel should not be in direct contact with the animal in order to avoid risk of explosion (AVMA 2013, Blackmore 1995, RSPCA 1997). If there is uncertainty regarding the proper location or penetration, placing three shots in line through the target area is recommended (Suisted 1999).

<u>Euthanasia of Sperm and Baleen Whales Using Ballistics</u>

Although mentioned in the literature (Geraci and Lounsbury 2005, Greer et al. 2001), explosives and ballistics are generally not recommended by the majority of experts for use in the euthanasia of sperm and baleen whales.

Ballistics is considered inappropriate for the humane destruction of these animals and information from the International Whaling Commission workshops on whale killing methods indicated that firearms cannot guarantee a rapid or humane death in animals larger than 7 meters (International Whaling Commission 2006).

The New Zealand Department of Conservation does include recommendations regarding the shooting of sperm and baleen whales with very specific guidelines (Suisted 1999). They have developed the "Sperm Whale Euthanasia Device" (SWED) specifically to enable the humane shooting of these animals, however further studies are being conducted on its use (Marsh and Bamber 1999). There are few people licensed to possess this weapon (as of 2006 only Craig Bamber) and the cost of time and effort may be significant. Initial trials of the weapon included the euthanasia of two sperm whales. The first was killed after a single shot while the second required two shots and remained alive for over two hours.

In regards to shooting small stranded baleen and sperm whales (<7 meters), high caliber bullets (7.2 mm, 9.3 mm, 30.06, .375 or .458 inch) appear to be the most reliable (IWC 2006). Suisted (1999) recommended using .303 rifles with MK.6 projectiles, .458 and solid nosed projectiles, or the SWED and then only by specially trained personnel. Studies found that .375 and .458 round nosed full metal jacket projectiles can penetrate and kill minke whales when hit in or near the brain (Øen and Knudsen 2007). Øen (2003) also euthanized a 7m sperm whale using a rifle and .458 full metal jacketed ammunition. Ultimately, Blackmore et al. (1997) summarized that for larger cetaceans, until further work on dead cetaceans has been carried out, euthanasia using ballistics on live animals would be unethical and inappropriate.

<u>Explosives</u>

The majority of sources we reviewed condemned the use of explosives to euthanize cetaceans. However, the use of explosives for cetacean euthanasia has been explored in different areas of the world. Sources mention potential placements of explosive charges for euthanasia, either deep in the mouth (Gerasci and Lounsbury 2005, Needham 1993) or externally over the cranium (Coughran *et al.* 2012, Greer 2001). Western Australia has developed sound methodology with repeated success in the use of explosives and has recently published its method (Coughran *et al.* 2012). This method involves the specialized use of shape charges that result in cranial implosion to euthanize large baleen whales. While the method takes some time to set up, death is nearly instantaneous once the charge is exploded. In Western Australia, stranding responders are all state employees who work closely with state police and other officials in the safe use of explosives (Coughran *et al.* 2012). In the United States, purchase and use of the explosives described by Coughran *et al.* (2012) is strictly regulated and certification for use by stranding organizations is likely to be expensive and difficult.

The whaling industry uses explosive harpoons to kill whales by detonating a penthrite grenade within the head or thorax, inducing neurotrauma and death (Knudsen and Øen 2003). Penthrite is one of the most powerful high explosives. In general when explosives are used, firearms are instituted as a secondary killing method when necessary (International Whaling Commission 2006). Øen (2006) and Knudsen & Øen (2003) reported on the examination of 37 minke whales killed in Norwegian commercial whale hunts between 1998 and 2000. The study reported that neurotrauma induced by blast-generated pressure of a single 30 gram penthrite grenade was the primary cause for immediate or very rapid loss of consciousness and death. Approximately four out of five animals died immediately based on IWC time to death (TTD) criteria in the 2000 – 2002 hunts (Øen 2006), and those that did not were judged insensible during the TTD interval based on pathologic findings (Knudsen and Øen 2003). Although IWC TTD criteria have been criticized from opposing perspectives as inconsistent and inadequate (Butterworth et al. 2004; Øen 2006), even under the worst case scenario for the approximately 20% of whales that did not die instantly, all expired substantially more rapidly (several minutes at most) than some live stranded whales have been documented to linger in a badly compromised state (days) before intervention was possible (Daoust & Ortenburger 2001, Kolesnikovas et al. 2012, Harms et al. 2014). Where equipment and expertise are available, and the technique culturally acceptable, use of explosive harpoon would be a reasonable consideration to end prolonged suffering of a stranded live whale. Lack of public acceptance, equipment and expertise would make this technique unlikely in the U.S.

Exsanguination

Sources that refer to exsanguination as a form of euthanasia say it is only considered humane when the animal is unconscious and should only be used as a secondary method to ensure death (AVMA 2007, 2013, Baer 2006). It is not mentioned in the CRC Handbook of Marine Mammal Medicine (Dierauf and Gulland 2001). In large whales and mass stranding cases where carcasses cannot be appropriately disposed of and toxic chemicals cannot be used, NOAA Fisheries has recommended exsanguinations by severing flukes at the peduncle (Walton unpublished data). An instance of exsanguination as the final step of a multi-step euthanasia involved a right whale stranded on a shoal in North Carolina. Responders had limited access to the animal and it could not be moved. After two days and use of all available less toxic chemical agents for euthanasia, exsanguination was deemed to be the most humane course of action (Harms et al. 2014). Following the first cut to the peduncle, 40 minutes elapsed prior to the pronouncement of death. In another case, a pygmy sperm whale stranded alive in a military live fire range could be neither moved nor buried after euthanasia, and military firearms had been ruled out as an option. After high doses of acepromazine and xylazine rendered it unresponsive, the caudal peduncle was severed and it expired 19 min later (Harms unpublished data). In these and other cases, the negative effect on the public and the responders is difficult to overcome, and more so if visual images are broadcast by commercial or social media.

In the Faroe Islands during pilot whale hunts, animals are exsanguinated by a dorsal cut into the neck just caudal to the blowhole and deep enough to sever the spinal cord and surrounding blood vessels leading to the brain. Loss of consciousness, when appropriately preformed, results in 5-10 seconds (Olsen 1996). Historically the cutting of carotid arteries has been advocated as humane for the euthanasia of cetaceans, however, the brain of these species is not supplied by the carotid arteries (Vogl and Fisher 1981) and death is prolonged. The trauma inflicted by this method, and extended time to death make this method inhumane (Suisted 1999). Similarly, Smithsonian mammal staff collected reports from former Ocracoke, North Carolina *Tursiops* fisheries of using a long, thin knife inserted just ventral and caudal to the left flipper to slice open the left ventricle to achieve quick death (Potter pers. comm.). These methods, however, are unlikely to be quick or simple in larger cetaceans such as sperm and baleen whales.

Another method of exsanguination utilized in hoofstock is the severing of the brachial vasculature. The front limb is lifted and the knife inserted deeply into the axillary area at the elbow, cutting until the limb can be laid back away from the thorax (Shearer et al. 2002). This method (the severing of the brachial vasculature) was successfully used on a heavily sedated 8.4 meter sperm whale (Tristan pers. comm.). It resulted in fairly rapid blood flow and safer access than the more commonly used peduncle area. It also enabled the actual cutting procedure to be more readily shielded from the crowd of on looking general public.

The potential to release infectious agents and cause dangerous conditions (slippery) must be considered during exsanguination (Geering et al. 2001).

Bilateral Thoracotomy

This method has only been mentioned as an adjunctive method, to be used as a secondary means to verify death (Greer et al. 2001). This method should not be considered an option for cetaceans because of their physiological adaptations for deep diving in their marine environment.

Pithing

This has only been recommended as a secondary means of euthanasia. A pithing rod is used for the humane destruction of sheep, pigs and cattle after stunning (captive bolt) (Butterworth et al. 2003). A form of pithing has been employed in the Japanese dolphin drive fishery in Taiji as a slaughter method, indicating that it is technically feasible in small cetaceans, but as a sole method it is questionable as humane slaughter, let alone euthanasia (Butterworth et al. 2013). It would require an appropriate primary method to be considered as the last part of a multi-stage euthanasia method.

Electrocution

Electrocution is considered unacceptable because humane death cannot be achieved by this method and the safety of the responders would be severely compromised (Barnes et al. 2006, McLachlan 1995). It has been found to be ineffective and potentially cause pain and suffering.

Verification of Death

Verifying death is imperative when the euthanasia/killing of any animal is involved (Greer et al. 2001). Close (1996) states that the only reliable method of death confirmation is the absence of a heartbeat. While this is possible to determine in some smaller, shallow diving cetaceans, a heartbeat is not always detectable using standard methods in large or deep diving species.

Multiple techniques should be utilized to confirm death. Criteria other than a lack of heartbeat which may be useful to assist in the confirmation of death in cetaceans include (Butterworth et al. 2003):

- Lack of jaw tone
- Absence of menace, palpebral and corneal reflexes
- Pupils are fixed and dilated
- Absence of respiration
    - Can be hard to assess in animals that normally hold their breath for extended periods
- Lack of response to painful stimulus
- No capillary refill time
- Ocular/skin temperature differential
    - The temperature of the eye surface and surrounding skin evaluated through thermography. The eye surface cools more rapidly after blood circulation ceases

Secondary physical means for euthanasia, including exsanguination and ballistics, should be performed if there is any doubt (Close et al. 1996).

Literature Review Conclusion

The goal of this Prescott project is to develop recommendations for stranding networks to facilitate the humane euthanasia of live stranded cetaceans. Although there is limited published information available, we have attempted to identify and present a comprehensive collection of available information and methods as well as identify areas where more data are needed. In general, we found each method has disadvantages and implementation becomes even more challenging for euthanasia of large whales.

There is a general consensus that a two-step process, particularly administration of pre-euthanasia agent accompanied by at least one other method, appears to be the most widely accepted. In regards to chemical methods, a combination of chemical agents, especially in smaller cetaceans (<8 meters), seems to have the best potential. However, more information is needed concerning chemical methods, including: research into various administration routes, doses, and techniques; best specific agents and combinations; potential species specific and idiopathic reactions; and minimizing the potential for personnel and eco-toxicological hazards.

Regarding available physical methods, most resources discuss the use of ballistics as a technique, generally shooting through the brain. Appropriately trained and experienced personnel are a necessity when considering this method. There is a great need to further determine and define ballistics techniques and specifics regarding various species and equipment required.

There is less of a consensus on exsanguinations and explosives due to safety and acceptability. These, and potentially other methods we may ultimately propose, may not be deemed "acceptable" from a traditional perspective. However, given the diversity of cetacean stranding events, and especially those involving large whales, the alternative of allowing an animal to "die a natural death" rather than alleviate extended suffering is often even less acceptable from an animal welfare perspective.

Regardless of method used, death of the animal must be confirmed. Criteria and measurable parameters to verify death, especially in large whales, are not always readily accessible or reliable. As a result, promising techniques like the ocular/skin temperature differential appears to warrant further investigation.

In our quest for the best and most current information available, we continued to search and review literature throughout this Prescott project. Ultimately, we incorporated this information and results from our assessments of historic stranding data to develop a repertoire of possible actions available, given the current state of knowledge. We took into consideration not only the method of euthanasia but also recommendations regarding the safety of personnel and the environment, protection of biological samples, the need for appropriate training for optimal execution of any given method, and how methods would affect stranding personnel and the public. This enables a stranding responder to identify a safe and practical euthanasia method to facilitate the best possible outcome given their specific situation.

Cetacean Euthanasia Data Review

Crucial to the accomplishment of this project was the collection of historic cetacean euthanasia data. We began by introducing the project and stressing the importance of network participation at regional and national marine mammal stranding conferences. To ensure the highest participation rate and most effective data collection, we directly contacted stranding facilities by email, phone and face to face meetings. We also solicited the participation of several stranding response organizations to utilize a new euthanasia data sheet. Developed as part of this two year project, the data sheet would be used to standardize data collection from cetacean euthanasia events occurring throughout the study period. A database was then developed to record all compiled data. It consisted of three basic sections:

Stranding data: level A data (basic data including species, sex, length, location, date, carcass condition), response times, necropsy and disposal information

Animal data: pre-, peri- and post- euthanasia behavioral and physiological observations and details

Euthanasia data: method(s), drug information, times of procedures and death

One of the most extensive and time-consuming challenges of this project was the task of collecting historic cetacean euthanasia data. We sought to collect data from 53 stranding response organizations for cetacean euthanasia events listed in national and regional marine mammal stranding databases for the United States from 1990-2009. Twenty-seven of the response organizations failed to respond to our inquiries. In general, data consistency and completeness of euthanasia records was poor.

We also identified discrepancies that exist between information in the national database and the data available from the responding organizations. For example, according to the national database, VAQS had a total of 38 cetacean euthanasia events, as compared to our database which included 58 such events. In another case, an organization had only five records in its database compared to the 18 events listed in the national database. Ultimately, we compiled data on 468 events from 26 organizations from 1990 through 2011.

Cetacean euthanasia demographics summary

Of the 468 events we investigated, 300 provided adequate information for demographic analysis of cetacean strandings that resulted in euthanasia. Greater than 90% occurred in the southeast and northeast regions of the U.S. There was more mass stranding euthanasia events in the northeast and more single stranding euthanasia events in the southeast, but nearly twice as many single stranding euthanasia events overall. Temporally, single stranding euthanasia events were fairly steady from February through September, with a mean of 25 each month for all years combined, and lower from October through January, with a mean of 11 per month. The highest number of single stranding euthanasia events was in February (n=33) and the lowest in October (n=5). The peak months for mass stranding euthanasia were January (n=43), December (n=23) and March (n=21). Among all years combined, the mean for other months was four, and no mass stranding euthanasia events were reported in May or June. Differences in numbers of euthanasia events per year were largely driven by mass stranding events, with single stranding euthanasia events relatively low (mean<10) prior to 2003 and higher (mean=24) after 2003. The number of euthanasia events increased from 15 to 34 per year after the Prescott program began providing funding in 2003.

Cetacean euthanasia method, drug & dose summaries

Of the total of 468 cetacean euthanasia events we collected nationwide, 283 (60%; representing 20 species) met project inclusion criteria regarding euthanasia methods, drugs and doses. Chemical administration represented the most common practice during a euthanasia event (281 of 283; Table 1).

Physical methods performed with chemical pre-medication included three exsanguinations (*Physeter macrocephalus*, *Kogia breviceps*, *Eubalaena glacialis*). Each of the three events cited logistical inability to remove the carcass and the resulting concern regarding potential eco-toxicological impacts as the factor that determined the decision to exsanguinate rather than administer sodium pentobarbital. Physical methods without chemical administration included a gunshot to the head of two animals (2 of 283) (sub-adult *Delphinus delphis*, approximately 160 cm, and *Phocoena phocoena*,106 cm).

Injection with sodium pentobarbital alone represented 166 (59%) of the 281 cetacean euthanasias, while premedication followed by sodium pentobarbital accounted for 26% (N=72) (Table 1). The number of premedication agents administered preceding sodium pentobarbital varied from one to three: one agent (N=45); two agents (N=23); and three agents (N=4). The most common single premedication agents included diazepam (N=19) and xylazine (N=12). The most frequently administered two-agent premedication included acepromazine/xylazine (N=7) and diazepam/xylazine (N=7) combinations.

Forty-two chemical euthanasia events (15%) did not include the administration of sodium pentobarbital. Of these, single agent chemical euthanasias accounted for 10 events, with xylazine being the most common single agent administered (N=5). Other single agents included diazepam (N=2), Telazol (N=2) and acepromazine (N=1). Based on VAQS experiences along with personal communications with other network responders, these single agent events occurred because the animal expired prior to further agent administration, rather than actual intended euthanasia protocol.

In twenty-eight (67%) of the 42 events in which sodium pentobarbital was not administered, two agents were used. The most common combination consisted of acepromazine and xylazine (N=23) and included one exsanguination post-sedation. Four-agent euthanasias accounted for two of the 42 events in which sodium pentobarbital was not administered and included one event utilizing KCL and another utilizing KCL along with air embolization. Three- and six-agent euthanasia events each involved one animal, with the six-agent event spanning a two day period and culminating in exsanguination.

There was a tendency for acepromazine and xylazine to be dosed lower when followed by euthanasia solution than when not followed by euthanasia solution, but the differences are not statistically significant (Wilcoxon rank sums test, $p = 0.0529$ for acepromazine and $p = 0.1336$ for xylazine).

Whether or not animals were premedicated had little effect on dose of pentobarbital used ($p = 0.7042$). Pentobarbital doses are, however, somewhat lower when an alpha-2 agonist (xylazine, detomidine, or medetomidine) is included as a premedication ($p = 0.0808$).

There was no statistically significant difference in pentobarbital doses when a benzodiazepine (diazepam or midazolam) was included as a premedication ($p = 0.2071$). In fact, the pentobarbital dose was slightly higher when preceded by a benzodiazepine. Pentobarbital doses were significantly lower when acepromazine was included as a premedication ($p = 0.0067$), but in all 10 cases, xylazine was also included as a premedication. No other premedications were employed frequently enough for meaningful statistical analysis.

Doses were significantly different among species for pentobarbital ($p < 0.0001$), but did not differ significantly for any other drug. Pentobarbital doses for *Delphinus delphis* were significantly higher than for *Grampus griseus*, *Tursiops truncatus*, *Kogia breviceps* and *Lagenorhynchus acutus*, but did not differ significantly between any other species pairs.

Suitable weight/length charts could not be generated or located for *Globicephala* spp., so in order to retain and present useful data, doses for these two species are presented per meter of length.

Calculations based on weight are preferable when possible, because doses do not scale linearly with length, but do scale close to linearly with weight (see Appendix 1). Median number of agents used was one, with a minimum of one and maximum of three agents.

Calculating doses from the compiled data was one of the most challenging aspects of this project. In many cases, no weight was available in the records we collected. In some cases, we could not retrieve information on drug concentration. A reliable means of calculating minimum effective doses of pre-euthanasia sedatives and euthanasia drugs is highly desirable. The hazards of under-dosing are widely appreciated (*e.g.* unnecessarily prolonging animal suffering) leading to an understandable tendency to administer more than the required amounts of euthanasia drugs in order to ensure rapid and safe death. The concept of overdosing euthanasia drugs initially appears oxymoronic. But administering more euthanasia drugs than necessary carries its own undesirable consequences, including greater potential for relay toxicosis and environmental contamination, tissue changes that can interfere with pathologic evaluation (particularly congestion), and higher costs (magnified in large animals). Whenever possible, doses should be calculated and recorded based on weight rather than length, because weight increases non-linearly with length, and at different rates for different species. Weight can rarely be determined directly for a stranded animal on the beach, but length is usually readily measured post-mortem. Length to weight equations and curves have been generated as part of this project for several cetacean species that commonly strand in the mid-Atlantic U.S. coasts, utilizing data from VAQS, NCSU CMAST, and UNCW (Table 2; Appendix 1). Despite sample size limitations and potential regional specificity, these equations and figures can be used to approximate weights with sufficient accuracy for the purposes of calculating doses for pre-euthanasia sedatives and euthanasia drugs, adjusting as appropriate for subjective assessments of body condition and degree of debilitations. These are not meant to supplant actual weights for the purposes of survival medical treatments, although in some cases they may provide a starting point. Whenever weight is calculated from length in euthanasia cases, the carcass should be weighed postmortem if at all possible to allow back-calculation of actual doses used.

Workshop and Working Group Results

The project proposal included a number of co-investigators and collaborators who were to make up an expert panel with funding to attend a workshop on cetacean euthanasia. Panel members included veterinarians, researchers, stranding responders and NOAA Marine Mammal Health and Stranding Response Program staff. As the project progressed, we added several additional people and expanded the expert panel to a Cetacean Euthanasia Working Group (CEWG).

The CEWG reviewed and provided comments on the literature review, survey and survey data, historic euthanasia data, cetacean euthanasia data sheet and instructions, and participated in the workshop held in October of 2011. The workshop was held in Virginia Beach for 2.5 days and was facilitated by Mark Swingle and Susan Barco of VAQS. The members of the workshop reviewed the compiled information, discussed details not captured in data sheets or in the survey, and developed the following recommendations for final products of the project.

The group discussed the need for more guidance on making the decision to euthanize, but in accordance with the main goal of this project, specifically addressed the process of euthanizing a stranded cetacean AFTER the decision has been made. Utilizing the compiled information, as well as the expertise and experience of working group members, we developed recommendations to assist stranding networks in making more informed decisions and facilitating the humane and safe euthanasia of live stranded cetaceans.

The euthanasia guidelines we developed attempt to balance the ideal of minimal pain and distress with the reality of the many environments in which cetacean euthanasia could be performed. The group agreed that NOAA/NMFS authorized veterinarians and/or responders with appropriate training and expertise for the species involved should be consulted to ensure that proper procedures are used.

Applying the definition of euthanasia for the workshop (adapted from AVMA 2007)

The CEWG started by discussing the definition of euthanasia (AVMA 2007, the most current version at the time of the workshop) and how it applies to stranded cetaceans. Throughout the workshop the definition's application to cetacean strandings was edited. We agreed to the following:

> *Euthanasia, or the induction of humane death, of stranded cetaceans is often the sole choice for authorized responders to relieve pain, distress and suffering. The panel recognizes that without humane euthanasia, animals may be subject to hours or even*

days of pain, suffering and stress if left to die naturally. The techniques used should result in rapid loss of consciousness, followed by cardiac or respiratory arrest and the ultimate loss of brain function (AVMA 2007, 2013). Euthanasia methods should also be safe for all responders and minimize the risk for secondary intoxication of (scavengers and) predators and environmental contamination upon disposal.

General Recommendations for Euthanizing Stranded Cetaceans

The CEWG felt that a combination of general guidelines and species-specific observations would be the most effective way to impart information. These guidelines and observations are not meant to be rigid directives and the species-specific observations are living documents based largely on the collective experience of the group and not on systematic research (see Appendix 1). As more data and observations become available, we expect that changes will occur.

Responders and veterinarians should be aware that there are many uncontrolled and often unrecognizable variables associated with every live stranding event and, as a result, that each event will be different. The CEWG recognized that we cannot develop a 'cookbook' for cetacean euthanasia, and practitioners must understand that these are guidelines and not directives.

1) Whenever possible, it is preferable to avoid or minimize pain or distress resulting in a "gentler death" that may take a longer time, rather than a more rapid but stressful death. The CEWG recognizes that in some events pre-euthanasia sedation is appropriate prior to euthanasia, especially when administration of such is safe and there is sufficient time for the agent(s) to take effect prior to administration of the killing agent. Sedation can be accomplished with a single agent or a combination of analgesic and/or sedative agents IM and/or IV. Use of sedation and drug combinations MAY result in less drug volume (especially of euthanasia solution) which can help mitigate, but not eliminate, potential ecotoxicosis and secondary intoxication.

AND

The CEWG recognizes that for other events pre-euthanasia sedation is not indicated and believes that single agent administration of a euthanasia solution (IV) as approved by AVMA (2007, 2013) is appropriate and humane.

AND

Single agent administration of a euthanasia solution via intracardiac injection (IC) is not ideal, but is conditionally acceptable in stranded cetaceans in extenuating circumstances.

2) The CEWG DOES NOT recommend use of KCl alone.
3) When use of standard euthanasia solution(s) is not an option, pre-euthanasia sedation followed by KCl to effect is acceptable. Note that high volume of KCl may be needed and should be readily available.
4) The CEWG recognizes that chemical euthanasia involves toxic substances and all efforts should be made to minimize secondary intoxication and environmental contamination (ecotoxicity). The panel urges all practitioners to dispose of waste drugs properly and in an environmentally acceptable manner.
5) The CEWG suggested the development of a matrix of cetacean euthanasia method choices available to stranding responders and veterinarians. Matrices for small and large cetacean euthanasia are included in the cetacean euthanasia guidelines (see Appendix 1).

Species-specific observations

Cetacean species are adapted to a wide variety of habitats and niches and these adaptations likely result in different behaviors and reactions to live stranding events and to drug administration. Below are observations by the CEWG regarding specific species:

Delphinus delphis

For this species, handling is very important. *Delphinus* seem to be very sensitive compared to other species and require minimal handling and minimal sound. As much as is safe, a hands-off approach to this species appears to work best

Observing and working with animals in enclosed spaces (*e.g.* enclosed vehicles) seems to result in calmer animals as compared to on beach exposed to the elements. However, safety of responders is paramount

Single agent euthanasia (IV) may be the most effective chemical means of euthanasia for this species

Violent movement at the time of death (with or without human intervention) is not uncommon and may be more likely in warmer weather

Kogia spp.

Pre-euthanasia sedation is recommended for these species. Using acepromazine IM @ (1 mg/kg IM) followed by xylazine IV (2 mg/kg IM) 15 minutes later or midazolam alone

IV (0.1 mg/kg) appears to be an effective sedative. Administration of diazepam and meperidine at 2mg/kg IM and after 20min administration of 2mg/kg euthanasia solution IV has also been used effectively on *Kogia* spp.

Kogia sima has a greater tendency to react more violently to sedatives than *Kogia breviceps*. Sedation alone, when responders are limited to non-controlled or non-barbiturate drugs, may take a long time (45 min - several hours) in *K. sima*

Grampus griseus

Alpha-II agonists (xylazine, medetomidine) are NOT recommended for this species as an initial treatment (adverse reactions have been observed including spinning, flipping and violent retching)

Diazepam IV (or IM if IV not possible) seems to be effective at sedation

Tursiops spp.

The *Tursiops* spp. that strand on U.S. beaches are currently considered different morphotypes, but are likely to be multiple species and could potentially react differently.

Pre-euthanasia sedation has been effective with stranded *Tursiops*

Globicephala spp.

Globicephala melas were euthanized with a sedative combination of acepromazine and xylazine during a mass stranding event in 2005 and the animals went down quietly but slowly (45 min)

Globicephala macrorhynchus were euthanized with acepromazine and xylazine followed by KCl during a mass stranding event in 2014, and some of the animals went through a prolonged (15-20 min) agonal response when KCl was administered (D. Boyd and G. Lovewell *pers. comm*)

Steno bredanensis

Pre-euthanasia sedation has been effective with *Steno sp.*

Safety Recommendations

Human safety should always be the top priority during these events. It is important to know your team's experience, skill and limitations and to continually assess the safety of a situation. Mitigation of safety concerns should always include personal protective equipment (PPE) including gloves, masks and face shields.

Concerns/risks include, but are not limited to:

- Operating in water is risky, and NOT PREFERRED. The CEWG does not recommend swimming near a stranded animal. The CEWG DOES NOT RECOMMEND working close to a live cetacean in surf, especially in water deeper than knee deep

- Someone at the scene should have general first aid/CPR (EMS certification). Stranding response equipment should always include a first aid kit minimally. Floatable back boards, splints, and neck braces are optimal.
- There is always a risk of accidental injection of potent drugs. Responders should also be aware of the possibility of broken hub/syringe and spray back of potent drugs. Be ready to respond quickly if spray back occurs (flush in first aid kit)
    - Know the location of and quickest route to a hospital
    - Whenever possible use Luer lock syringes to reduce the likelihood of spray back
    - Used needles and other sharps should always be disposed of in a sharps container
    - NEVER assume that local EMS will have sedative reversal agents
    - Reversal agents, if available for the drugs used, should be included in stranding first aid kits for EMS to administer @ site if potent drugs are used.
        - *Veterinarians and stranding response personnel are not licensed to treat human patients and should not administer reversals, but should be aware of the dose and concentration of potent drugs and the appropriate reversal agents and doses*
- Concentrated sedatives ARE NOT RECOMMENDED for euthanasia for most stranding response organizations. These require specialized safety measures.
- The responding organization should establish a 'Zone of Safety' around animal/drugs/tools (good use of local enforcement and/or inexperienced or volunteer responders).
    - When a euthanasia is conducted in a public area, educating the public about the process is important
    - Prepare the public by explaining why the choice to euthanize was made and what will happen. Have talking points for less experienced volunteers and staff. It is often valuable for an experienced staff member to address the public.
    - Allow time for members of the public to walk away.
    - Answer questions prior to the event and afterwards if needed.

Research Needs

The group recognized the need for more information on many aspects of cetacean euthanasia, including:

- The fate and effects of agents need to be better understood prior to the development of a euthanasia protocol in the context of a particular disposal option. Research must be conducted and data compiled on carcasses that are left *in situ*, rendered, buried and composted. We need to consider the degradation of individual drugs, drug combinations and assess risks to scavengers and overall environmental quality.
    - Specific questions include: What tissues have higher/lower toxicity?

- The CEWG routinely administers between 0.5 and 2.0 times the dose recommended in domestic species (1ml/10lb) of euthanasia solution (390mg/ml sodium pentobarbital).
    - There is a need to determine whether this dose can be decreased (with and without pre-euthanasia agents)
- In addition to dose information, there is a need for better means of calculating drug amount when weight is not available. Especially useful would be species or genus specific weight/length graphs. Some of these have been developed and are available in the cetacean euthanasia guidelines (Appendix 1).
- The CEWG needs to further investigate the combination of pre-euthanasia sedation (using both controlled and non-controlled drugs) and KCl to effect. More data are needed on KCl volume and route in cetaceans.
- Other sedatives and drug combinations need to be tried on most species
- Need better information/training on physical methods of euthanasia (when/where NO chemicals can be used):
    - Ballistics – animal size vs. weapon/bullet size – location well established (without chemical agent administration)
    - Knife/lance (to create internal hemorrhage, with heavy sedation)
    - Exsanguination target areas (Appendix 1)
        - Peduncle/fluke
        - Heart/great vessels
        - Brachial plexus
    - Cervical separation
    - Other methods that could be used in U.S. realistically
        - Shape charges are unlikely because of legal issues

Training Needs

There is a need/interest from the group to address the '*decision to euthanize'* and the veterinary process of triage. This is covered to a degree in Marine Mammals Ashore (Geraci and Lounsbury 2005) and in the CRC Handbook of Marine Mammal Medicine (Dierauf and Gulland 2001).

The CEWG agreed that when a decision whether to euthanize must be made, that process constitutes practicing veterinary medicine and should involve a veterinarian, either directly or indirectly, via consultation and/or carrying out previously agreed upon protocols.

The CEWG agreed that the physical process of euthanasia AFTER a decision is made to euthanize may be conducted by NOAA/NMFS authorized non-veterinary stranding response personnel who have received adequate training. The authority to euthanize should be explicitly stated in the SA. It should also be discussed with the DEA and state officials with concerns regarding the euthanasia of federally protected species. This will help to alleviate concerns regarding controlled drug use and euthanasia of wildlife not covered under state authority.

The CEWG suggests that NOAA Fisheries policies to euthanize based on condition, species, size and/or age class should be made with the input of veterinarians, stranding response personnel, and the public display community. There is definite interest from the panel in development of a NOAA approved euthanasia training process.

The training/authorization/apprenticeship should minimally include:

- Classroom training to include:
    - Region-specific items
        - Cultural concerns
        - Species diversity within the region
        - Endangered species
        - NMFS/DEA and other guiding policies
    - Physical/manual restraint techniques
    - Parenteral drug administration (injections)/phlebotomy training (including various routes, IM, IV, IP, IC, etc.)
    - Explanation and training of euthanasia methods with an emphasis on training for physical methods
    - Basic clinical pharmacology information; ecotoxicity concerns/carcass disposal options
    - Standardized and continued data collection and observations
- Field training to include:
    - Observation followed by assistance with euthanasia events
        - Parenteral drug administration (injections)/venipuncture practice (on carcasses and/or rehab patients scheduled for blood collection and/or injections)
- Approval by DEA license holder and veterinarian listed on license if controlled drugs are used

There is also a need for:

- Practical training for veterinarians who do not have marine mammal experience
- Matrix of needle length and gauge needed for various routes in various sized cetaceans (included in Appendix 1)
- Summary of drugs and dose ranges that have been used effectively (included in Appendix 1)

Summary

In summary, responses from the CEWG and network participants who collaborated on data collection and the survey have all been very positive and encouraging. There is clearly an interest from the marine mammal stranding network for guidance on cetacean euthanasia. While this report cannot answer all of the questions responders have about safely and humanely conducting euthanasia on stranded cetaceans, it represents a good first step in understanding what we know, what we need to know and where we should focus future energy on the topic.

Appendix 1 of this report is designed to be a stand-alone document that summarizes information on euthanasia methods, historically effective drug choices, equipment needs and safest practices. We intend for it to be a first draft providing the marine mammal stranding network with information to help with euthanasia events. We encourage stranding organizations to share this information with staff and to provide it to outside veterinary medical personnel (who may be called upon to conduct cetacean euthanasia) before an event occurs as well as use it as a reference during an event. Further refinement of this document will be needed as more information is developed, and we encourage the NMFS MMHSRP to continue to support work that will provide further data and refined recommendations to the stranding network.

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Table 1: Combined Summary Statistics of all euthanized cetaceans for which doses could be calculated: 282 individuals of 20 species met inclusion criteria for calculating mg/kg drug doses. Actual weights were determined for 148 individuals, estimates were made for 95, and method of weight determination was not recorded for 39.

Species	Number	Percentage of Total (%)	Median (minimum, maximum) Weight (kg)	Mean (+/- sd) Weight (kg)	Median Length in cm (minimum, maximum)	Mean Length in cm (+/- sd)
Delphinus delphis	80	28.3	78 (26, 132)	79.7 (+/- 28.1)	198 (129, 240)	194.2 (+/-25.6)
Lagenorhynchus acutus	42	14.9	104 (30, 255)	188.9 (+/- 58.5)	204 (156, 280)	208.1 (+/- 41.7)
Kogia breviceps	36	12.8	300 (30,455)	269.0 (+/- 116.7)	276 (121, 328)	253 (+/- 55.9)
Phocoena phocoena	35	12.4	23 (7.7 – 52)	24.7 (+/-6.9)	116 (77, 156)	116.3 (+/- 14.2)
Grampus griseus	25	8.9	225 (71, 323)	205.6 (+/- 71.5)	270 (187, 290)	256 (+/- 30.1)
Tursiops truncatus	21	7.4	193 (16, 400)	186.0 (+/- 97.4)	254 (106, 331)	241.1 (+/- 54.3)
Kogia sima	8	2.8	102 (10, 191)	88.8 (+/- 58.5)	176 (84, 244)	169.2 (+/- 50.4)
Globicephala melas	5	1.8	347 (103, 560)	302.8 (+/-186.2)	295 (201, 352)	285.4 (+/- 54)
Stenella coeruleoalba	5	1.8	36 (28, 45)	93.1 (+/-36.9)	203 (156, 216)	194.2 (+/- 24.7)
Balaenoptera acutorostrata	4	1.4	345 (211, 500)	350.3 (+/- 158.8)	329 (284, 387)	332.4 (+/-51.5)
Megaptera novaeangliae	3	1.1	8500 (6500, 9000)	8000 (+/- 1323)	830 (762, 878)	823.3 (+/-58.3)
Mesoplodon europaeus	3	1.1	550 (166, 823)	512.9 (+/- 329.9)	432 (201, 444)	359.0 (+/- 137.0)
Peponocephala electra	3	1.1	160 (153, 166)	160.0 (+/- 6.6)	248 (246, 249)	247.8 (+/- 1.7)
Balaenoptera physalus	2	0.7	22437 (10000, 34875)	22437 (+/- 17589)	1402 (129, 240)	1502 (+/- 462)
Eubalaena glacialis	2	0.7	5763 (1526, 10000)	5463 (+/- 5992)	735 (495, 975)	735 (+/-339)
Feresa attenuata	2	0.7	125 (122, 128)	125.2 (+/- 4.0)	206 (205, 207)	206.0 (+/- 1.4)
Physeter macrocephalus	2	0.7	14200 (11400, 17000)	14200 (+/- 3960)	1110 (1021, 1200)	1110 (+/- 127)
Stenella frontalis	2	0.7	36 (28, 45)	36.5 (+/- 12.0)	144 (137, 150)	143.7 (+/- 9.5)
Globicephala macrorhynchus	1	0.4	332	332	299	299
Stenella clymene	1	0.4	92	92	202	202

Table 2: Regression equations for total body length (L, in cm) versus total body weight (kg) for eight cetacean species stranded in North Carolina and Virginia, USA.

Species	N	Weight (kg) =	R^2
Delphinus delphis	49	$0.0044927*(L – 205.912)^2 + 1.2063757*L – 161.63$	0.828
Grampus griseus	20	$0.0056553*(L – 241.59)^2 + 1.9757271*L – 312.08$	0.897
Kogia breviceps	42	$0.0050229*(L – 238.419)^2 + 2.1917378*L – 303.50$	0.895
Kogia sima	33	$0.0049206*(L – 202.221)^2 + 1.7564184*L – 210.58$	0.899
Phocoena phocoena	53	$0.0072798*(L - 116.683)^2 + 0.3895724*L – 22.00$	0.573
Stenella coeruleoalba	31	$0.003495*(L – 201.903)^2 + 1.2171744*L – 150.09$	0.946
Stenella frontalis	29	$0.0061927*(L – 203.924)^2 + 1.525497*L – 207.59$	0.837
Tursiops truncatus	171	$0.004468*(L – 196.833)^2 + 1.3728948*L – 168.61$	0.939

Appendix 1: Stand Alone Field Reference

Recommendations for Euthanasia of Stranded Cetaceans

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Background

The goal of this project was to compile published information and existing euthanasia data and to develop recommendations for stranding networks to facilitate the euthanasia of live stranded cetaceans once veterinary medical personnel have determined this to be the best option.

In order to achieve this goal we:
- Developed standardized methods of data collection for cetacean euthanasia events
- Performed an extensive literature search, including "gray-literature" (personal communications, medical records, stranding reports, government documents) and peer-reviewed publications and compiled pertinent information
- Compiled and analyzed data from historical euthanasia events to develop a cetacean euthanasia database
- Convened an expert advisory panel whose tasks were to:
    - Critically examine euthanasia data
    - Based on the literature, data, and their experience, develop recommendations for safe and humane cetacean euthanasia and subsequent carcass disposal

The working group included nationwide participants including: veterinarians experienced in stranding response and cetacean euthanasia, field response personnel from areas with high live cetacean stranding rates from two different regions and NOAA Marine Mammal Health and Stranding Response (MMHSRP) staff. The recommendations included in this document are meant to assist with stranding networks in the process of cetacean euthanasia and not to be used as directives or requirements.

The cetacean euthanasia guidelines we developed attempt to balance the ideal of minimal pain and distress with the reality of the many environments in which cetaceans may be euthanized. NOAA/NMFS-authorized veterinarians and/or responders with appropriate training and expertise for the species involved should be consulted to ensure that proper procedures are used.

Definition of euthanasia for Stranded Cetaceans (adapted from AVMA 2007, 2013)

The Cetacean Euthanasia Working Group (CEWG) started by discussing the definition of euthanasia and throughout the workshop edited the definition. We agreed to the following:

> *Euthanasia, or the induction of humane death, of stranded cetaceans is often the sole choice for authorized responders to relieve pain, distress and suffering. The panel recognizes that without (humane) euthanasia, animals may be subject to hours or even days of pain, suffering and stress if left to die naturally. The techniques used should result in rapid loss of consciousness, followed by cardiac or respiratory arrest and the*

ultimate loss of brain function (AVMA 2007, 2013). Euthanasia methods should also be safe for all responders and minimize the risk for secondary intoxication of (scavengers and) predators and environmental contamination upon disposal.

General recommendations for euthanizing stranded cetaceans

The CEWG felt that a combination of general guidelines and species-specific observations would be the most effective way to impart information. These guidelines and observations are not meant to be rigid directives and the species-specific observations are living documents based largely on the collective experience of the group and not on systematic research. As more data and observations become available, we expect that changes will occur.

Responders and veterinarians should be aware that there are many uncontrolled and often unrecognizable variables associated with every live stranding event and, as a result, that each event will be different. The CEWG recognized that we cannot develop a 'cookbook' for cetacean euthanasia, and practitioners must understand that these are guidelines and not directives.

1) Whenever possible, it is preferable to avoid or minimize pain or distress resulting in a "gentler death" that may take a longer time, rather than a more rapid but stressful death. The CEWG recognizes that in some events pre-euthanasia sedation is appropriate prior to euthanasia, especially when administration of such is safe and there is sufficient time for the agent(s) to take effect prior to administration of the killing agent. Sedation can be accomplished with a single agent or a combination of analgesic and/or sedative agents IM and/or IV. Use of sedation and drug combinations MAY result in less drug volume (especially of euthanasia solution) which can help mitigate, but not eliminate, potential ecotoxicosis and secondary intoxication.

   AND

   The CEWG recognizes that for other events pre-euthanasia sedation is not indicated and believes that single agent administration of a euthanasia solution (IV) as approved by AVMA (2007, 2013) is appropriate and humane.

   AND

   Single agent administration of a euthanasia solution via intracardiac injection (IC) is not ideal, but is conditionally acceptable in stranded cetaceans in extenuating circumstances.
2) The CEWG DOES NOT recommend use of KCl alone.
3) When use of standard euthanasia solution(s) is not an option, pre-euthanasia sedation followed by KCl to effect is acceptable. Note that high volume of KCl may be needed and should be readily available. A rock salt form of KCl marketed for water softeners

is slow to go into solution, and should be mixed ahead of time. A finer granular form of KCl is available that can be mixed into solution more rapidly.

4) The CEWG recognizes that chemical euthanasia involves toxic substances and all efforts should be made to minimize secondary intoxication and environmental contamination (ecotoxicity). The panel urges all practitioners to dispose of waste drugs properly and in an environmentally acceptable manner.
5) The CEWG suggested the development of a matrix of cetacean euthanasia method choices available to stranding responders and veterinarians. Matrices for small and large cetacean euthanasia are included in the cetacean euthanasia guidelines.

Safety recommendations

Human safety should always be the top priority during these events. It is important to know your team's experience, skill and limitations, and to continually assess the safety of a situation. Mitigation of safety concerns should always include personal protective equipment (PPE) including gloves, masks or face shields.

Concerns/risks include, but are not limited to:

- Operating in water is risky, and NOT PREFERRED. The CEWG does not recommend swimming near a stranded animal. The CEWG DOES NOT RECOMMEND working close to a live cetacean in surf, especially in water deeper than knee deep
- Someone at the scene should have general first aid/CPR (EMS certification). Stranding response equipment should always include a first aid kit minimally. Floatable back boards, splints, and neck braces are recommended but optimal
- There is always a risk of accidental injection of potent drugs. Responders should also be aware of the possibility of broken hub/syringe and spray back of potent drugs. Be ready to respond quickly if spray back occurs (flush in first aid kit)
- Know the location of and quickest route to a hospital
- Carry or have ready access to MSDS and labels of chemicals and drugs used in the field
- Whenever possible use Luer lock syringes to reduce the likelihood of spray back
- Used needles and other sharps should always be disposed of in a sharps container
- NEVER assume that local EMS will have sedative reversal agents
- Reversal agents, if available for the drugs used, should be included in stranding first aid kits for EMS to administer @ site if potent drugs are used
- *Veterinarians and stranding response personnel are not licensed to treat human patients and should not administer reversals, but should be aware of the dose and concentration of potent drugs and the appropriate reversal agents and doses*

- Concentrated sedatives ARE NOT RECOMMENDED for euthanasia for most stranding response organizations. These require specialized safety measures.
- The responding organization should establish a 'Zone of Safety' around animal/drugs/tools (good use of local enforcement and/or inexperienced or volunteer responders)
- When a euthanasia is conducted in a public area, educating the public about the process is important
    - Prepare the public by explaining why the choice was made and what will happen and have talking points for less experienced volunteers and staff. It is often valuable for an experienced staff member to address the public
    - Allow time for members of the public to walk away
    - Answer questions prior to the event and afterwards if needed

Small Cetacean Euthanasia Matrix

#	Method	Drugs	Dose(s)	Route(s)	Pros	Cons	Concerns specific to euthanasia: Responder safety	Ecotoxicity	Public perception	Needs	Research needs
	Not chemically limited										
1	Sedation followed by euthanasia solution	see table of effective drugs and dosages	see table of effective drugs and dosages	IM (sedatives) IV, IC, IP,	we know it works	disposal concerns	moderate	high	low	Need more data on best sedatives and drug combinations, moderate length needles needed for larger animals, better guidance on safe IV delivery	Need to research tissue residue levels by disposal method; need more data on lowest effective dose & better weight estimators
2	euthanasia solution only		1ml/10lbs	IV, IC, IP	we know it works; relatively inexpensive	possible negative reactions, disposal concerns	moderate, if not in surf and not administering via fluke	high	low if no rxn		Need more data on lowest effective dose & better weight estimators; tissue residue levels
	Barbiturate limited										
3	Over-sedation	see table of effective drugs and dosages	see table of effective drugs and dosages	IM, IV	no barbiturate, can be achieved without controlled drugs	may take longer (~hour) in deep diving species, higher doses may still be a risk for relay toxicity	moderate	moderate	low	need to compile dosage data and protocol for determining when animal is over-sedated	
4	Sedation followed by KCl	see table of effective drugs and dosages	see table of effective drugs and dosages	IM, IV, IC, IP,	fewer chemicals, no barbiturate; volume KCl readily available and less expensive	requires deep sedation	moderate	low/moderate	low	need to compile dosage data and protocol for determining when animal is sedated enough to adminster KCl	
5	Sedation followed by exsanguination	see table of effective drugs and dosages	see table of effective drugs and dosages	IM, IV	fewer chemicals, no barbiturate, less expensive	requires deep sedation; needs training; disturbing to responders and public	high	low/moderate	high	need to compile dosage data and protocol for determining when animal is sedated enough, need better training and development specialized tools (two-bladed knives)	training and diagram(s) needed
	Chemically limited (for various reasons)										
6	exsanguination		NA	NA	no drugs; inexpensive	method of last resort; disturbing to responders and public	high	zero	high	need better training especially in thorascic cuts, two bladed knife?	training and diagram(s) needed
7	ballistics		NA	NA	no drugs, ammunition inexpensive	some responders need training and access to firearm, may be disturbing to responders and public	low	low	high	need better training, need to acquire appropriate weapons and ammunition; permitting	training and diagram(s) needed
8	natural death		NA	NA	no drugs	can take a long time; public safety; requires constant site/PR management	low	zero	high	need to educate local enforcement; NOAA deputize local enforcement; public safety issues	

Large Whale Euthanasia Matrix

#	Method	Drugs	Dose(s)	Route(s)	Pros	Cons	Concerns specific to euthanasia: Responder safety	Ecotoxicity	Public perception	Needs	Research needs
	Not chemically limited										
1	Sedation followed by euthanasia solution	see table of effective drugs and dosages	see table of effective drugs and dosages	IM, IV, IC, IP, IT, RB, IN	we know it works if we have enough drugs	need large drug volumes on hand; expensive, availabilty issues	moderate	high	low	Safe delivery of drugs for animal in water ; very long needles just developed, may need to stock-pile drugs	Need to research pole delivery of sedation; tissue residue levels
2	Euthanasia solution only			IV, IC, IP, IT	we know it works; inexpensive	need large drug volumes on hand; possible animal reactions	high	high	low if no rxn	Safe delivery of drugs for animal in water; very long needles just developed; may need to stock-pile euthanasia solution	Need to research pole delivery of euthsolution; tissue residue levels
	Barbiturate limited										
3	Over-sedation	see table of effective drugs and dosages	see table of effective drugs and dosages	IM, IV	no barbiturate, can be achieved without controlled drugs	drug volumes needed may not be readily available, may take >hr, higher doses may still be a risk for relay toxicity	moderate	moderate	low	need to compile dosage data and protocol for determining when animal is over-sedated; may need to stock-pile sedatives	
4	Sedation followed by KCl	see table of effective drugs and dosages	see table of effective drugs and dosages	IM, IV, IC, IP, IT, RB, IN	fewer chemicals, no barbiturate; volume KCl readily available and inexpensive	requires deep sedation	moderate	low/moderate, depending on agent, more info needed	low	safe delivery of drugs for animal in water ; very long needles just developed; need to compile dosage data and protocol for determining when animal is over-sedated;	
5	Sedation followed by exsanguination	see table of effective drugs and dosages	see table of effective drugs and dosages	IM, IV	fewer chemicals, no barbiturate	requires deep sedation; need training	high	low/moderate, depending on agent, more info needed	high	need better training; need to compile dosage data and protocol for determining when animal is over-sedated; need to design knife or lance (long, thin double bladed semi-stiff, knife-stiletto)	training and diagram(s) needed
	Chemically limited (for various reasons)										
6	Exsanguination		NA	NA	no drugs; inexpensive	method of last resort	high	zero	high	need better training, need to design knife or lance	training and diagram(s) needed
7	Cranial implosion (Cochran et al. 2012)		NA	peri-cranial	no drugs	logistics, training, carcass destruction, not foolproof	high	zero	unknown/needs education	legal issues regarding explosives handling, supply,	Can only be a reality in the US if federally supported for training/deployment (DOD; mining, demolition expts) directed by NOAA
8	Ballistics		NA	NA	no drugs, ammunition inexpensive	not on animals >7m	low	zero	high	need better training, need to acquire appropriate weapons and ammunition; permitting	training and diagram(s) needed
9	Natural death		NA	NA	no drugs	can take a long time; public safety; requires constant site/PR management	low	zero	high	need to educate local enforcement NOAA deputize; local enforcement public safety issues	

Summary of the Most Effective Euthanasia Methods as per the Cetacean Euthanasia Working Group (1 of 2)

(This is NOT a list of recommended drugs, but rather a list of drugs found to be historically effective.)

			AGENT 1				AGENT 2				AGENT 3			
Category	**# of agents**	***Acceptability**	**Generic Name**	**Dose**	**Route**	**Comments**	**Generic Name**	**Dose**	**Route**	**Comments**	**Generic Name**	**Dose**	**Route**	**Comments**
NOT CHEMICALLY LIMITED														
Heavy Sedation followed by euthanasia solution *When possible allow 20 -30 minutes between sedatives and euthnasia solution*	2	*Acceptable	Meperidine	4 mg/kg	IM	Usual dose of 2 mg/kg IM, wait 20 minutes then collect samples. If animal still too alert, may be followed by 2 mg/kg IV prior to euthansia solution adminstration.	Sodium Pentabarbitol	80 mg/kg or to effect	IV	1.5X dose if adminstered IP or IH				
Heavy Sedation followed by euthanasia solution *When possible allow 20 -30 minutes between sedatives and euthnasia solution*	3	*Acceptable	Acepromazine	1 mg/kg	IM	Adverse reaction has been observed when administered to *Delphinus* .	Xylazine	2 mg/kg	IM	Allow approximately 10 min for acepromazine to take effect prior to xylazine administration. Adverse reactions have been observed when administered to Grampus.	Sodium Pentabarbitol	80 mg/kg or to effect	IV	1.5X dose if adminstered IP or IH
Euthanasia solution only	1	*Acceptable	Sodium Pentobarbital	80 mg/kg or to effect	IV	May be associated with mild to severe reaction post injection eliciting a negative public response. (if administered IV). Conditionally acceptable for other routes (e.g. IP, IC) if animal is heavily sedated.								

"Normal Sedation" drug dosages for cetaceans

Butorphanol	0.05 to .1mg/kg IM	Has been used in rigtht whales at 0.1 mg/kg with midazolam at 0.1 mg/kg
Diazepam	0.05 to 0.1 mg/kg IM	Less consistant than midazolam IM
Meperidine	0.5 to 2 mg/kg IM	Has been used in conjunction with midazolam at 1 mg/kg or higher
Midazolam	.05 to .1 mg/kg IM	Can be given at higher levels and in combination with other drugs (*e.g.* Meperidine , Butorphanol)

Cetacean stranding situations and species can be extremely complex and diverse. Taking this into consideration ,below we have listed dosages for several agents NOT typically used in clinical cetacean sedation for those times when these are the only options available. These drugs are not generally used in healthy animals but may be considered for euthanasia if no other drugs are available. Typically the dosage for euthanasia is three times the theorectical dose in order to avoid suffering and promote a quiet death.

Other drugs used for cetacean euthanasia protocols BUT NOT USED FOR CLINICAL CETACEAN SEDATION

Summary of the Most Effective Euthanasia Methods as per the Cetacean Euthanasia Working Group (1 of 2)

(This is NOT a list of recommended drugs, but rather a list of drugs found to be historically effective.)

			AGENT 1				AGENT 2				AGENT 3				AGENT 4			
Category	# of agents	*Acceptability	Generic Name	Dose	Route	Comments	Generic Name	Dose	Route	Comments	Generic Name	Dose	Route	Comments	Generic Name	Dose	Route	Comments
CONTROLLED DRUG LIMITED																		
Oversedation *It may require 3 x the normal dosage to accomplish euthanasia. Over-sedation with other drugs would also be acceptable, refer to the survey-based tables and use clinical judgement*	2	*Acceptable *(Once effects of IM administration are evident, it is acceptable to administer subsequent doses IC)*	Acepromazine	1 mg/kg	IM	Allow approximately 10 minutes between admintrations. Repeat as needed to accomplish euthanasia. Adverse reaction has been observed when administered to *Delphinus*.	Xylazine	2 mg/kg	IM	Allow approximately 10 min for acepromazine to take effect prior to xylazine administration. Adverse reactions have been observed when administered to Grampus without prior administration of another sedative/tranquilizer. Repeat as needed to accomplish euthanasia.								
Heavy Sedation followed by KCl		**Conditionally Acceptable ONLY if animal is heavily sedated.	Midazolam	0.05-0.1 mg/kg	IM	Omit if not available or if controlled drugs are not an option. Allow approximately 10 minutes between admintrations. Administer sedatives prn sequentially to effect prior to KCL.	Acepromazine	0.2-1 mg/kg	IM	Allow approximately 10 minutes between admintrations. Administer sedatives prn sequentially to effect prior to KCL. Adverse reaction has been observed when administered to *Delphinus*.	Xylazine	3-4 mg/kg	IM *(or IV if safe)*	Allow approximately 10 minutes between admintrations. Administer sedatives prn sequentially to effect prior to KCL. Adverse reactions have been observed when administered to *Grampus*.	KCL Administer supplementary doses of sedatives as necessary to render animal unresponsive to KCL injection	1-2 mmol/kg (75-150 mg/kg)	IC	IV may require higher dose than IC. Especially when carcass removal is an issue (ecotoxicological impact)
Heavy Sedation followed by exanguintation		**Conditionally Acceptable ONLY: 1) If animal is heavily sedated; 2)personnel is skilled and knowledge about proper severing sites	Midazolam	0.05-0.1 mg/kg	IM	Omit if not available or if controlled drugs are not an option. Allow approximately 10 minutes between admintrations. Administer sedatives prn sequentially to effect prior to exsanguination.	Acepromazine	0.2-1 mg/kg	IM	Allow approximately 10 minutes between admintrations. Administer sedatives prn sequentially to effect prior to KCL. Adverse reaction has been observed when administered to *Delphinus*.	Xylazine	3-4 mg/kg	IM *(or IV if safe)*	Allow approximately 10 minutes between admintrations. Administer sedatives prn sequentially to effect prior to KCL. Adverse reactions have been observed when administered to Grampus.	Exsanguinate	Need better information and training. This method requires a solid anatomic knowledge, training and appropriate tools. Sites may include: ventral peduncle, brachial artery; *deep neck area *(*noting that cetaceans do not have large superficial carotid arteries)*		
Ballistics		**Conditionally Acceptable in small cetaceans ONLY if personnel are: well trained and knowledgeable	Need better information and training. Techniques can be found in RSPCA (1997) for small cetaceans, Blackmore (1995) for pilot whales and Suisted (1999) for baleen whales				Shooter must have proper equipment and knowledge of cetacean cranial anatomy. Site must have strict crowd contral.			Disarticulating the brain from the spinal cord at the occipital condyles is the goal. The recommended target area in guidelines is 1/2 way between the eye and flipper insertion with either dorsal or lateral entry. Never fire with barrel in direct contact with the animal. Be sure caliber size matches animal. Projectiles track off target with thick soft tissue coverage. Do NOT fire at the melon from the front of the animal, bullet may ricochet.					Not acceptible with sperm whales >7m. Conditionally acceptable in cetaceans under 7m in length.			
Explosives		Not currently acceptable in US	See Coughran *et al.* 2012				Must have qualified, permitted personnel and directional or shaped charges (see reference).								Permitting is likely to be a problem in the US.			

"Normal Sedation" drug dosages for cetaceans

Butorphanol	0.05 to .1mg/kg IM	Has been used in rigtht whales at 0.1 mg/kg with midazolam at 0.1 mg/kg
Diazepam	0.05 to 0.1 mg/kg IM	Less consistant than midazolam IM Meperidine 0.5 to 2 mg/kg IM. Has been used in conjunction with midazolam at 1 mg/kg or higher
Midazolam	.05 to .1 mg/kg IM	Can be given at higher levels and in combination with

Cetacean stranding situations and species can be extremely complex and diverse. Taking this into consideration ,below we have listed dosages for several agents NOT typically used in clinical cetacean sedation for those times when these are the only options available. These drugs are not generally used in healthy animals but may be considered for euthanasia if no other drugs are available. Typically the dosage for euthanasia is three times the theorectical dose in order to avoid suffering and promote a quiet death.

Other drugs used for cetacean euthanasia protocols BUT NOT USED FOR CLINICAL CETACEAN SEDATION

Acepromazine (1 mg/kg); Medatomadine (40-80 mcg/kg); Detomidine (30-60 mcg/kg); Xylazine (2mg/kg)

*** Acceptable methods: most consistently result in most humane cetacean euthanasia and fewer public safety hazards.***

*****Conditionally Acceptable methods: not considered humane and greater potential for public safety hazards.***

Regression equations for total body length (L, in cm) versus total body weight (kg) for eight cetacean species stranded in North Carolina and Virginia, USA

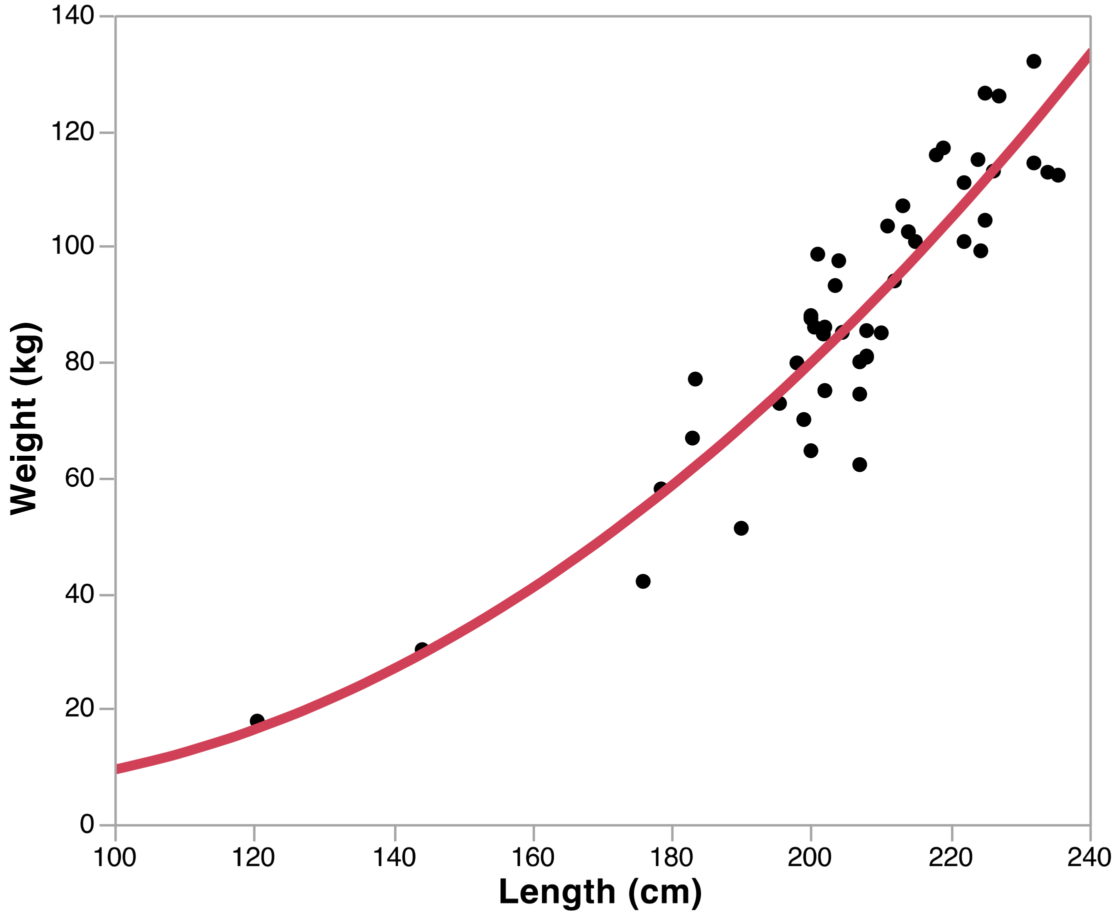
(see figures on following pages for weight ranges used to create equations)

Species	N	Weight (kg) =	R^2
Delphinus delphis	49	$0.0044927*(L – 205.912)^2 + 1.2063757*L – 161.63$	0.828
Grampus griseus	20	$0.0056553*(L – 241.59)^2 + 1.9757271*L – 312.08$	0.897
Kogia breviceps	42	$0.0050229*(L – 238.419)^2 + 2.1917378*L – 303.50$	0.895
Kogia sima	33	$0.0049206*(L – 202.221)^2 + 1.7564184*L – 210.58$	0.899
Phocoena phocoena	53	$0.0072798*(L - 116.683)^2 + 0.3895724*L – 22.00$	0.573
Stenella coeruleoalba	31	$0.003495*(L – 201.903)^2 + 1.2171744*L – 150.09$	0.946
Stenella frontalis	29	$0.0061927*(L – 203.924)^2 + 1.525497*L – 207.59$	0.837
Tursiops truncatus	171	$0.004468*(L – 196.833)^2 + 1.3728948*L – 168.61$	0.939

Delphinus delphis length-weight regression graph

$$W = 0.0044927*(L - 205.912)^2 + 1.2063757*L - 161.63$$

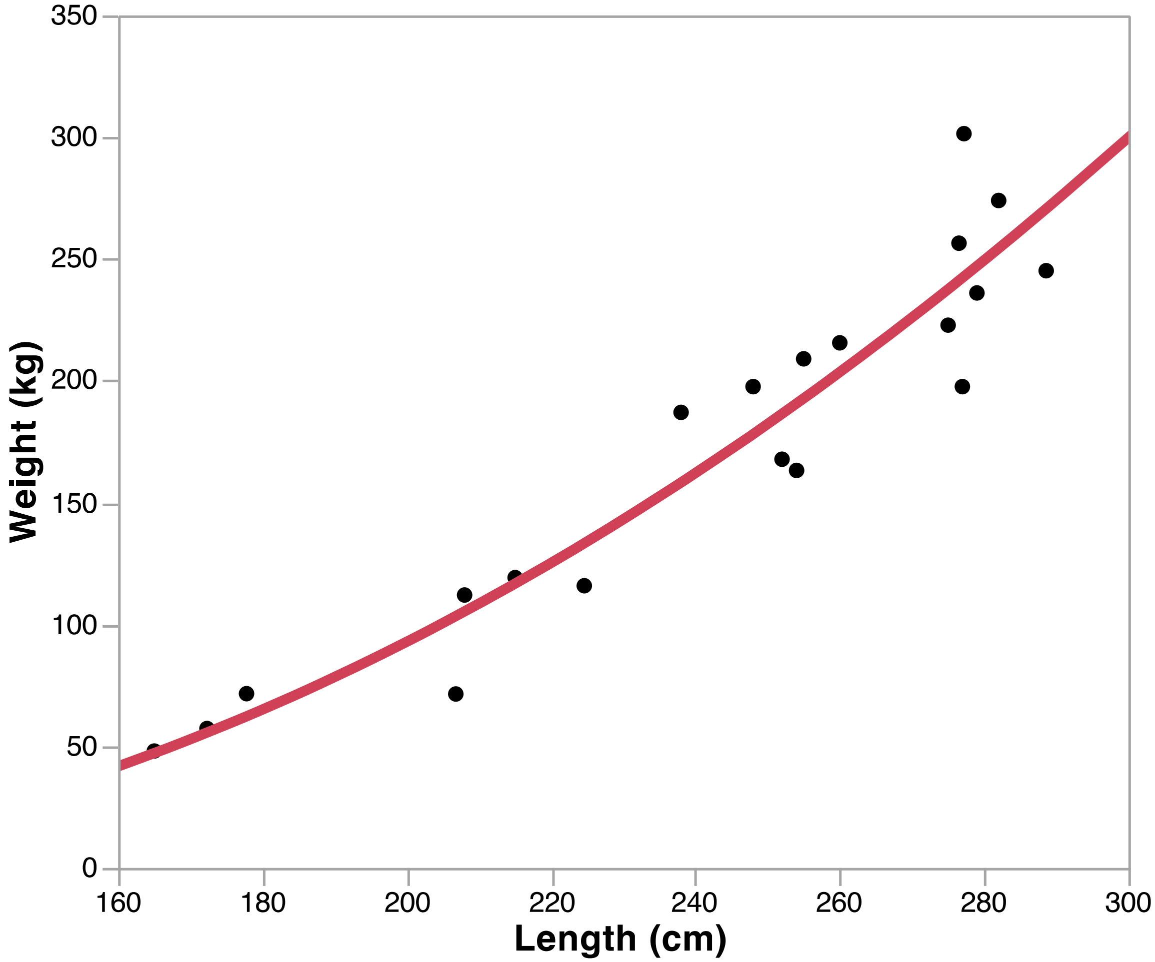
$N = 49$, $R^2 = 0.828$



Grampus griseus length-weight regression graph

$$W = 0.0056553*(L - 241.59)^2 + 1.9757271*L - 312.08$$

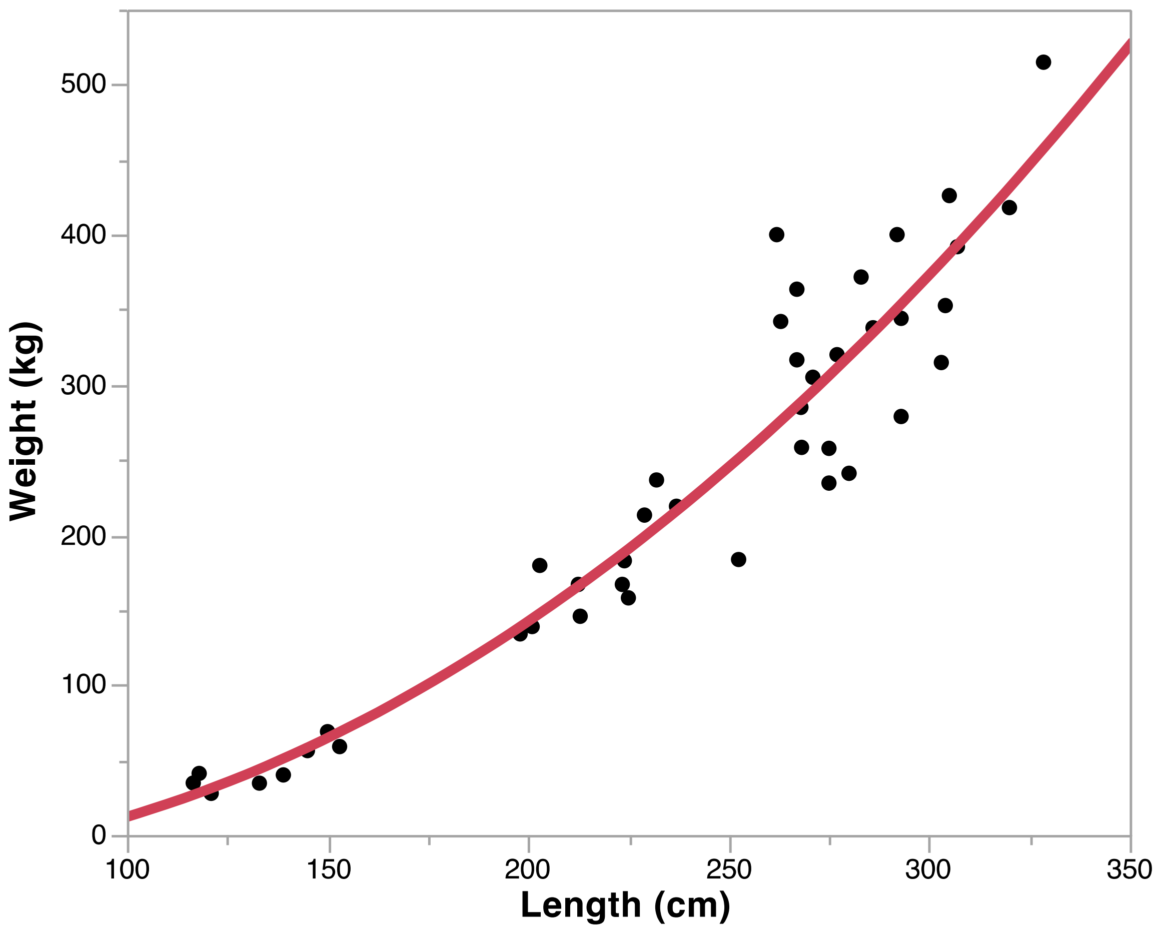
$N = 20$, $R^2 = 0.897$



Kogia breviceps length-weight regression graph

$$W = 0.0050229*(L - 238.419)^2 + 2.1917378*L - 303.50$$

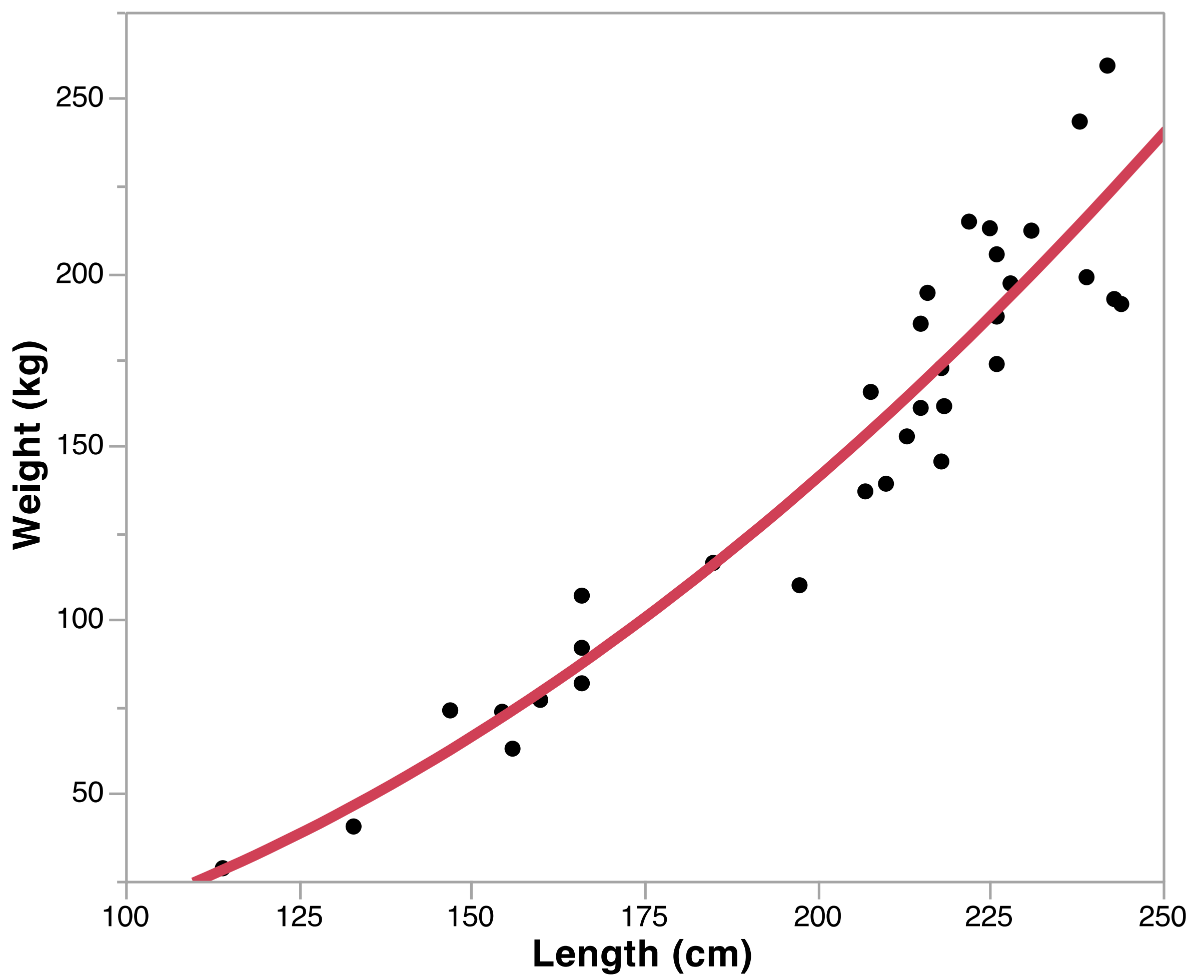
$N = 42$, $R^2 = 0.895$



Kogia sima length-weight regression graph

$$W = 0.0049206*(L - 202.221)^2 + 1.7564184*L - 210.58$$

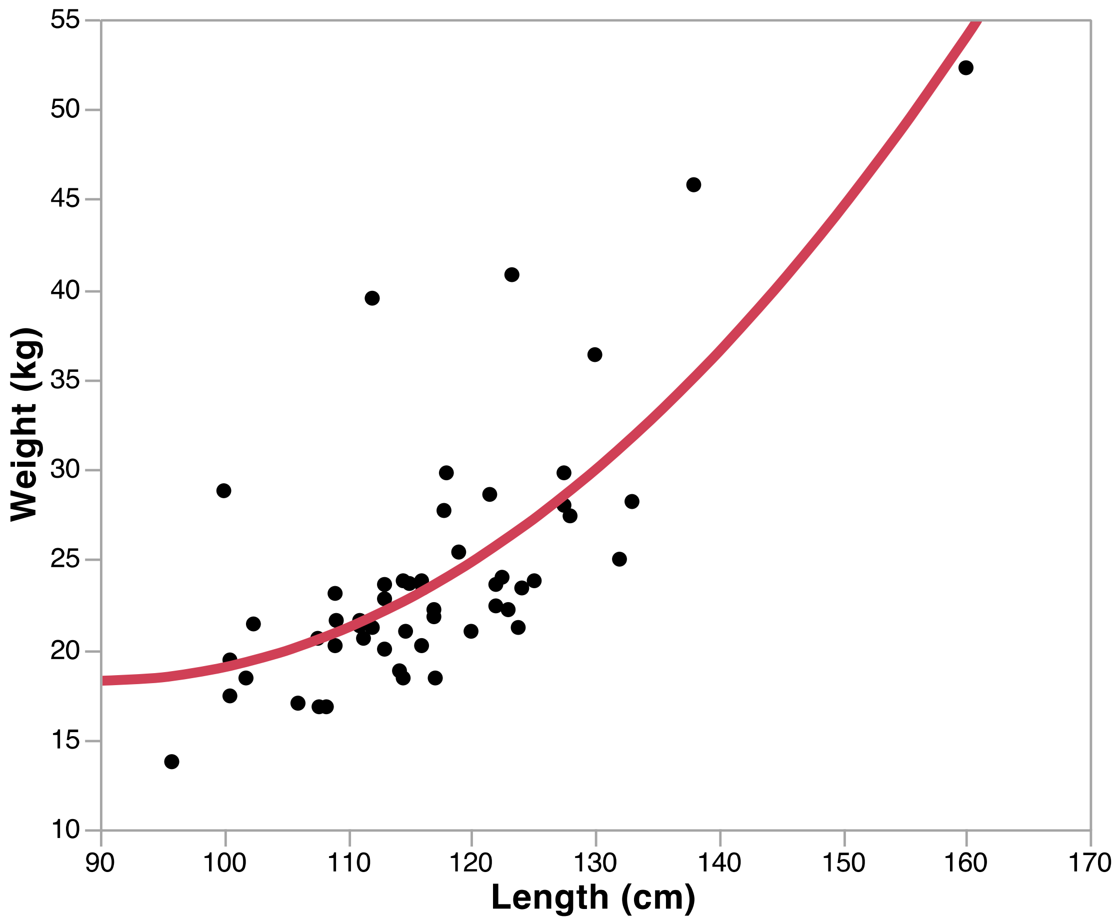
$N = 33$, $R^2 = 0.899$



Phocoena phocoena length-weight regression graph

$$W = 0.0072798*(L - 116.683)^2 + 0.3895724*L - 22.00$$

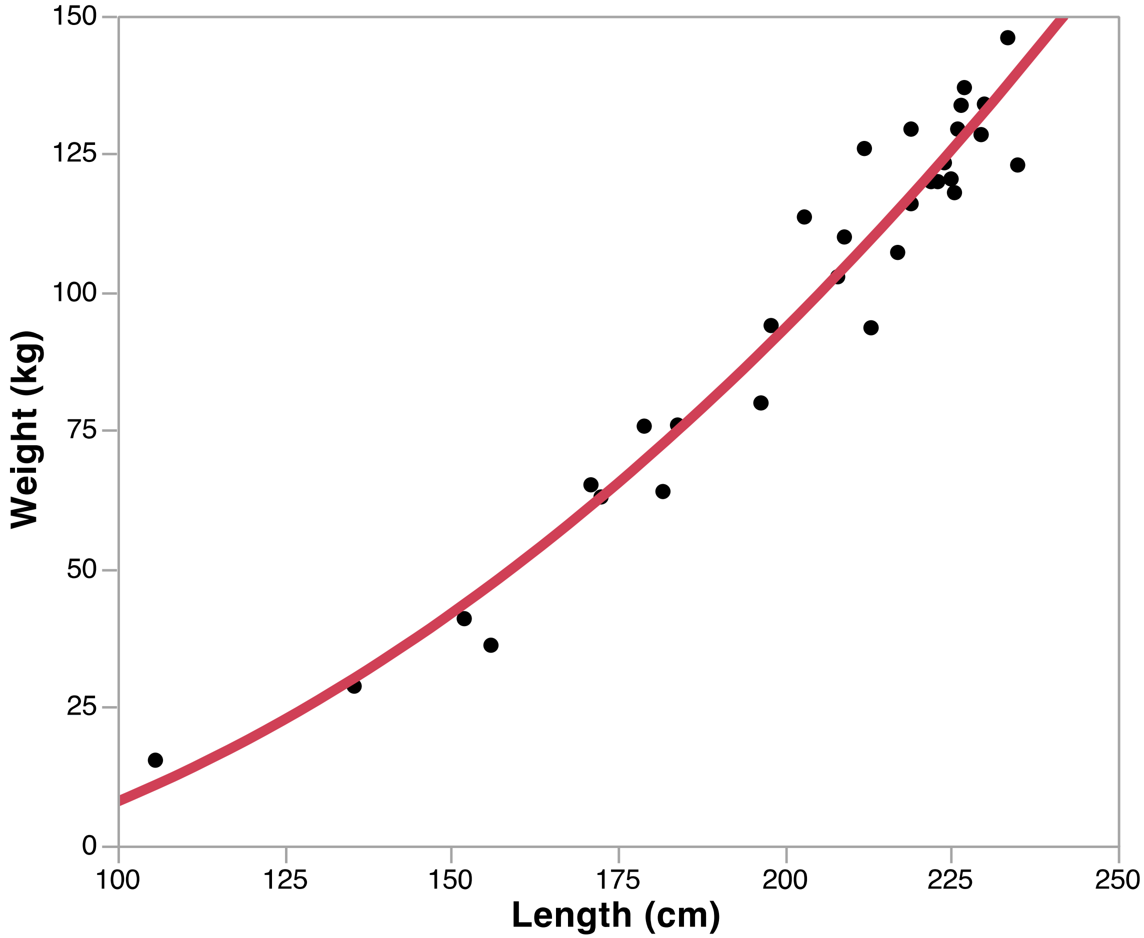
$$N = 53, R^2 = 0.573$$



Stenella coeruleoalba length-weight regression graph

$$W = 0.003495*(L - 201.903)^2 + 1.2171744*L - 150.09$$

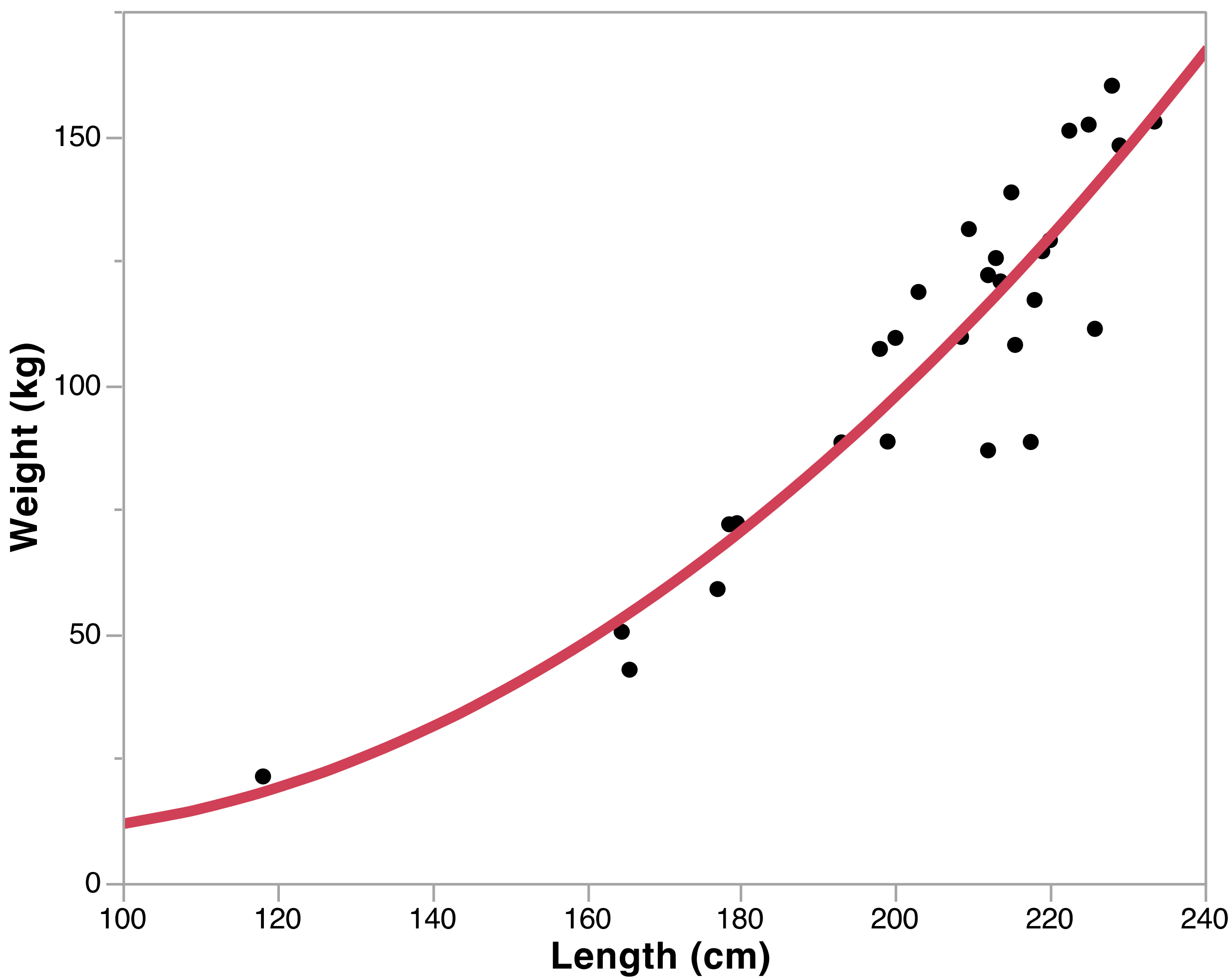
$$N = 31, R^2 = 0.946$$



Stenella frontalis length-weight regression graph

$$W = 0.0061927*(L - 203.924)^2 + 1.525497*L - 207.59$$

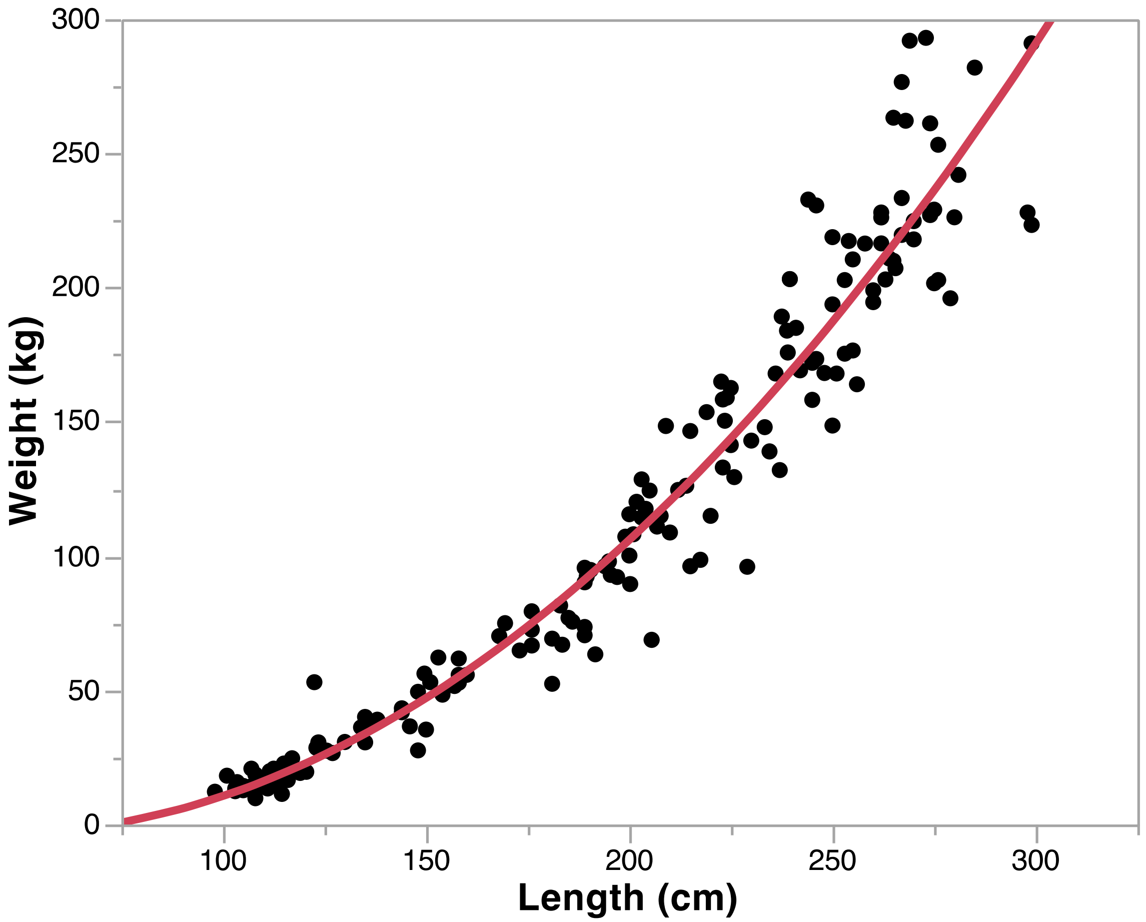
$N = 29$, $R^2 = 0.837$



Tursiops truncatus **length-weight regression graph**

$$W = 0.004468 * (L - 196.833)^2 + 1.3728948 * L - 168.61$$

$$N = 171, R^2 = 0.939$$



SPECIES-SPECIFIC OBSERVATIONS

Cetacean species are adapted to a wide variety of habitats and niches and these adaptations likely result in different behaviors and reactions to live stranding events and to drug administration. Below are observations by the CEWG regarding specific species:

Delphinus delphis

- For this species, handling is very important. *Delphinus* seem to be very sensitive compared to other species and require minimal handling and minimal sound. As much as is safe, a hands-off approach to this species appears to work best
- Observing and working with animals in enclosed spaces (ex, enclosed vehicles) seems to result in calmer animals as compared to on beach exposed to the elements. However, safety of responders is paramount
- Single agent euthanasia (IV) may be the most effective chemical means of euthanasia for this species
- Violent movement at the time of death (with or without human intervention) is not uncommon and may be more likely in warmer weather

Kogia spp.

- Pre-euthanasia sedation is recommended for these species. Using acepromazine IM @ (1 mg/kg IM) followed by xylazine IV (2 mg/kg IM) 15 minutes later or midazolam alone IV (0.1 mg/kg) appears to be an effective sedative. Administration of diazepam and meperidine at 2mg/kg IM and after 20min administration of 2mg/kg euthanasia solution IV has also been used effectively on *Kogia*
- *Kogia sima* has a greater tendency to react more violently to sedatives than *Kogia breviceps*. Sedation alone, when responders are limited to non-controlled or non-barbiturate drugs, may take a long time (>45min- up to several hours) in *K. sima*

Grampus griseus

- Alpha-II agonists (xylazine, medetomidine) are NOT recommended for this species as an initial treatment (adverse reactions have been observed including spinning, flipping and violent retching)
- Diazepam IV (or IM if IV not possible) seems to be effective at sedation

Tursiops spp.

- The *Tursiops* that strand on U.S. beaches are currently considered different morphotypes, but are likely to be multiple species could potentially react differently.
- Pre-euthanasia sedation has been effective with stranded *Tursiops*

Globicephala spp.

- *Globicephala melas* were euthanized with a sedative combination of acepromazine and xylazine during a mass stranding event in 2005 and the animals went down quietly but slowly (45 min)
- *Globicephala macrorhynchus* were euthanized with acepromazine and xylazine followed by KCl during mass stranding events in 2014, and some of the animals went through a prolonged (15-20 min) agonal response when KCl was administered (D. Boyd and G. Lovewell, pers. comm.).

Steno bredanensis

- Pre-euthanasia sedation has been effective with *Steno*

Common Needle Choices for Various Sizes of Cetaceans & Methods of Injection

Things to remember when choosing appropriate needles:

- When deciding on needle size (gauge and length), consider the thickness of the skin, blubber and muscle as well as the amount and consistency of agent, the potential temperature impact on agent, etc.
- Luer locking syringes decrease the chance of a disconnection between needle and syringe during administration
- When using > 2" needles, styleted needles are optimal to decrease chance of lumen impaction
- Use of butterfly needles or extension sets decrease chances of dislodging needles if an animal moves AND allows for easier ability to switch syringes

Animal Size	Example of Species	Intramuscular (IM)		Peripheral Intravenous (IV)		Caudal Peduncle (lateral or ventral approach)		**Intraperitoneal (IP)		**Intrahepatic		+Intracardiac (IC)		Via Blowhole (Use flexible catheter, or no catheter and administer on inspiration)		+Retrobulbar		++Common Brachiocephalic Vein	
		Gauge	*Length	Gauge	*Length	Gauge	*Length	Gauge	*Length	Gauge	*Length	Gauge	*Length	Gauge	*Length	Gauge	*Length	Gauge	*Length
Small (<6')	Harbor Porpoise, Dall's Porpoise, calves, juveniles	20-22G	1-1.5"	20-22G	0.5-1"	20-22G	1-1.5"	20-22G	2.5-3.5"	20-22G	2.5-3.5"	18-20G	3.5-6"	5–9Fr (14–16 G)	2-3"	20-22G	2.5-3.5"	16-18G	3-5."
Med (6-12')	Common Dolphin, Bottlenose Dolphin	18-22G	2-3.5"	18-22G	1-1.5"	18-22G	1.5-3"	18-22G	3.5-8"	18-22G	3.5-8"	18-20G	6-12"	9–12Fr	2.5–5"	18-22G	3.5-8"	16G	5.5"
Large (12-25')	Pilot Whale	18-21G	3.5-6"	18-22G	1-2"	18-22G	3-5"	18-22G	8-12"	18-22G	8-12"	16-20G	12-15"	9–15Fr	6-10'	18-22G	8-12"	16G	5.5"
Large Whales (>25')***	Baleen Whales, Sperm Whales	16-20G	>8-10"	16-20G	1.5-3.5"	16-20G	5-10"	16-20G	>12"	16-20G	>12"	>18"G	12->20"	9–15Fr	>12"	16-20G	>12"	NA	NA

*Custom made needles (ex. length over 8-10") may be required

**IP and IH Injections

- A higher dose of barbiturate is required for intraperitoneal and intrahepatic euthanasia
- An animal should be sedated prior to administration

***See Harms *et al.* 2014

+IC and Retrobulbar Injections

- An animal must be heavily sedated or comatose prior to administration

++Common Brachiocephalic Vein

- Ultrasound guidance, when available, simplifies accession of the vessel, otherwise inserting the needle roughly 2 cm cranial to the cranial aspect of the sternum, directly perpendicular to the long access of the animal, and perfectly parallel to the ground increases the likelihood of access
- The animal should always be placed in right lateral recumbency for this procedure. Animals tend to accept lateral recumbency with less resistance if they have been previously sedated.

Aggregate drug doses by weight, in mg/kg, for 282 cetacean euthanasias.

The median number of drugs used for euthanasia was 1, with a minimum of 1 and a maximum of 6 agents.

Drug	N	Median	Quartiles	Minimum, Maximum	Mean	Standard Deviation
Midazolam	19	0.10	0.03, 0.40	0.0085, 0.96	0.235	0.274
Diazepam	31	0.19	0.12, 0.27	0.015, 1.4	0.263	0.277
Acepromazine	39	0.78	0.33, 1.3	0.03, 5.8	1.0	1.1
Acepromazine not followed by pentobarbital	28	0.95	0.44, 1.3	0.04, 5.8	1.2	1.2
Acepromazine followed by pentobarbital	11	0.43	0.10, 0.90	0.03, 1.8	0.56	0.54
Telazol*	7	1.2	0.9, 6.6	0.4, 8.4	3.4	3.3
Ketamine	1	5			5	
Butorphanol	9	0.05	0.04, 0.2	0.01, 0.9	0.17	0.28
Meperidine	7	1.2	0.2, 2.5	0.04, 13.7	2.8	4.9
Morphine	1	0.11			0.11	
Xylazine	66	3.6	1.3, 11.8	0.14, 152	12.8	27.1
Xylazine not followed by pentobarbital	35	4.1	1.7, 13.9	0.42, 152	17.8	35.6
Xylazine followed by pentobarbital	31	2.6	1.1, 10.5	0.14, 44.6	7.1	9.6
Detomidine	2	0.22	0.11, 0.33	0.11, 0.33	0.22	0.16
Medetomidine	3	0.084	0.0022, 0.62	0.0022, 0.62	0.237	0.338
Pentobarbital	239	130	87.3, 190	1.8, 621	152	104
Pentobarbital with alpha-2 agonist premedication	34	100	58, 214	1.8, 339	125	98
Pentobarbital without alpha-2 agonist premedication	166	133	95, 178	2.3, 546	145	82
Phenytoin**	33	8.8	4.4, 13.0	0.22, 46.0	10.0	8.9
Embutramide***	1	2.4			2.4	
KCl	2	166	111, 222	111, 222	166	78.5

Notes:
**dose listed as the combined tiletamine and zolazepam total*
***always combined with pentobarbital, typically as sodium pentobarbital 390 mg/ml and phenytoin sodium 50 mg/ml in DEA schedule III formulations*
****as T-61 Euthanasia Solution: Embutramide (200 mg/ml) is combined with mebezonium iodide (50 mg/ml) and tetracaine hydrochloride (5 mg/ml)*

There is a tendency for acepromazine and xylazine to be dosed lower when followed by euthanasia solution than when not followed by euthanasia solution, but the differences are not statistically significant (Wilcoxon rank sums test, $p = 0.0529$ for acepromazine and $p = 0.1594$ for xylazine). Use of any premedication or not has little effect on dose of pentobarbital used ($p = 0.5861$), so pentobarbital doses are not separated by use or non-use of a premedication. Pentobarbital doses are, however, somewhat lower when an alpha-2 agonist (xylazine, detomidine, or medetomidine) is included as a premedication ($p = 0.047$), and these differences are included in the table. There is no statistically significant difference in pentobarbital doses when a benzodiazepine (diazepam or midazolam) is included as a premedication ($p = 0.2071$), in fact the pentobarbital dose is slightly higher when preceded by a benzodiazepine (data not shown). Pentobarbital doses are significantly lower when acepromazine is included as a premedication ($p = 0.0035$), but in all 11 cases where acepromazine precedes pentobarbital use, xylazine is also included as a premedication (data not shown). No other premedications were employed frequently enough for meaningful statistical analysis.

Drug doses by length for *Globicephala* spp. (*G. macrorhynchus* (n=5), *G. melas* (n=19)

Suitable weight/length charts could not be generated or located for *Globicephala* spp., so in order to retain and present useful data, doses for these two species are presented per unit length. Calculations based on weight are preferable when possible, because doses do not scale linearly with length, but do scale close to linearly with weight. Median number of agents used is 1, with a minimum of 1 and maximum of 3 agents.

Drug	N	Median (mg/meter of length)	Quartiles (mg/meter of length)	Min, Max (mg/meter of length)	Mean (mg/meter of length)	Standard Deviation
Diazepam	3	9.5	7.5, 16.0	7.5, 16.0	11.0	4.5
Acepromazine	5	27.9	18.1, 54.5	8.3, 80.3	34.6	26.9
Telazol	1	215			215	
Xylazine	7	415	279, 1538	163, 1675	699	632
Pentobarbital	20	7932	5086, 13231	218, 16619	8604	4785
Phenytoin	3	492	28, 785	28, 785	435	381

Pentobarbital doses (mg/kg) for the six most commonly encountered species, irrespective of premedication

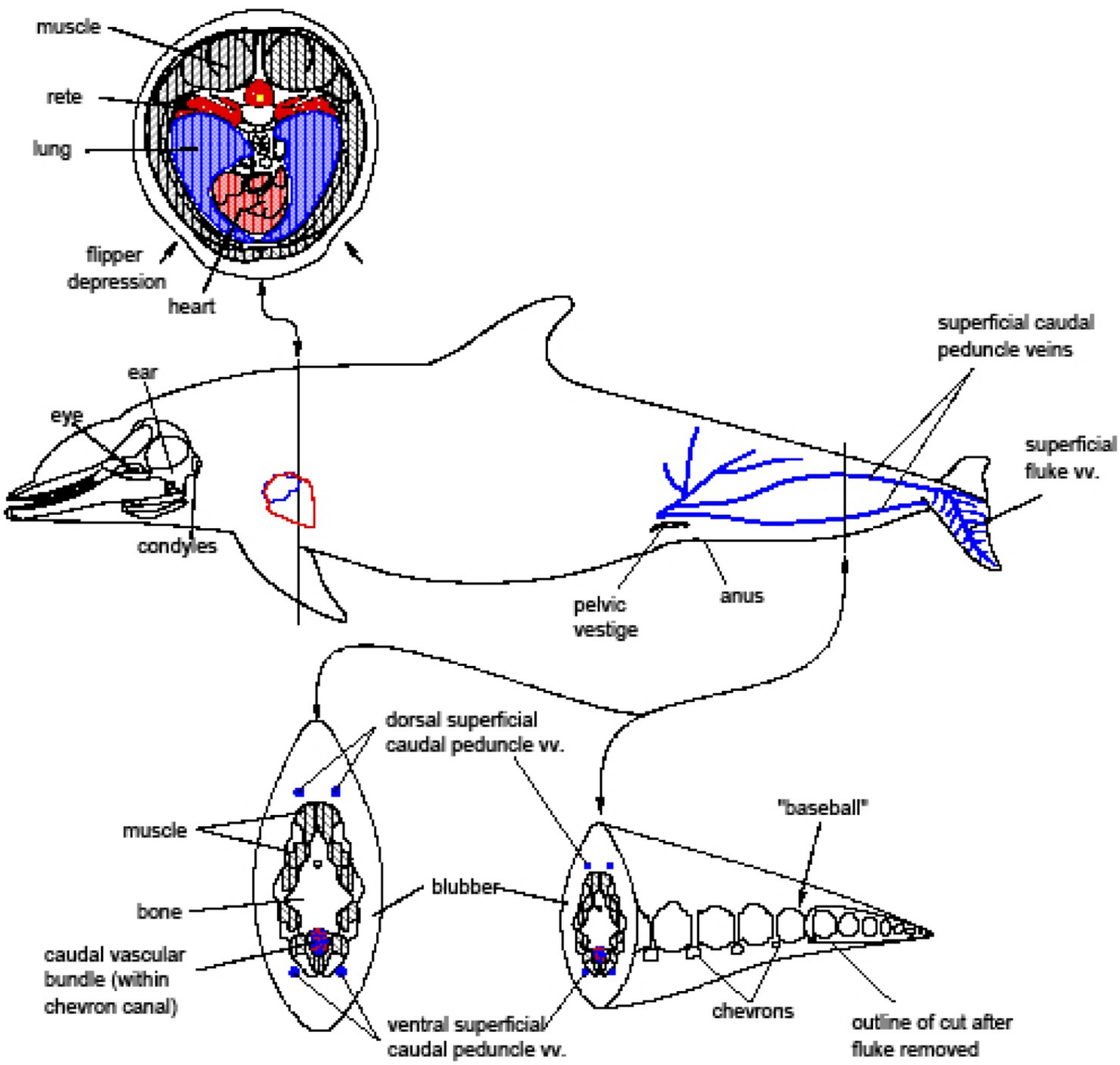
Doses were compared among species by the Kruskal-Wallis test, followed by the Steel-Dwass method for multiple comparisons. Doses were significantly different for pentobarbital ($p < 0.0001$), but did not differ significantly for any other drug. Pentobarbital doses for *Delphinus delphis* were significantly higher than for *Grampus griseus*, *Tursiops truncatus*, *Kogia breviceps* and *Lagenorhynchus acutus*, but did not differ significantly between any other species pairs.

Species	N (receiving pentobarbital)	Median	Quartiles	Minimum, Maximum	Mean	Standard Deviation
Delphinus delphis	73	151	128, 194	46, 471	168	65
Phocoena phocoena	31	143	116, 214	1.8, 553	198	156
Grampus griseus	21	115	70, 163	34, 233	120	56
Tursiops truncatus	16	103	65, 154	3.1, 260	111	71
Lagenorhynchus acutus	40	101	78, 166	15, 504	129	95
Kogia breviceps	27	81	69, 173	2.7, 325	115	81

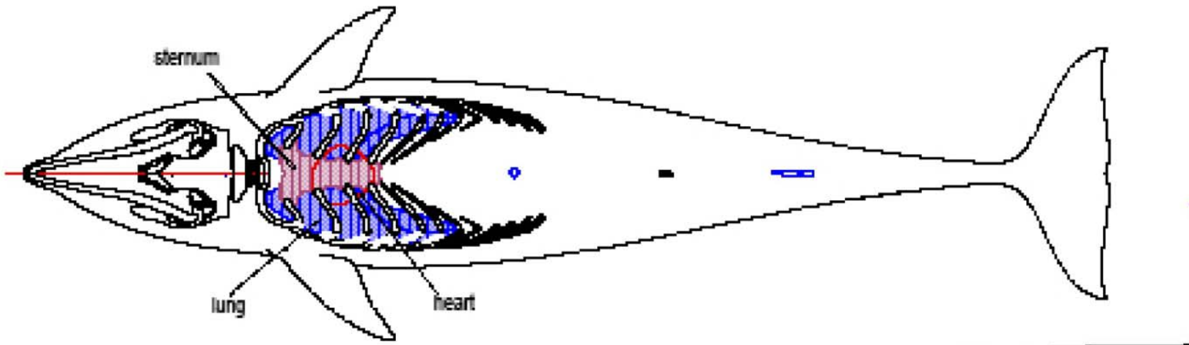
Small Cetacean Illustrations

(Illustrations provided by Sentiel Rommel, UNC Wilmington)

Location of heart in cross section and laterally with flipper as landmark for intracardiac injection and knife/lance access for exsanguination.

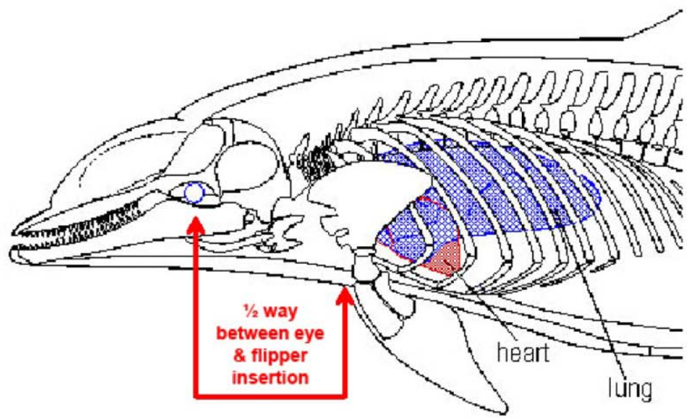


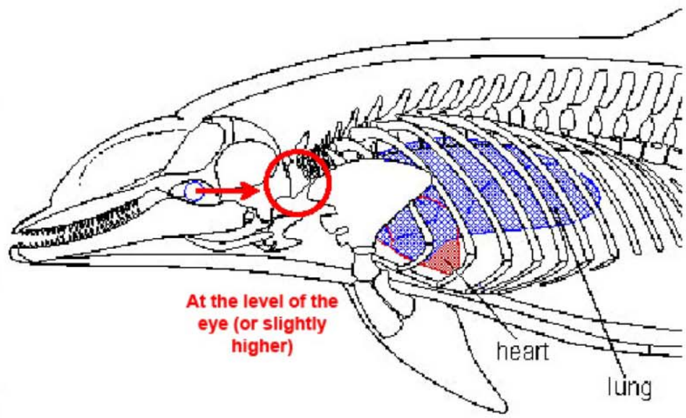
Lateral and cross sectional view of peduncle vessels for intravenous access and knife/lance placement for exsanguination. The caudal vascular bundle is the target for exsanguination. Note that caudal to the anus, access to this vascular bundle is blocked by the chevron bones and a cutting instrument must cut between the chevron bones.

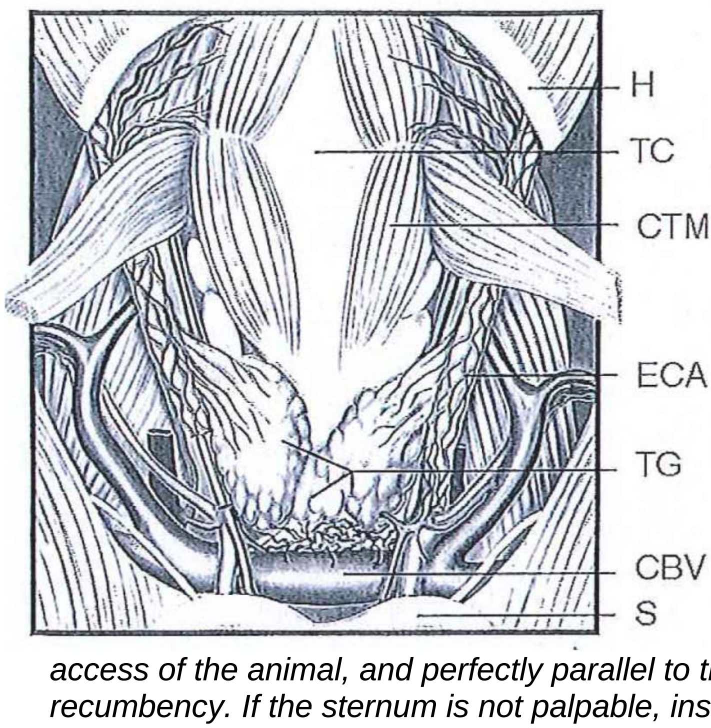


Outline of skeletal elements and target area for euthanasia of small cetaceans using ballistics. The goal is to aim at the occipital condyles and to disarticulate the skull from the spinal column. (Images above and to the right by S. Rommel)

Donoghue (2006) recommends a series of three shots in a line halfway between the eye and the insertion of the flipper at the level of the eye. The area can be accessed laterally, dorsally or ventrally (above).







The common brachiocephalic vein (CBV) in the ventral neck of a bottlenose dolphin. This vessel, although ideally located with the aid of ultrasound, can be approached 'blind' in the following manner: if the cranial aspect of the sternum is palpable, insert the needle roughly 2cm cranial to the cranial aspect of the sternum, directly perpendicular to the long access of the animal, and perfectly parallel to the ground in a (sedated) animal in right lateral recumbency. If the sternum is not palpable, insert the needle on the ventral midline at the level of the pectoral fins, advancing the needle until blood is seen entering the syringe.

[H-hyoid, TC-thyroid cartilage, CTM-cricithyroid muscle, ECA-external carotid artery, TG-thyroid gland, CBV-common brachiocephalic vein, S-sternum] Drawing by Barbara S. Irvine from Ridgway and Patton 1971.

Cetacean Euthanasia Data Record (Dec 2011)

Field Number: ____________________ **Species:** ___________________________ **Date:** ______________

Euthanizing Agency: ________________________ **Initial Report Time:** _______ **Time of Arrival:** ________

Officiating Veterinarian: ________________________ **Lead Responder:** ___________________________

Location Description (Circle): ND Marsh/mudflat Beach In surf Still/shallow water Bar/shoal Other _______________

Decision to perform euthanasia authorized by ____________________ @ NOAA and reason for euthanasia:

Pre- Euthanasia Data Time taken:

Heart rate	Palpate/auscultate ___ beats/1 min	**Attitude**	Alert Lethargic Non-responsive
Resp. rate; exudate	_____ breaths/ ____ min ; Y N	**Body position**	ND Upright Left side up Right side up
Resp. character	Strong Weak Regular Irregular Other	**Eyes** (Open / Closed)	Palpebral reflex- Y N Menace reflex- Y N
Body condition	ND Robust Normal Thin Emaciated	**Movement**	None Arch Fluke Swim Tremble Other
Body temp. (°F C)	Skin @ ________ Warm Cool ND	**Other:** *(describe in comments)* Vocalize Vomit Feces Urine Lesions	

Method of Euthanasia: **Pre-euthanasia Sedation** **Chemical Method** **Non-Chemical Method** *(describe)*

(Circle all that apply)

Agent 1:

Location of animal for administration of Agent 1 (Circle): Water Shore Vehicle Facility Other

Time of admin.		**Heart rate**	Palpate/auscultate ___ beats/1 min
Drug & conc.		**Resp. rate**	_____ breaths/ ____ min
Amount		**Resp. character**	Strong Weak Regular Irregular
Route	IM IV IP IH IC Other	**Resp. exudate**	Y N (Clear Foam Blood Other)
Injection site	R L D V Body area:	**Attitude**	Alert Lethargic Non-responsive
Response to agent	Y N	**Movement**	None Arch Fluke Swim Tremble Other
Time observed		**Eyes** (Open/Closed)	Palpebral reflex- Y N Menace reflex- Y N
Type of response	↑ ↓ Activity ↑ ↓ Responsiveness ↑ ↓ Respirations ↑ ↓ Heart rate ↑ ↓ Other		

Agent 2:

Location of animal for administration of Agent 2 (Circle): Water Shore Vehicle Facility Other

Time of admin.		**Heart rate**	Palpate/auscultate ___ beats/1 min
Drug & conc.		**Resp. rate**	_____ breaths/ ____ min
Amount		**Resp. character**	Strong Weak Regular Irregular
Route	IM IV IP IH IC Other	**Resp. exudate**	Y N (Clear Foam Blood Other)
Injection site	R L D V Body area:	**Attitude**	Alert Lethargic Non-responsive
Response to agent	Y N	**Movement**	None Arch Fluke Swim Tremble Other
Time observed		**Eyes** (Open/Closed)	Palpebral reflex- Y N Menace reflex- Y N
Type of response	↑ ↓ Activity ↑ ↓ Responsiveness ↑ ↓ Respirations ↑ ↓ Heart rate ↑ ↓ Other		

Additional sections for agents and comments are located on the back of this sheet.

Post Euthanasia Data:

Time of death		**Carcass necropsied**	Y N ND
How determined		**Carcass disposal method**	Bury Landfill Render Other

Please use comments section on back of form for additional notes on process and reaction(s)

Cetacean Euthanasia Data Record (Dec 2011)

Agent 3:

Location of animal for administration of Agent 3 (Circle): Water Shore Vehicle Facility Other

Time of admin.		**Heart rate**	Palpate/auscultate ___ beats/1 min
Drug & conc.		**Resp. rate**	_____ breaths/ ____ min
Amount		**Resp. character**	Strong Weak Regular Irregular
Route	IM IV IP IH IC Other	**Resp. exudate**	Y N (Clear Foam Blood Other)
Injection site	R L D V Body area:	**Attitude**	Alert Lethargic Non-responsive
Response to agent	Y N	**Movement**	None Arch Fluke Swim Tremble Other
Time observed		**Eyes** (Open/Closed)	Palpebral reflex- Y N Menace reflex- Y N
Type of response	↑ ↓ Activity ↑ ↓ Responsiveness ↑ ↓ Respirations ↑ ↓ Heart rate ↑ ↓ Other		

Agent4:

Location of animal for administration of Agent 4 (Circle): Water Shore Vehicle Facility Other

Time of admin.		**Heart rate**	Palpate/auscultate ___ beats/1 min
Drug & conc.		**Resp. rate**	_____ breaths/ ____ min
Amount		**Resp. character**	Strong Weak Regular Irregular
Route	IM IV IP IH IC Other	**Resp. exudate**	Y N (Clear Foam Blood Other)
Injection site	R L D V Body area:	**Attitude**	Alert Lethargic Non-responsive
Response to agent	Y N	**Movement**	None Arch Fluke Swim Tremble Other
Time observed		**Eyes** (Open/Closed)	Palpebral reflex- Y N Menace reflex- Y N
Type of response	↑ ↓ Activity ↑ ↓ Responsiveness ↑ ↓ Respirations ↑ ↓ Heart rate ↑ ↓ Other		

Use additional data sheets if needed for additional comments or if more than 4 agents were used

Comments: On a scale of 1-5, where 1=very poorly and 5=very well, how did this event go? 1 2 3 4 5

Please check any areas where problems occurred and elaborate below:

- ☐ Personnel/public safety
- ☐ Animal response/behavior
- ☐ Drug/tool availability
- ☐ Disposal
- ☐ Media/public response
- ☐ Other (please explain)

Please return form to:

Virginia Aquarium
Stranding Response
717 General Booth Blvd
Virginia Beach, VA 23451
vaqstranding@gmail.com
fax 757-437-4933

Instructions for Filling Out the Cetacean Euthanasia Record

Overview

This cetacean euthanasia record has been developed and distributed as part of a NOAA John H. Prescott Marine Mammal Rescue Assistance Grant Program project titled: "The Collaborative Development of Stranded Cetacean Euthanasia Recommendations". The goal of this project is to develop recommendations for stranding networks to facilitate the humane euthanasia of live stranded cetaceans when rehabilitation or release is not an option.

The collection and compilation of data from this euthanasia record is a critical part of the project. These data, as well as national historic cetacean euthanasia information, will be compiled and entered into a database that was specifically designed with this data record for our project. An expert advisory panel will examine and evaluate this information. Potential correlations regarding effects and outcomes as they relate to various methods of euthanasia (ex. techniques, drug types and doses, etc.) and the stranding situations encountered (ex. mass or single stranding, species specific reactions, logistics, environmental conditions, etc.) will be investigated. This information, as well as published information, will be used to assist in the determination of cetacean euthanasia recommendations. A final workshop report including the recommendations will be distributed to all contributors and participants. All participants will be acknowledged for their contribution to this project in any resulting products.

This cetacean euthanasia record will also facilitate standardizing data collection for cetacean euthanasia efforts.

Below we have provided instructions and explanations for entering information and data into the cetacean euthanasia record.

Identification Section

Field Number: Unique identifying number originally assigned to the animal by responder/responding organization. This number should coincide with the same information on the Marine Mammal Stranding Report – Level A Data.

Species: Genus and species and/or common name of the animal

Date: Date that the euthanasia was performed

Euthanizing Agency: Name of the lead Stranding Agreement holder or agency performing the euthanasia

Initial Report Time: Time of earliest known report of the animal to the responding agency

Time of Arrival: Time of arrival on scene by the responding agency

Be sure to use the ***Comments*** *section on the back of this record to expand on observations, descriptions, etc.*

***Decision to perform euthanasia authorized by __________ @ NOAA and reason for euthanasia*:** enter the name of the NOAA staff person who authorized the euthanasia and document the factor(s) that influenced the decision to euthanize the animal (e*xample: severe injury, species, size, age, logistics, lack of rehab facility, etc.)*

Pre-Euthanasia Data Section

Pre-euthanasia data time taken: record the time of initial examination

Heart rate: record the number of heart beats per one minute (e*xample: 30 beats/1 min*)

Circle the method used to determine heart rate:

Palpate: feel heart beat with hand

Auscultate: listen for internal heart sounds, generally with a stethoscope

Resp. rate; exudate (Respiratory exudate): record the number of breaths per 3 minutes (e*xample: 4 breaths/3 min*)

Circle **Y** if exudate is observed around blowhole and write description in comments section

(*ex. clear, foam, blood, other*); Circle **N** if none is observed

Resp. character (Respiratory character): the quality, depth, ease of breaths

Circle most accurate descriptions regarding strength and regularity:

Strong: "normal" forceful breaths

Weak: depressed or shallow respirations

Pre-Euthanasia Data Section *(continued)*

Body temp.: ***(˚F C)*** If actual body temperature taken, record degrees Fahrenheit or Celsius

Skin @ _______***:*** describe anatomical area used to determine skin temperature

Circle most accurate description of palpated skin temperature:

Warm: a slight, comfortable heat evident

Cool: lacking heat, "clammy"

ND: no data

Attitude: the animal's behavior/action (or lack thereof)

Circle most accurate description(s):

Alert: animal is alert and aware, follows movement with eyes

Lethargic: animal is sluggish or inactive, eyes closed, little reaction to touch/sound

Non-responsive: animal is not reactive to stimuli

Body position: the position of the animal's body upon initial discovery

Circle the most accurate description(s):

ND: no data

Upright: laying on ventrum

Left side up: laying on right side, right lateral recumbency

Right side up: laying on left side, left lateral recumbency

Method of Euthanasia:

Circle all methods that apply:

Pre-euthanasia Sedation: Circle if chemical agent(s) administered to the animal prior to euthanasia, including: tranquilizers (*ex. acepromazine maleate*), sedatives (*ex. xylazine*), immobilizers (*ex. ketamine*) and/or general anesthetics (ex. *tiletamine-zolazepam*)

Chemical Method: Circle if chemical agent(s) administered to euthanize the animal

Non-Chemical Method: Circle if euthanasia method included the use of non-chemical methods

Describe: describe method(s) used (*ex. ballistics – include type of firearm and ammunition used; exsanguination – include type of equipment used and anatomical location; etc.*)

Agent # Sections

We have provided "Agent #" sections for you to record the administration of up to four chemical agents (*#1 & #2 on first page, #3 & #4 on second*). If you did not administer any chemical agents please record N/A after Agent 1. If you used more than 4 agents, please use additional data sheets.
The Agent sections refer to the animal's response to the actual drug or agent (or the effect of the agent on the animal), not the physical reaction to the administration of the agent(s).

Location of animal during administration of Agent #:

Circle most appropriate answer:

Water: Circle if "Agent #" was administered while the animal was being maintained in water body

Shore: Circle if "Agent #" was administered while the animal was being maintained on a beach, marsh, mudflat, sandbar, etc.

Vehicle: Circle if "Agent #" was administered while the animal was being maintained in a vehicle

Facility: Circle if "Agent #" was administered while the animal was being maintained at a facility

Other: Circle if "Agent #" was administered while the animal was in an area not listed (e*xample: on boat, etc.*)

Time of admin. (administration): record the time that "Agent #" was administered

Drug and conc. (concentration): record the name of drug(s) administered and its concentration(s) (*example: number of mg/ml*)

Amount: Record the total amount of "Agent #" administered to the animal (*total mls or mgs*)

Route: Circle most appropriate answer(s):

IM (Intramuscular): into the muscle

IV (Intravenous): into the vein

IP (Intraperitoneal): into the peritoneal cavity

IH (Intrahepatic): into the liver

IC (Intracardiac): into the heart

Other: circle if route used is not listed and write in most appropriate answer

Injection site: Circle most appropriate answer(s) and then write the anatomical location(s) in which the agent was administered:

R: right

L: left

D: dorsal

V: ventral

Body area: anatomical location (*ex. fluke, epaxial muscle, etc.*)

(*ex. If administered in the dorsal side of right fluke =* ***R L D V*** ***body area:*** *fluke*)

Response to agent: Did animal exhibit any reaction to the drug administered

Circle most appropriate answer:

Y: if a response was observed after "Agent #" administered (*ex: animal becomes more sedate/agitated, heart rate or respirations decrease/increase, etc.*) and complete the next sections for "time of response" and "type of response"

N: if no response was observed. Skip "time of response" and "type of response" and move on to "heart rate", etc.

Time observed: Record time(s) response(s) was observed

Type of response: Describe response(s) to agent(s)

Circle most appropriate answer(s) (↑= increase, ↓= decrease)

Activity: amount of body movement (*ex. Twitching, fluking, etc*)

Responsiveness: animal's reaction to stimuli

Respirations: number and/or strength of respirations/minute

Heart Rate: number and/or strength of heartbeats/minute

Agent # Sections (*continued*)

Heart rate: same definitions and instructions as in the "pre-euthanasia" section
Resp. rate: same definitions and instructions as in the "pre-euthanasia" section
Resp. character (Respiratory character): same definitions and instructions as in the "pre-euthanasia" section
Resp. exudate (Respiratory exudate): same definitions and instructions as in the "pre-euthanasia" section
Attitude: same definitions and instructions as in the "pre-euthanasia" section
Movement: same definitions and instructions as in the "pre-euthanasia" section
Eyes: same definitions and instructions as in the "pre-euthanasia" section

Post Euthanasia Data Section

Time of death: record time that the animal was presumed deceased
How determined: record method(s) used to determine death of the animal (*example: no heart beat or respirations for specified amount of time, no palpebral reflex, rigor mortis present, etc.*)
Carcass necropsied: circle **Y** if the animal was necropsied, **N** if carcass was not necropsied or **ND** (no data) if unknown
Carcass disposal method: record method of disposal of the carcass
Bury: carcass placed in the ground and covered
Landfill: carcass taken to landfill
Render: carcass melted down
Other: sunk, towed, incinerated, chemically dissolved, etc.

Comments and Observations Section

Indicate, on a scale of 1 to 5 with 1 being worst case and 5 being best case, how the euthanasia proceeded. From the list provided, check any areas where you had concerns about the event and elaborate in the comment area below.

Also use this section to document details and/or explanations regarding the event that may not be captured in other documents.
Information may include:

- weather or other environmental conditions
- comments on the outcome of the event (*ex. did the event go well or poorly and reasons for this opinion*)
- details and/or a timeline of reactions to specific agents
- safety issues
- other personal observations or comments

Use extra pages if needed.