Presented at the DLSU Research Congress 2017
De La Salle University, Manila, Philippines
June 20 to 22, 2017

Using Carbon Dioxide Resulting from the Chemical Reaction of Sodium bicarbonate and Vinegar to Euthanise Rodents

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Abstract **– While rodent specimens are invaluable and essential in biological research, the ethical euthanasia of these animal specimens using Carbon dioxide is cost-prohibitive, especially for smaller institutions or less-funded researchers. Amateur rodent and reptile breeders use Carbon dioxide that arises from the chemical reaction of Sodium bicarbonate and acetic acid to euthanize their animals, however this methodology has not been standardized for use in research. This initial study reported the use of this method in euthanizing a wild rodent, and recommends methods by which this methodology can become further tested.**

Keywords: rodents; euthanasia; lab animals

1. INTRODUCTION

The use of rodents as laboratory specimens has long been the standard practice of many research laboratories and institutes. However, the methods by which these rodents are studied have changed across the years, with a focus on ethical treatment of these research animals. While there are many approved methods of sacrificing organisms for research, majority of these, such as the injection of barbiturates, can only be properly administered by a licensed veterinarian. A researcher without the training and licensing necessary to employ other methods is recommended to sacrifice laboratory rodents by euthanizing them with Carbon dioxide.

Euthanizing rodent specimens with Carbon dioxide can be done in two ways: firstly, by gradually increasing the amount of Carbon dioxide in the container of the specimen to either 10 or 30% within one minute, and secondly, by performing a two-step immersion method where the specimen is first sedated and then placed in a container of 100% Carbon dioxide. By increasing the concentration of Carbon dioxide inhaled by a lab animal in an enclosed space, a researcher can cause respiratory acidosis, leading to failed ventilation and depressed nervous system in the rodent specimen, which will eventually lead to coma, then death.

While there are many different places where a researcher or institution can purchase a Carbon dioxide canister and the prerequisite materials necessary for its use, such as the regulator and refills of the canister, it is financially prohibitive to do so. Larger institutions such as universities and research laboratories are more capable of purchasing and maintaining these containers and their accoutrements, but smaller learning institutions or those that are located less centrally may find it more difficult, shrinking the scope of work that their students and researchers can engage in.

Amateur rodent and reptile breeders have been able to develop their own method of euthanizing small rodents using baking soda, Sodium bicarbonate, and vinegar, diluted food grade acetic acid. When both are mixed together, a chemical reaction results in the release of Carbon dioxide gas, which can, in varying concentrations, either sedate or euthanize a rodent. Similar to the plight of smaller institutions, amateur rodent and reptile breeders have found a need to develop a cost-effective method of euthanizing rodents, whether mice or rats, that does not require them to

commit to buying and maintaining a Carbon dioxide canister. Baking soda and vinegar are readily available in grocery stores in varying amounts that can be chosen depending on the number of rodent specimens to be euthanized. Since the materials are easily accessible and researchers do not need to commit to purchasing amounts beyond what is necessary for their purposes, this methodology seems to be well-suited for small research purposes.

30% of the volume of the container must contain Carbon dioxide within 1 minute to euthanise rodents [1]. The interaction of 1 g of baking soda and 15 mL of vinegar results in 291 mL of Carbon dioxide and is sufficient to kill purebred mice, however wild rodents are much larger and heartier, and the interaction of vinegar and baking soda is also imperfect as the percentage of acetic acid in vinegar varies, so modifications were made and the measurements below have been found to work on a rodent with a body 19 cm long, not counting the tail. and 10.15 cm in circumference, around the belly, and less than 500 grams in weight.

2. METHODOLOGY

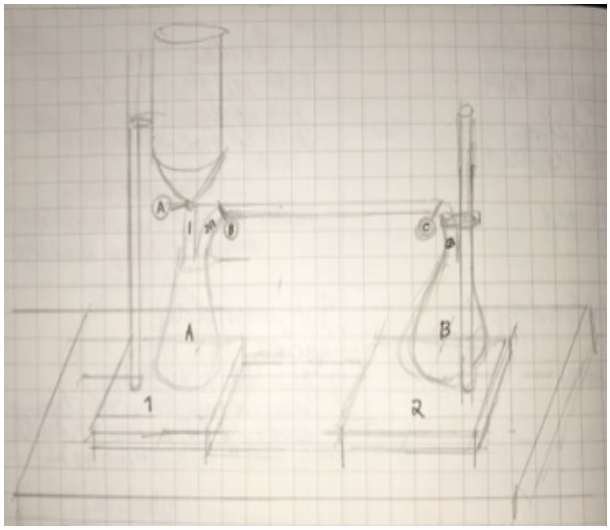

Fig. 1. Idealized ad hoc euthanasia chamber diagram.

13.5 tablespoons, roughly 191.76 grams, of baking soda was put into a 10-L volume transparent bag (Fig 1A). A 3-in piece of rubber tube (Fig. 1-1) and a 6-in piece of rubber tubing (Fig. 1-2A) was affixed to the bag. Tube 1 was connected to a bladder containing 1L of vinegar. The rodent specimen was then put in the second 10-L volume transparent bag (Fig. 1B). The vinegar was then allowed to pour from the bladder down tube 1 and into Bag A. The researcher had to agitate the resulting mixture until the bag filled with Carbon dioxide, then they compressed the bag until the gas travelled through tube 2A to Bag B. The researcher continued to agitate the mixture until it suddenly cooled, indicating the completion of the reaction.

3. RESULTS

Fig. 2. Rodent specimen euthanized with the method.

The rodent ceased to respire within 3 minutes since the gas filled Bag B, and was left in the gas-filled bag for up to 5 minutes after it ceased respiring, whereupon it was tested for response to external stimuli. The rodent failed to respond any external stimuli and did not present any challenges to physical manipulation.

4. CONCLUSION

While the rodent specimen was euthanized using the methodology delineated above, the findings themselves cannot be considered conclusive as the results from a single sample cannot be, in good conscience, extrapolated over all rodent specimens, and the rodent specimen in question was not checked in advance of the experiment itself, meaning that it could have possessed pre-existing conditions that would make it predisposed to be more sensitive to the methodology.

The researcher has several recommendations on that score: 1) to increase the number of rodent samples used, 2) to control the type and size of rodents to determine a proper range of size and weight that the methodology is effective upon, 3) to have the rodents checked by a veterinarian before experimentation to ensure that they do not possess pre-existing conditions that may affect the outcome of the study, 4) to have the rodents examined and dissected by a veterinarian after being euthanized so as to ensure that the methodology does not inflict additional physical harm on the rodents that could affect experimentation, e.g. internal bleeding.

If the recommendations can be applied and the findings are consistent that this methodology can ethically euthanize rats without inflicting additional damage to their internal organs, this could be a less financially prohibitive method of euthanizing rodent specimens that most researchers and institutions can do with little expense and effort, enabling more people to perform ethical euthanasia at a lower price point.

5. ACKNOWLEDGMENTS

P.P.S. thanks DOST-SEI ASTHRDP-NSC, Dr. Divina Amalin, Dr. Florencia Claveria, Dr. Mary Jane Flores, and Dr. Rodel Vitor.

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