Date of Approval: May 20, 2005

FREEDOM OF INFORMATION SUMMARY

NADA 141-245

TRIBUTAME Euthanasia Solution

embutramide/chloroquine phosphate/lidocaine

TRIBUTAME Euthanasia Solution is indicated for euthanasia in dogs only.

Sponsored by:
Phoenix Scientific, Inc.
3915 South 48th Street Ter.
St. Joseph, MO 64503

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1. GENERAL INFORMATION:

a.	File Number:	NADA 141-245
b.	Sponsor:	Phoenix Scientific, Inc. 3915 South 48th Street Ter. St. Joseph, MO 64503 Drug Labeler Code: 059130
c.	Established Name:	embutramide/chloroquine phosphate/lidocaine
d.	Proprietary Name:	TRIBUTAME* Euthanasia Solution
e.	Dosage Form:	Injectable Solution
f.	How Supplied:	100 mL bottles
g.	How Dispensed:	Rx
h.	Amount of Active Ingredients:	Each mL contains 135 mg Embutramide, 45 mg Chloroquine Phosphate, USP, 1.9 mg Lidocaine, USP.
i.	Route of Administration:	Intravenous Injection
j.	Species/Class:	Dogs
k.	Recommended Dosage:	Inject **1 mL** of TRIBUTAME Euthanasia Solution for each **5 pounds** body weight (0.45 mL/kg). Administer the calculated dose as a single intravenous bolus injection within a 10- to 15-second period. Do not overdose. **Do not use in cats.**
l.	Pharmacological Category:	Euthanasia Solution
m.	Indications:	TRIBUTAME Euthanasia Solution is indicated for euthanasia in dogs only.

* During this product's development, the proposed trade name was EMBUTANE. This trade name was changed to TRIBUTAME late in the development process. Therefore, most study titles refer to the product as EMBUTANE. However, the reader should be aware that the two trade names refer to the identical product and the word EMBUTANE has been changed to TRIBUTAME wherever possible.

2. EFFECTIVENESS:

a. Dosage Characterization:

TRIBUTAME Euthanasia Solution is a true solution of three active components, embutramide, chloroquine phosphate and lidocaine, in water and alcohol. The purpose of the ethyl alcohol is to increase the solubility of the three active components.

Embutramide is a general anesthetic that has never been used clinically in animals for that purpose. During initial testing, it was found to have a very narrow margin of safety, providing narcosis at 50 mg/kg but being lethal at 75 mg/kg[1]. Testing of various doses of embutramide resulted in rapid onset of anesthesia, but also resulted in apnea and short periods of ventricular asystole followed by severe ventricular dysrhythmias[2]. Embutramide dissolved in alcohol was found to be lethal in dogs at intravenous (IV) doses exceeding 61 mg/kg. Additionally, embutramide at these doses also caused pain at the injection site.

Chloroquine phosphate is an aminoquinoline used in the control and treatment of malaria in humans. It has undesirable cardiovascular effects such as arterial hypotension, depression of myocardial function and disturbances in ventricular conduction and rhythm[3]. In dogs, chloroquine phosphate is a myocardial depressant. Hypotension induced by chloroquine phosphate is attributable both to its myocardial depressant action as well as reduction of peripheral vascular resistance[4]. Studies indicated that the time to death was shortened when chloroquine phosphate was combined with embutramide, which further supports the cardiotoxic effects of chloroquine phosphate[2].

Lidocaine is used as a local anesthetic and is used in this formulation to counteract the burning sensation that can accompany the IV administration of the alcohol. Lidocaine is also used in human medicine as a cardiac depressant, anti-arrhythmia agent. At elevated dosages, lidocaine can cause central nervous depression, which leads to respiratory arrest[5]. Therefore, the local anesthetic effects and the cardiac and central nervous system depression are the basis for including lidocaine in this formulation.

In summary, any of the three active components in the formula are capable of causing death in the dog if given in large enough doses. However, these drugs individually can be slow to produce the desired result and can cause or intensify undesirable side effects. The combination of a general anesthetic, a cardiotoxin and a local anesthetic each contribute a different mechanism of action to work concurrently to produce a quick and painless death while reducing the occurrence of undesirable side effects.

Dose Titration Evaluation of EMBUTANE as an Euthanasia Solution in Dogs.
Study Number: PSI-6833-01-01-94-SP

Investigator: Donald Sawyer, D.V.M, Ph.D.
Michigan State University
East Lansing, MI

Study Purpose: The purpose of this study was to determine the optimum dose of TRIBUTAME Euthanasia Solution for euthanasia in dogs.

Test Animals: A total of 104 dogs were divided based on their health status. The dogs weighed between 1.7 to 40.8 kilograms and ranged in age between 4 months and 10 years. Sixty-four dogs were classed as healthy and 40 were classed as sick. Animals were judged to be healthy by observation and physical examination with no obvious evidence of systemic disease. Animals in poor health were determined to be sick by observation, physical examination, or advanced aged. Signs of cough, ocular discharge, loss of weight, poor appetite, and lethargy were criteria used to place animals in the sick category.

Control: Each animal acted as its own control.

Treatment Groups: The dogs were randomly assigned to one of four treatment groups (see Table 1). Each of the four treatment groups for the healthy dogs contained 8 male and 8 female dogs. Each of the four treatment groups for the sick dogs contained 5 male and 5 female dogs. All of the treatment groups received TRIBUTAME Euthanasia Solution, but at differing doses.

Table 1: Treatment Groups

Treatment Group	**Dose for Healthy Dog Group[1]**	**Dose for Sick Dog Group[1]**
A	0.45 mL/kg	0.15 mL/kg
B	0.25 mL/kg	0.25 mL/kg
C	0.15 mL/kg	0.35 mL/kg
D	0.35 mL/kg	0.45 mL/kg

[1] If death did not occur within 5 minutes, an additional dose of 0.2 mL/kg was injected via the intracardiac route of administration.

Masking: A separate individual, who was not involved in any observations or data acquisition, prepared the dose for administration. An individual, who was masked to the dose, made all of the observations and recorded the timed events.

Dosage Form: Solution for injection. The formulation used in this study was not final formulation. The final formulation is the same formulation that was used in this study, except that the bitter agent, denatonium benzoate, has been added to this formulation so that the solution is less likely to be taken orally by accident or without knowledge.

Dosage: 0.15, 0.25, 0.35, or 0.45 mL/kg

Route of Administration: Intravenous injection.

Variables Measured:

1. Reaction to Injection Score:
   The observer subjectively assessed the clinical emergence of pain or discomfort at the injection site and assigned a score, based on the following:
   0 = No reaction
   1 = Look at leg
   2 = Lift/pull leg
   3 = Cry, struggle

2. Timed Events:
   The observer determined the study clock time (to the second) of start of initial injection, end of initial injection, (and if needed - start of additional injection, end of additional injection), collapse, apnea, loss of palpebral reflex, loss of visible heartbeat, and death. Time of death was defined as no visible, audible or palpable heartbeat.

3. Adverse Reactions:
   The observer continually observed the dog from the start of injection until time of death for the presence of the following adverse reactions:
   Agonal Breathing
   Opisthotonus
   Vocalization
   Other

Results:

1. Reaction to Injection Score:
   In both the healthy and sick dogs, all dogs in the three lowest dose groups (0.15, 0.25 and 0.35 mL/kg) experienced no reaction to the injection (Score=0). Two healthy and one sick dog in the 0.45 mL/kg dose group lifted or pulled the leg (Score=2) and one healthy and two sick dogs in the 0.45 mL/kg dose group cried or struggled (Score=3).

2. Timed Events:
   In the 0.15 mL/kg dose group, only one healthy and one sick dog died within five minutes. In the 0.25 mL/kg dose group, 5/16 healthy dogs and 4/10 sick dogs died within five minutes. The remaining dogs all required additional administration of TRIBUTAME Euthanasia Solution. The results for the timed events for the two higher dose groups are shown in Table 2 below.

Table 2: Arithmetic Mean (range) in Seconds of Timed Event Results in the Two Higher Dose Groups (0.35 mL/kg and 0.45 mL/kg)

Timed Event[1]	0.35 mL/kg		0.45 mL/kg Dose Group	
	Healthy	Sick	Healthy	Sick
Collapse	17 (11-50)	13 (10-16)	13 (11-23)	14 (8-28)
Apnea	34 (15-60)	22 (15-47)	26 (15-70)	26 (9-70)
Loss of Palpebral Reflex	64 (38-105)	64 (51-86)	65 (30-80)	65 (51-79)
Loss of Palpable Heart Beat	91 (35-181)	78 (20-187)	89 (25-220)	64 (10-135)
Death	188 (61-246)[2]	186 (110-286)	158 (90-225)	191(143-252)[2]

[1] All times were measured from the start of the injection until the event.
[2] This result excludes one dog whose time of death was greater than five minutes and required additional administration of TRIBUTAME Euthanasia Solution.

3. Adverse Reactions:
Table 3 shows the incidence of adverse reactions between the treatment groups.

Table 3: Number of Dogs (%) with Each Adverse Reaction

Adverse Reaction	0.15 mL/kg		0.25 mL/kg		0.35 mL/kg		0.45 mL/kg	
	Healthy	Sick	Healthy	Sick	Healthy	Sick	Healthy	Sick
Agonal Breathing	0 (0%)	0 (0%)	1 (6.3%)	3 (30%)	8 (50%)	3 (30%)	11 (68.8%)	6 (60%)
Vocalization	0 (0%)	0 (0%)	0 (0%)	2 (20%)	1 (6.3%)	0 (0%)	5 (31.3%)	2 (20%)
Opisthotonus	0 (0%)	0 (0%)	0 (0%)	1 (10%)	3 (18.8%)	2 (20%)	1 (6.3%)	1 (10%)
Resumption of Breathing	3 (18.8%)	1 (10%)	6 (37.5%)	1 (10%)	1 (6.3%)	0 (0%)	0 (0%)	0 (0%)
Rearing on Hind Legs	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (6.3%)	0 (0%)
Tremors/Movement	1 (6.3%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (6.3%)	0 (0%)
Urination	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (6.3%)	0 (0%)

Conclusions: Because the two lower doses (0.15 and 0.25 mL/kg) did not produce death within five minutes in the majority of animals, the minimum acceptable dosage for euthanasia of both healthy and sick dogs was 0.35 mL/kg body weight. Because of an increased incidence of opisthotonus and resumption of breathing in the 0.35 mL/kg dose group, the 0.45 mL/kg dose was selected as the intended labeled dose.

b. Substantial Evidence:

Dose Confirmation/Physiology Study to Define the Cessation of Critical Vital Functions in the Dog following the Administration of EMBUTANE (embutramide solution) at Two Intravenous Dosages.
Study number PSI-0736-99C-004

Investigator: William Muir, III D.V.M., Ph.D.
Ohio State University
Columbus, Ohio

Study Purpose: The purpose of the study was to determine the time to cessation of vital physiological signs in dogs following administration of one of two doses of TRIBUTAME Euthanasia Solution using either a single bolus or split bolus injection technique.

Test Animals: Thirty-two dogs, between 5.6 and 7.6 kg, were randomly assigned to one of four gender equalized groups.

Control: Each animal acted as its own control.

Treatment Groups: The dogs were randomly assigned to one of four groups, with 8 dogs (4 male and 4 female) in each group (see Table 4). All of the treatment groups received TRIBUTAME Euthanasia Solution, but at differing doses and bolus administrations.

Table 4: Treatment Groups

Treatment Group	**Type of Intravenous Bolus**	**Dose (mL/kg)[1]**
1	Single	0.35 mL/kg over 5 seconds
2		0.45 mL/kg over 5 seconds
3	Split	0.35 mL/kg total with 0.15 mL/kg initially followed by 0.2 mL/kg within 5 seconds of collapse
4		0.45 mL/kg total with 0.15 mL/kg initially followed by 0.3 mL/kg within 5 seconds of collapse

[1] If additional drug was needed to complete euthanasia, one half of the original dose was given at 5-minute intervals until euthanasia was complete.

Masking: A separate individual, who was not involved in any observations or data acquisition, prepared the dose for administration. An individual, who was masked to the dose, made all of the observations and recorded the timed events.

Dosage Form: Solution for injection. The formulation used in this study was not final formulation. The final formulation is the same formulation that was used in this study, except that the bitter agent, denatonium benzoate, has been added to this formulation so that the solution is less likely to be taken orally by accident or without knowledge.

Dosage: 0.35 mL/kg or 0.45 mL/kg as a single or split bolus intravenous injection.

Route of Administration: Intravenous injection.

Variables Measured:
1. Quality of Induction Score:

The masked observer subjectively assessed the clinical attainment of unconsciousness and assigned a score, based on the following definitions:
0 = Excellent: Quietly collapses without any sign of excitement, struggling or distress.
1 = Good: Quietly collapses with only minor, fleeting signs of excitement, struggling or distress.
2 = Poor: Exhibits a brief period of excitement before collapsing with noticeable signs of struggling or distress.
3 = Unacceptable: Exhibits marked excitement during induction with vigorous struggling or marked signs of distress.

2. Timed Events:
   The masked observer determined the study clock time (to the minute) of start of initial injection, end of initial injection, (and if needed - start of additional injection, end of additional injection), collapse, apnea, last audible heartbeat, and death, which was defined as no audible heartbeat and cessation of electroencephalograph (EEG) activity.

3. Adverse Reactions:
   The masked observer continually observed the dog from the start of injection until time of death for the presence of any indicators of perceived pain or distress. The adverse reactions were defined as follows.
   Agonal Breathing: Gasp(s) of breath occurring at the moment of or just before death.
   Opisthotonus: Tonic spasm in which the head is bent backward, the limbs extended, the back arched and the chest bowed forward.
   Vocalization: Crying, whining, growling or barking.
   Other: The observer described any other adverse reactions.

4. Electrocardiograph (ECG) Measurements:
   Baseline readings were recorded immediately prior to dosing. Readings (25 mm/sec) were recorded continuously through dosing until time of death (minimum of 3 minutes), then for 15 seconds at 15 to 20 minutes post-injection to confirm death. The heart rate (beats/min), rhythm (normal or arrhythmic) and time to cessation of electrical activity were evaluated.

5. Electroencephalograph (EEG) Measurements:
   Baseline readings were recorded immediately prior to dosing. Readings (25 mm/sec) were recorded continuously through dosing until time of death (minimum of 3 minutes), then for 15 seconds at 15 to 20 minutes post-injection to confirm death. The time to cessation of electrical activity was evaluated.

Results:
1. Quality of Induction Score:

All dogs, with the exception of one dog in Group 2, scored a zero (excellent). The one dog in Group 2 scored a one (Good).

2. Timed Events:
   The timed events were recorded to the minute, not the second. Therefore, events are rounded to the nearest minute. Clarification between 1 and 59 seconds is not possible.

   For all dogs in Groups 1, 2 and 4, the time to collapse, apnea and heartbeat was less than one minute. In Group 3, the time to apnea was one minute for two dogs and the time to the last audible heartbeat was 2 and 4 minutes for two dogs.

   For all dogs in Groups 2 and 4, the time to death was less than a minute. In Group 1, the time to death was less than a minute for four dogs, one minute for two dogs and four minutes for two dogs. In Group 3, the time to death was less than one minute for 3 dogs, one minute, two, four, six and seven minutes for each of the remaining seven dogs.

   The average time to confirmation of death was 18.5, 16.2, 19.4 and 16.9 minutes for Groups 1, 2, 3, and 4, respectively.

3. Adverse Reactions:
   Table 5 shows the incidence of adverse reactions between the four treatment groups.

Table 5: Incidence of Adverse Reactions

Adverse Reaction	**Number of Dogs (%)**			
	Group 1	**Group 2**	**Group 3**	**Group 4**
Agonal Breathing	0 (0%)	0 (0%)	1 (12.5%)	3 (37.5%)
Vocalization	0 (0%)	1 (12.5%)	0 (0%)	0 (0%)
Minor Respirations	0 (0%)	0 (0%)	2 (25%)	0 (0%)
Minor ECG Electrical Activity	0 (0%)	0 (0%)	1 (12.5%)	0 (0%)
Return of Audible Heart Sounds	1 (12.5%)	0 (0%)	3 (37.5%)	0 (0%)
Additional Drug Required	0 (0%)	0 (0%)	2 (25%)	0 (0%)

4. Electrocardiograph Measurements:
   In Group 1, electrical activity ceased between 15 and 20 minutes in 7 of the 8 dogs. One dog demonstrated infrequent nonpulsatile and abnormal ventricular ECG activity for 24 minutes, which began approximately 4 minutes after TRIBUTAME Euthanasia Solution administration. This activity was considered to be nonfunctional electrical activity.

   In Group 2, electrical activity ceased by 20 minutes in all eight dogs.

In Group 3, electrical activity ceased between 15 and 20 minutes in 4 of the 8 dogs. Four dogs demonstrated infrequent nonpulsatile and abnormal ventricular activity lasting between 21 and 26 minutes. This ventricular activity began approximately 4 minutes after TRIBUTAME Euthanasia Solution administration. This activity was considered to be nonfunctional electrical activity.

In Group 4, electrical activity ceased between 15 and 20 minutes in 5 of the 8 dogs. Three dogs demonstrated infrequent nonpulsatile and abnormal ventricular activity lasting between 15 and 20 minutes. This ventricular activity began approximately 4 minutes after TRIBUTAME Euthanasia Solution administration. This activity was considered to be nonfunctional electrical activity.

5. Electroencephalograph (EEG) Measurements:
   Table 6 shows the arithmetic mean and range time to a cessation of electrical activity on the electroencephalograph. On average, the dogs in group 2 achieved cessation of electrical activity faster than the dogs in the other three treatment groups.

Table 6: Time to Cessation of Electrical Activity in the Brain

Treatment Group	Mean (in seconds)	Range (in seconds)
1	25.9	20-30
2	23.3	16-32
3	41.4	25-61
4	33.9	26-44

Conclusion: The results of this study show that the 0.45 mL/kg dose given as a single intravenous bolus produces a smooth induction and apnea, collapse, loss of audible heartbeat and a flatline EEG within one minute of the injection. The ECG continued to show electrical activity for up to 20 minutes in this group. The adverse reaction of vocalization was seen in one dog in this group.

A Clinical Field Trial for Euthanasia of Dogs with EMBUTANE Solution.
Study Number: PSI –0736-00C-002

Investigators:
The study was performed at six geographical locations within the U.S. Study sites consisted of five veterinary practices and one humane society/animal control facility. Several sites are missing because those investigators agreed to participate in the study but then subsequently left the study prior to enrolling any animals. The remaining Investigators that enrolled animals are shown below.

Site B: James Preuter, D.V.M.
Bedford Heights, OH

Site G: Robert Herzog, D.V.M.
Lee's Summit, MO

Site C: Signe Plunkett, D.V.M
Phoenix, AZ

Site D: John Mulnix, D.V.M.
Fort Collins, CO

Site E: Darrell Haeker, D.V.M.
St. Joseph, MO

Site H: Sarah Colburn, D.V.M.
Mesa, AZ

Site I: Bernard Mangone, D.V.M.
Phoenix, AZ

Study Purpose: The purpose of this study was to determine the field safety and effectiveness of TRIBUTAME Euthanasia Solution in veterinary practices in the U.S.

Test Animals: Eighty-five dogs of both genders and representing 46 breeds, met the study entrance criteria. These dogs were between 0.2 to 15.5 years old and weighed between 1.1 to 58.3 kilograms. One dog was hit by a car and died before administration of the TRIBUTAME Euthanasia Solution. Therefore, 84 dogs received the test article. Three dogs were removed from the study, leaving 81 valid cases that were included in the data evaluation. Of the three dogs that were removed from the study, two dogs were removed due to TRIBUTAME Euthanasia Solution overdose and one dog was removed due to concomitant treatment with acepromazine.

Control: Each animal acted as its own control.

Treatment Groups: This study contained one treatment group. All dogs received the TRIBUTAME Euthanasia Solution.

Masking: Because all dogs were assigned to a single treatment group, this study was not masked.

Dosage Form: Solution for injection. The formulation used in this study was not final formulation. The final formulation is the same formulation that was used in this study, except that the bitter agent, denatonium benzoate, has been added to this formulation so that the solution is less likely to be taken orally by accident or without knowledge.

Dosage: 0.45 mL/kg

Route of Administration: Single-bolus intravenous injection.

Variables Measured:

1. Quality of Induction Score:
   The Investigator or backup veterinarian subjectively assessed the clinical attainment of unconsciousness and assigned a score, based on the following definitions.

0 = Excellent: Quietly collapses without any sign of excitement, struggling or distress.
1 = Good: Quietly collapses with only minor, fleeting signs of excitement, struggling or distress.
2 = Poor: Exhibits a brief period of excitement before collapsing with noticeable signs of struggling or distress.
3 = Unacceptable: Exhibits marked excitement during induction with vigorous struggling or marked signs of distress.

2. Reaction to Injection Score:
   The Investigator or backup veterinarian subjectively assessed the clinical emergence of pain or discomfort at the injection site and assigned a score, based on the following definitions.
   0 = No pain/discomfort: Animal does not react to injection; appears unaware of injection.
   1 = Slight pain/discomfort: Animal turns head toward injection site, but makes no other movement or sign of pain or discomfort.
   2 = Moderate pain/discomfort: Animal lifts leg or tries to pull away from handler.
   3 = Severe pain/discomfort: Animal cries, struggles violently to pull away from handler.

3. Timed Events:
   The Investigator or a backup veterinarian(s) determined the study clock time (to the second) of start of initial injection, end of initial injection, (and if needed - start of additional injection, end of additional injection), collapse, apnea, loss of palpebral reflex, loss of visible heartbeat, and death.

4. Adverse Reactions:
   The Investigator or backup veterinarian(s) continually observed the dog from the start of injection until time of death for the presence of any indicators of perceived pain or distress. The adverse reactions were defined as follows.
   Agonal Breathing: Gasp or gasps occurring at the moment of or just before death.
   Opisthotonus: Tonic spasm in which the head is bent backward, the limbs extended, the back arched and the chest bowed forward.
   Vocalization: Crying, whining, growling or barking.
   Other: The Investigator or backup veterinarian described any other adverse reactions.

Results:

1. Quality of Induction Score:
   Table 7 shows the distribution of Quality of Induction Score.

Table 7: Distribution of Quality of Induction Score

Quality of Induction Score	Number of Dogs (%)
0 (excellent)	60 (74.1%)
1 (good)	16 (19.7%)
2 (poor)	5 (6.2%)
3 (unacceptable)	0 (0%)

2. Reaction to Injection Score:
   Table 8 shows the distribution of Reaction to Injection Score.

Table 8: Distribution of Reaction to Injection Score

Reaction to Injection Score	Number of Dogs (%)
0 (no pain)	69 (85.2%)
1 (slight)	3 (3.7%)
2 (moderate)	1 (1.2%)
3 (severe)	8 (9.9%)

3. Timed Events:
   Table 9 shows the arithmetic mean and range of the timed events.

Table 9: Timed Events Results

Timed Event[1]	Mean (in Seconds)	Range (in Seconds)
Collapse	20	5-58
Apnea	31	9-78
Loss of Palpebral Reflex	34	10-78
Loss of Palpable Heart Beat	44	8-205
Death	53	12-282

[1] All times were measured from the start of the injection until the event.

4. Adverse Reactions:
   Adverse reactions were recorded for 35.8% (29/81) of the study animals. The lowest reported incidence of side effects occurred at site B with 2 out of 15 animals (13.3%), and the highest reported incidence occurred at site E with 10 out of 16 animals (62.5%). Table 10 shows the distribution of adverse reactions in the 81 study animals. In two of the cases of agonal breathing, the Investigator reported the occurrence of agonal breathing 104 seconds and two minutes after the time of death.

Table 10: Incidence of Adverse Reactions

Adverse Reaction	Number of Dogs (%)
Agonal breathing	20 (24.6%)
Vocalization	8 (9.9%)
Muscle twitching[1]	4 (4.9%)
Opisthotonus	1 (1.2%)
Excitability	1 (1.2%)
Swallow reflex	1 (1.2%)
Attempt to stand	1 (1.2%)
Anxious	1 (1.2%)
Front limb extension	1 (1.2%)

[1] Includes muscle tremors and fasciculations.

Two dogs received an overdose of TRIBUTAME Euthanasia Solution. Both dogs experienced muscle twitching and one dog experienced agonal breathing that did not cease until approximately one minute after the time of death. Additionally, one dog was administered acepromazine maleate to facilitate diagnosis. This dog experienced opisthotonus and agonal breathing at the time of death and 100 seconds after death.

Conclusions:

This study shows that TRIBUTAME Euthanasia Solution induces collapse and death in an average of 20 and 53 seconds, respectively. However, the study also shows that 35.8% of the dogs experienced at least one adverse reaction. The most frequent adverse reaction was agonal breathing (24.6%). These adverse reactions, although potentially unacceptable to observers, did not result in undesirable side effects for the dogs that unacceptably compromised induction of unconsciousness and death. It is necessary to use the correct dose as overdose may increase the type and magnitude of adverse reactions. The use of tranquilizers or sedatives, such as acepromazine, prior to administration of TRIBUTAME Euthanasia Solution is not recommended due to the adverse reaction that was observed in one case.

A Clinical Study for the Comparison of EMBUTANE Euthanasia Solution with denatonium benzoate and EMBUTANE Euthanasia Solution in Regard to Injection Site Irritation in Dogs.
Study Number: PSI –0736-03C-019

Investigator: This study was performed at one site, an animal shelter.
Robert Hertzog, D.V.M.
Lee's Summit, MO

Study Purpose: The purpose of this study was to compare the injection response of dogs treated with TRIBUTAME Euthanasia Solution formulated with and without denatonium benzoate.

Test Animals: Twenty-two dogs, ranging in age between 4 months and 13 years and weighing between 3.8 and 33.2 kilograms were included in the study.

Control: TRIBUTAME Euthanasia Solution (without denatonium benzoate).

Treatment Groups:
The 22 dogs were randomly assigned to one of two equal treatment groups in blocks of four animals (two animals of each group in each block). See Table 11 below.

Table 11: Treatment Groups

Treatment Group	Test Article	Dose (mL/kg) injected IV within 10 to 15 seconds[1]	Number and Sex of Animals
I	TRIBUTAME with denatonium benzoate	0.45	4 males, 7 females
II	TRIBUTAME without denatonium benzoate	0.45	7 males, 4 females

[1] One dog in Group I was administered the test article in 16 seconds.

Masking:
Both test articles are identical in appearance. A separate individual (Test Article Technician), who was not involved in any observations or data acquisition, prepared the dose and handed the syringe with the test or control article to the Investigator. The Investigator made all injections and observations.

Dosage Form: Solution for injection. The formulation used in this study was the final formulation.

Dosage: 0.45 mL/kg

Route of Administration: Single-bolus intravenous injection.

Variables Measured:
1. Quality of Induction Score:
   The Investigator subjectively assessed the clinical attainment of unconsciousness and assigned a score, based on the following definitions:
   0 = Excellent: Quietly collapses without any sign of excitement, struggling or distress.
   1 = Good: Quietly collapses with only minor, fleeting signs of excitement, struggling or distress.
   2 = Poor: Exhibits a brief period of excitement before collapsing with noticeable signs of struggling or distress.
   3 = Unacceptable: Exhibits marked excitement during induction with vigorous struggling or marked signs of distress.

2. Reaction to Injection Score:
   The Investigator subjectively assessed the clinical emergence of pain or discomfort at the injection site and assigned a score, based on the following definitions.
   0 = No pain/discomfort: Animal does not react to injection; appears unaware of injection.
   1 = Slight pain/discomfort: Animal turns head toward injection site, but makes no other movement or sign of pain or discomfort.
   2 = Moderate pain/discomfort: Animal lifts leg or tries to pull away from handler.
   3 = Severe pain/discomfort: Animal cries, struggles violently to pull away from handler.

3. Timed Events:
   The Investigator determined the study clock time (to the second) of start of initial injection, end of initial injection, collapse, apnea, and death, which was defined as the inability to visualize, palpate or auscultate a heartbeat.

4. Adverse Reactions:
   The Investigator monitored the emergence of adverse reactions from start of the injection until 5 minutes after time of death. The Investigator categorized the adverse reaction into "Major" or "Minor" based on the following criteria. See Table 12 below.

   Table 12: Adverse Reactions

Major Adverse Reactions	**Agonal Breathing**	Gasps or gasps for air after injection of euthanasia solution. Exchange of air is evident.
	Opisthotonus	Tonic spasm in which the head is bent backward, the limbs extended, the back arched and the chest bowed forward.
	Vocalization	Crying, whining, growling or barking after completion of injection of euthanasia solution.
	Post-Injection Struggling	Any attempts to struggle after completion of injection of euthanasia solution
Minor Adverse Reactions	**Minor to Normal Respiratory Efforts**	Shallow to normal respiratory efforts or slight movements of the chest wall with or without exchange of air after time of death. No gasping sounds are made
	Movement	Head, Limbs, Muscle Fasciculation or Other
	Other	The Investigator described any other adverse reactions (for example, swallowing or licking)

Results:

1. Quality of Induction Score:

Table 13 shows the distribution of Quality of Induction Scores between the two treatment groups.

Table 13: Distribution of Quality of Induction Score

Quality of Induction Score	Number of dogs (%)		Total
	Group I	Group II	
0 (Excellent)	8 (72.7%)	8 (72.7%)	16 (72.7%)
1 (Good)	3 (27.3%)	2 (18.2%)	5 (22.3%)
2 (Poor)	0 (0%)	1 (9.1%)	1 (4.5%)
3 (Unacceptable)	0 (0%)	0 (0%)	0 (0%)

2. Reaction to Injection Score:
   Table 14 shows the distribution of Reaction to Injection Score between the two treatment groups.

Table 14: Distribution of Reaction of Injection Score

Reaction to Injection Score	Number of dogs (%)		Total
	Group I	Group II	
1 (No pain/discomfort)	9 (81.8%)	6 (54.5%)	15 (68.2%)
2 (Slight pain/discomfort)	2 (18.2%)	3 (27.3%)	5 (22.7%)
3 (Moderate pain/discomfort)	0 (0%)	1 (9.1%)	1 (4.5%)
4 (Severe pain/discomfort)	0 (0%)	2 (18.2%)	2 (9.1%)

3. Timed Events:
   Table 15 shows the arithmetic mean and range of the timed events.

Table 15: Timed Events Results

Timed Event[1]	Group I		Group II	
	Mean (in Seconds)	Range (in Seconds)	Mean (in Seconds)	Range (in Seconds)
Injection Time	13	11-16	13	10-15
Time to Collapse	18	11-23	16	12-22
Time to Apnea	32	20-66	27	19-68
Time to Death	33	20-63	30	20-60

[1] From start of injection

4. Adverse Reactions:
   Table 16 shows the incidence of adverse reactions between the two groups.

Table 16: Incidence of Adverse Reactions

Adverse Reaction	Number of Dogs (%)		Total
	Group I	Group II	
Agonal Breathing	2 (18.2%)	3 (27.3%)	5 (22.8%)
Opisthotonus	0 (0%)	0 (0%)	0 (0%)
Vocalization	2 (18.2%)	0 (0%)	2 (9.1%)
Post-Injection Struggling	1 (9.1%)	1 (9.1%)	2 (9.1%)
Minor Respirations	5 (45.5%)	3 (27.3%)	8 (36.4%)
Movement	3 (27.3%)	2 (18.2%)	5 (22.7%)
Licking	3 (36.4%)	6 (54.4%)	9 (40.9%)
Upward Bending of Neck	1 (9.1%)	1 (9.1%)	2 (9.1%)

Conclusions:
The study showed that the addition of the bitter agent, denatonium benzoate, did not result in an increase of pain or discomfort during the injection process. The study also repeated the high incidence of adverse reactions, specifically agonal breathing and vocalization that was seen in the large field study. This observation indicates that these incidences of adverse reactions are likely to be seen in the general population.

3. TARGET ANIMAL SAFETY:

Within the framework of evaluating a euthanasia agent, safety in the dog refers to rapid, painless induction of unconsciousness and clinical and physiologic death. Although this product results in potentially objectionable side effects from the observer's perspective, the field and laboratory effectiveness studies adequately demonstrated unconsciousness and death. Therefore, no additional safety studies were required.

4. HUMAN SAFETY:

This drug is intended for use in dogs, which are non-food animals. Because this new animal drug is not intended for use in food-producing animals, data on human safety pertaining to drug residues in food were not required for approval of this NADA.

Human Warnings are provided on the product label as follows: "Not for use in humans. Keep out of reach of children. Not for use in animals intended for food. TRIBUTAME Euthanasia Solution is lethal when administered in accordance with label directions. Keep in a secured area with authorized access only. In case of ingestion or accidental injection in a human, call a poison control center immediately. There are no known reversal agents. It is recommended that TRIBUTAME Euthanasia Solution be administered by a veterinarian or by a trained technician under the direct supervision of a veterinarian. In case of accidental exposure to skin, this product may cause numbness or local anesthesia due to the presence of lidocaine

in the formulation. Wash with soap and water immediately. Upon exposure to the eyes, flush eyes with copious amounts of cold tap water for 15 minutes. Avoid contact of the drug with open wounds or inadvertent self-inflicted injections. The following procedures should be taken in case of accidental exposure. The wound should be thoroughly washed with cold tap water and compression applied laterally to the site of puncture. Regular first aid measures must be applied for prevention of infection of puncture wounds if accidental exposure occurs. The drug should be removed as much as possible from the exposure site (flushing with cold tap water and/or use of suction pump). For a copy of the Material Safety Data Sheet (MSDS), or to report possible adverse reactions, call 1-800-759-3664."

5. AGENCY CONCLUSIONS:

The data submitted in support of this NADA satisfy the requirements of section 512 of the Federal Food, Drug, and Cosmetic Act and 21 CFR Part 514 of the implementing regulations. The data demonstrate that TRIBUTAME Euthanasia Solution, when used under the labeled conditions of use is safe and effective for euthanasia in dogs only.

TRIBUTAME Euthanasia Solution is restricted to use by or on the order of a licensed veterinarian because professional expertise is needed to properly administer the product.

Under Section 512 (c)(2)(F)(ii) of the Federal Food, Drug, and Cosmetic Act, this approval qualifies for THREE years of marketing exclusivity beginning on the date of approval because new studies for substantial evidence of effectiveness were conducted.

TRIBUTAME Euthanasia Solution is under the following U.S. patent numbers:

U.S. Patent Number	Date of Expiration
5,290,775	March 1, 2011
5,281,611	January 25, 2011

6. ATTACHMENTS:

Facsimile Labeling is attached as indicated below:

Bottle Label with Insert
Shipping Carton Label

7. LITERATURE CITED:

1. Lindner. Internal document reporting the evaluation of embutramide as a potential hypnotic and analgesic. Germany. April 21, 1958.

2. Sawyer DC. Ecklamide: A new euthanasia solution. Michigan State University. Unpublished report, 1991. (Note: the name Ecklamide was later changed to Embutane and then Tributame).
3. Michael TAD and Alwassadeh S. The effects of acute chloroquine poisoning with special references to the heart. American Heart Journal. 79(6): 831-842, 1970.
4. Sodfola OA. The cardiovascular effect of chloroquine in anesthetized dogs. Canadian Journal of Physiology and Pharmacology. 58: 836-841, 1980.
5. Nath S, Haggmark S, Johansson G, et al. Differential depressant and electrophysiologic cardiotoxicity of local anesthetics: An experimental study with special reference to lidocaine and bupivacaine. Anesthesia Analgesia. 65: 1263-1270, 1986.