

Research in

ISSN : P-2409-0603, E-2409-9325

AGRICULTURE, LIVESTOCK and FISHERIES

An Open Access Peer-Reviewed International Journal

Article Code: 0376/2022/RALF Article Type: Research Article Res. Agric. Livest. Fish. Vol. 9, No. 2, August 2022: 213-219.

A CASE REPORT ON EUTHANASIA OF A RESCUED HORSE USING XYLAZINE HYDROCHLORIDE AND THIOPENTAL SODIUM

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ARTICLE INFO	ABSTRACT
Received 4 August, 2022	
Revised 22 August, 2022	Veterinary professionals are still looking for a suitable method of euthanasia that causes an easy death of animals with minimum pain, fear, and suffering. The present study describes a method of euthanasia in rescued horses using xylazine hydrochloride and thiopental sodium. The study was conducted at Veterinary Teaching Hospital, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Bangladesh. Xylazine hydrochloride (0.1 mg/kg; 20mg/ml) and thiopental sodium (10 mg/kg; 25% solution) was administered intravenously to the horse.
Accepted 24 August, 2022	
Online 31 August, 2022	
Key words:	The horse rapidly became unconscious and died within 4 minutes without any signs of excitement. This method can be practiced as an easy and convenient method of euthanasia of
Euthanasia Horse Thiopental sodium Xylazine hydrochloride Unconsciousness	horses.

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To cite this article: Rahman M. A, M. T. Rahman, M. Saha, R. S. Sadi, T. R. Labib, M. F. Hasan, M. G. Haider, M. M. Rahman and M. S. Alam, 2022. A case report on euthanasia of a rescued horse using xylazine hydrochloride and thiopental sodium. Res. Agric. Livest. Fish., 9 (2): 213-219.



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INTRODUCTION

Questions regarding the welfare of animals are becoming increasingly obvious as people and animals have a closer relationship due to recent social, economic and ecological motives. In veterinary, the animal well-being is extremely valued, and the methods that minimize animal suffering are routine (Amaral et al., 2011). Veterinarians play an important role in the tripartite association between animals, animal owners, and their environment. Veterinarians are obligated to improve animal welfare. These include disease prevention, treatment, good management, proper nutrition, humane handling, humane transportation, and humane killing when necessary. A veterinarian is not obligated to kill an animal with good health unless they are validated by the law. In certain instances, veterinarians do have the honor of being able to ease an animal's suffering by performing euthanasia (Royal College of Veterinary Surgeons, 2021). When conditions of disease or injury occur that reduce the quality of life or cause pain and suffering that cannot be effectively relieved by medical procedures, euthanasia is indicated (Shearer & Nicoletti, 2013). Furthermore, there are many reasons for a horse to be euthanized, including disease, an accident, a change in the owner's circumstances, or their quality of life has deteriorated due to old age or a pre-existing condition (The British Horse Society, 2020). Euthanasia can be performed by chemical or physical means (Sharp, 2016). Chemical means include chloral hydrate, potassium chloride, magnesium sulfate, and intrathecal lidocaine which are used exclusively for horse euthanasia (Kranenburg, 2020). Cinchocaine and quinalbarbitone, as well as the captive bolt method, are also used (Knottenbelt, 2006; Kranenburg, 2020). Sedation prior to euthanasia may be considered as a method of reducing possible anxiety, and distress, and considered as the most humane method (Close et al., 1996). Among many ways, euthanasia by lethal injection is one of the most widely used methods. The injection contains an overdose of anesthetic chemicals, causing the horse to progressively collapse, lose consciousness guickly, and suffer cardiovascular arrest (The British Horse Society, 2020). Sodium pentobarbitone alone may cause unacceptably high levels of excitement before unconsciousness (Reilly, 2001). In the case, xylazine premedication may be required (Reilly, 2001). Thiopental sodium is an anesthetic agent given intravenously for anesthesia (Royal Dutch Society for the Advancement of Pharmacy, 1994). Chloral hydrate is conditionally suitable for large animal euthanasia when administer intravenously and after sedation to reduce the aforementioned unpleasant side effects (AVMA, 2007). Potassium chloride or magnesium sulfate should only be used under general anesthesia. Many countries do not allow potassium chloride and magnesium sulfate to be used for animal euthanasia because they are not controlled substances (Kranenburg, 2020). Intrathecal administration of 2% lidocaine hydrochloride has been shown to cause euthanasia in horses within 30 seconds (Kranenburg, 2020). Cinchocaine and guinalbarbitone can also be used in some cases (Knottenbelt, 2006) and physical methods in horses, while, a well-placed gunshot can produce rapid insensibility and death (AVMA, 2007).

In the present case, we used both xylazine hydrochloride and thiopental sodium for euthanasia. Xylazine is a potent 2adrenergic agonist whose effects are mediated through central 2-receptor stimulation. The release of dopamine and norepinephrine in the central nervous system (CNS) is inhibited by 2-adrenergic stimulation, resulting in drowsiness, reduced perception of painful stimuli, and muscular relaxation (Saeed Kolahian, 2014).Furthermore, its actions may involve cholinergic, serotonergic, dopaminergic, 1-adrenergic, histaminergic, or opiate pathways (Hoffmann, Meister, Golle, and Zschiesche, 2001). Barbiturates depress the central nervous system (CNS) in descending order, starting with the cerebral cortex and progressing to general anaesthesia. An overdose, leads to apnoea owing to respiratory center depression, which is followed by cardiac arrest (Kranenburg, 2020). Before administering barbiturates, horse should be sedated with xylazine or other general anesthetics to avoid the risks to the individuals (Kranenburg, 2020). These are the most often used and accepted euthanizing agents for most animals (Close et al., 1996).

Ethical approval

The present research and experimental protocols were approved and performed according to the guidelines for the care and use of animals as established by Animal Welfare and Experimentation Ethics Committee, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur-1706, Bangladesh.

Case description

A rescued horse weighing 200 kg was found on the side of a road near Itakhola in Gazipur, Bangladesh, and was brought to the Veterinary Teaching Hospital of Bangabandhu Sheikh Mujibur Rahman Agricultural University for treatment. There were several wounds on the body filled with pus. It was cachectic. In close observation, the right hind limb was found paralyzed that was unable to get up and this was the cause of the numbness (Figure 1 and Figure 2). The horse tried to eat green grass indicating that it had an appetite. Furthermore, mucopurulent discharge from the vulva and vagina was found. The mare could not defecate on its own but the urination was normal. The wounds on the body indicated decubitus ulcers due to prolonged recumbency. The horse was cachectic because it did not get proper feed for a long time and there was a chronic nutritional deficiency. There was no sign of fracture or dislocation on the right hind limb. Though there was a lack of proper history, we diagnosed the case as traumatic nerve injury leading to paralysis. Vaginal mucopurulent discharge indicated chronic vulvovaginitis. The horse was given an anti-inflammatory drug, ketoprofen (Inj. Keto-A vet®, Acme Laboratories Ltd., Bangladesh) @ 4mg/kg body weight IM once daily for 5 days. An antihistaminic drug pheniramine maleate (Inj. Asta vet®, Acme Laboratories Ltd., Bangladesh) was given @ 11.375 mg/kg body weight IM once daily for 5 days. Oxytetracycline (Inj. Otetra vet-50®, Square Pharmaceuticals Ltd., Bangladesh) was selected as the antibiotic and was given @ 10 mg/kg body weight IM once daily for 5 days. Calcium gluconate (Cal-D-Mag vet®, Renata Ltd., Bangladesh) was also given @ 260 mg/kg body weight IV once daily for 2 days along with 2000 ml 5% dextrose saline (Libbot®, Libra Infusions Ltd., Bangladesh) intravenously. A 10 ml of a multivitamin preparation (Multivit vet®, Square Pharmaceuticals Ltd.) was injected intramuscularly (IM) once daily for 5 days. The mare received green grass and clean water ad-libitum. But it had no affinity towards concentrate feed. The side of recumbency was changed from time to time at a regular interval. The wounds were dressed using povidone-iodine solution (Sol. Betadine®, Mundipharma (Bangladesh Pvt. Ltd.) and local antibiotic Bacitracin (Powder Nebanol®, Square Pharmaceuticals Ltd.) was applied twice daily for 15 days. There was no sign of improvement after the treatment. The mare was really old and it was suffering from severe pain. The affected limb was irreversibly paralyzed. The rescuer was emotional and can't tolerate the suffering of the animal. The rescuer requested us to put an end to its pain. So, we decided to euthanize the mare. For euthanasia, we used 20 mg of xylazine HCI (0.1 mg/kg bodyweight, Xylazine®, Mesko Pharmaceuticals) given intravenously (IV) at the jugular vein. Immediately 25% solution of thiopental sodium (10 mg/kg bodyweight, Inj. TPS® 500mg, Square Pharmaceuticals Ltd., Bangladesh, IV) was injected rapidly at the jugular vein.



Figure 1. Recumbent mare

RESULTS

Palpebral reflex, lacrimation, nystagmus were absent within 2-5 seconds of injecting thiopental sodium. At the same time, two deep breaths were taken by the horse followed by complete cessation of respiration within 15 seconds. The heart beat was decreased in the first minute and then the beat was increased during the second minute. The heart beat again went down and slowly the beat was completely lost in 3 minutes and 40 seconds. The muscle of limb, neck and

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back were relaxed. There were no signs of salivation and vomition. The animal was declared clinically dead within 4 minutes. During this time the animal showed no hypersensitivity or anxiety or hyperesthesia. Finally, the horse was buried by maintaining all hygienic protocols. We have used trench burial method. The best cross-sectional shapes for the burial site are trapezoidal and rectangular. We used rectangular shape. At first the length of the trench was measured from the cross-sectional area of the trench geometry. The ratio of trench volume to carcass volume for burying carcasses was 2:1. Hydrated lime was used during burial to reduce the potential for spread of disease or odor control. Then the carcasses placed in the trench. The carcass was punctured to prevent gases from accumulating and exploding. After that 2 feet thick soil layer was used for covering the carcasses.



Figure 2. Right hind limb paralyzed mare

Discussion

Since many horses do not pass away naturally, owners must consider euthanasia in the event of an accident or a sickness that is incurable. No horse should have to go through unnecessary pain or distress and euthanasia can prevent a horse from suffering. Having to put a horse to die, is never an easy decision to make and it can also be very upsetting for an owner. In making that decision, the role of the veterinarian is to give them the best information regarding the horse's condition necessitating euthanasia and also specify the method and chemicals needed for this case. Likewise, a case that came to the veterinary teaching hospital was diagnosed with chronic vulvovaginitis and the affected animal had swelling of the vagina and vulva and mucopurulent vaginal discharge by their vulva. Several previous research findings confirm that vulvitis and vaginitis in horses frequently occur as a result of difficult labor, prolonged contamination of the reproductive system as a result of poor conformation, sexually transmitted infections, or mating (Gilbert, 2019). A previous study shows that traumatic nerve injury leads to paralysis due to the damage to the peripheral nerves and nerve injury can also be caused by irritants medication injected into or near the peripheral nerves (Thomas, 2019). Injury to the femoral, sciatic, peroneal, or tibial nerve in the leg and to the nerve roots in the lower back or tailbone is frequently the cause of hind leg paralysis and the most prevalent cause is traumatic injury for such types of paralysis (Schubert, 2020).

In current case; the horse showed very little progress and the complications were increasing as time going on regardless of the medication and care. The severity of vulvovaginitis was improved a little but the condition of the paralyzed limb and hindquarter was declining with time such as starting to get thinner and thinner. As mentioned earlier, we decided to perform euthanasia to free the horse from suffering. Different chemical methods are employed for this purpose but all those method have some like drug residues, longer time for performing euthanasia. We introduce an improved chemical method which should be compared with conventional chemical euthanasia for equine species. Lethal injection with a barbiturate, typically pentobarbital is the method most commonly employed by veterinarians in the United States (Lisa Nesson, 2019). This method does result in drug residues that require the careful disposal of carcass to prevent other scavenger animals from having access to the dead body or entering any food chain (Marchitelli, 2019).

Intravenous injection of a potassium chloride solution at a dose of 75-150 mg/kg is another method of euthanasia but takes longer time. Chloral hydrate is another choice of drug for euthanasia, but when applied it slowly depresses the cerebrum and restraint may be difficult in the case of some animal species (AVMA, 2013). Death is caused by hypoxemia, which is caused by increasing respiratory depression and distress may occur (AVMA, 2007). A saturated solution of Magnesium sulfate of 360g in 1L of distilled water (2ml/kg IV) will induce cardiac arrest and death. This method is not approved for euthanasia of animals in many countries (Kranenburg, 2020). Cinchocaine and quinalbarbitone are available as a single preparation for use with or without prior sedation but xylazine is not recommended to use as violent reactions can occur which then negatively affect the procedure (Knottenbelt, 2006). Thiopental Sodium is used intravenously for the euthanasia at a dose ranges from 5.5 to 22 mg/kg for horse are recommended. In both the Netherlands and Belgium, where active euthanasia is allowed by law, the standard protocol recommends thiopental sodium as the ideal agent to induce coma (Royal Dutch Society, 1994). In current case of euthanasia by using thiopental sodium in combination with xylazine, death occurs quickly and the reflexes disappeared within a few seconds. In the present report, use of xylazine hydrochloride (0.1mg/kg body weight, IV) and 25% solution of thiopental sodium (10 mg/kg bodyweight IV) were used. Similar article showed euthanasia of horse combination with xylazine and pentobarbitone (Derek knottenbelt, 1995).

To achieve optimal bioavailability, the muscle relaxant xylazine was also given intravenously. Cardiac arrest, leg movements, and involuntary jerking, as well as muscle tremors were recorded and reported. Therefore, it is revealed that xylazine-induced sedation and analgesia in animals could reduce the dose of barbiturate required for the induction of anesthesia (Green, 1999). Disposal by trench burial involves excavating a trough into the earth, placing carcasses in the trench, and covering with the excavated material (backfill). Relatively little expertise is required to perform trench burial, and the required equipment is commonly used for other purposes. The primary resources required for trench burial include excavation equipment and a source of cover material. Cover material is often obtained from the excavation process itself and reused as backfill. Important characteristics in determining the suitability of a site for burial include soil properties; slope or topography; hydrological properties; proximity to water bodies, wells, public areas, roadways, dwellings, residences, municipalities, and property lines; accessibility; and the subsequent intended use of the site. Trench burial is cited as a relatively economical option for carcass disposal as compared to other available methods. It is also reported to be convenient, logistically simple, and relatively quick, especially for daily mortalities, as the equipment necessary is generally widely available and the technique is relatively straightforward. If performed on-farm or on-site, it eliminates the need for transportation of potentially infectious material. The technique is perhaps more discrete than other methods (e.g., open burning), especially when performed on-site (on-farm). Disadvantages of trench burial include the potential for detrimental environmental effects, specifically water quality issues, as well as the risk of disease agents persisting in the environment (e.g., anthrax, transmissible spongiform encephalopathy [TSE] agents, etc. The residue within a burial site has been shown to persist for many years, even decades, ultimate elimination of the carcass material represents a longterm process, and there is a considerable lack of knowledge regarding potential long-term impacts. Trench burial may be limited by regulatory constraints or exclusions, a lack of sites with suitable geological and/or hydrological properties, and the fact that burial may be prohibitively difficult when the ground is wet or frozen. In some cases, the presence of an animal carcass burial site may negatively impact land value or options for future use (The National Agricultural Biosecurity Center Consortium Carcass Disposal Working Group, 2004).

CONCLUSION

A rapid intravenous injection of xylazine hydrochloride (0.1 mg/kg; 20mg/ml) and thiopental sodium (10 mg/kg; 25% solution) resulted in rapid and smooth unconsciousness (death confirmed within 4 minutes) with no reactions or excitement. In our opinion, this method is the most humane and safest technique to sedate and euthanize horses.

ACKNOWLEDGEMENT

The author thanked the rescuers for rescuing the horse. The work was supported by BSMRAU-RMC funding, FY 2021-2023-31 to M. Shah Alam.

AUTHORS CONTRIBUTIONS

Experiments were conceived and designed by MA Rahman, MT Rahman, and M Saha. The experiment was done by S Saadi, TR Labib, and MF Hasan. MG Haider provided facilities for conducting experiments at the Veterinary Teaching Hospital. The manuscript was written by MA Rahman, and M Saha, and critically revised by MM Rahman and MS Alam. The study was overall supervised by MS Alam.

AUTHORS' DECLARATION OF INTERESTS

No conflicts of interest have been declared.

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