Effects of Rhodamine B on Palatability of Invasive Wild Pig Baits

John C. Kinsey
Kerr Wildlife Management Area, Texas Parks and Wildlife Department, Hunt, Texas and Department of Environmental Science, University of Texas at San Antonio, San Antonio, Texas

Joshua R. Coward
Borderlands Research Institute, Sul Ross State University, Alpine, Texas

Justin A. Foster
Kerr Wildlife Management Area, Texas Parks and Wildlife Department, Hunt, Texas

Nathan P. Snow and Kurt C. VerCauteren
US Department of Agriculture, Animal and Plant Health Inspection Service, Wildlife Services, National Wildlife Research Center, Fort Collins, Colorado

ABSTRACT: Biomarkers have been used to quantify consumption of toxicants and other pharmaceutical baits by free ranging wildlife populations. Previous research has tested the efficacy and persistence of Rhodamine B (RB) as a biomarker in invasive wild pigs. However, little information is available about effects of RB on palatability of baits meant for invasive wild pigs, and studies have shown that the addition of RB to otherwise palatable baits reduces consumption by some species. HOGGONE® has been identified as an effective Sodium Nitrite-based oral toxicant for invasive wild pigs in trials conducted in captive pen trials. We simultaneously conducted five separate 2-choice tests to examine potential differences in consumption between HOGGONE® placebo paste (standard placebo) and HOGGONE® placebo paste containing 0.5% RB (RB placebo) in five groups of three invasive wild pigs. Each group was simultaneously presented with equal amounts of standard placebo and RB placebo paste for one night and monitored with remote cameras. Remaining bait was weighed and subtracted from the initial weight of both feed types to calculate consumption. There were no differences in the total amount of bait consumed or the time spent feeding between the two bait types across all five groups. Results of this study suggest that the addition of RB does not negatively impact consumption of HOGGONE® placebo paste by groups of invasive wild pigs. Thus, we provide more evidence that RB will be a useful tool for research on wild pigs, such as estimating proportions of free-ranging populations consuming baits that contain toxins or pharmaceuticals.

KEY WORDS: bait, biomarker, feral swine, palatability, Rhodamine B, Sus scrofa, vertebrate pest control, wild boar, wild pig

INTRODUCTION
Invasive wild pigs (Sus scrofa) are one of the most destructive invasive species in the world (Low et al. 2000). Pimentel (2007) reported that each invasive wild pig has an associated annual cost of $300, which means a hypothetical invasive wild pig population of 5 million could cost $1.5 billion annually. Most of the economic damage done by invasive wild pigs is to the agricultural industry, however, the ecological impacts of this species are widespread and impossible to quantify (Pimentel et al. 2002, Sward et al. 2004). Populations of invasive wild pigs are projected to continue growing and expanding their ranges, which will lead to increased economic and ecological costs associated with damages (Timmons et al. 2012, Snow et al. 2017c).

Trapping, shooting, aerial gunning, snaring, and other lethal means of population control have been found to be effective in the temporary reduction of localized invasive wild pig populations across the world (Choquenot et al. 1993, Mayer and Brisbin 2009, Campbell et al. 2010). These methods, though, have not been effective at reducing the invasive wild pig population long term or at a broad scale (Dickson et al. 2001). Additional methods of lethal population control, to be used in conjunction with the methods above, are necessary for large-scale reduction of invasive wild pig populations and their associated damages (Beasley et al. 2018).

A multidisciplinary, international team of scientists is currently working to develop HOGGONE® (Animal Control Technologies Australia P/L, Somerton, Victoria, Australia), a sodium nitrite-based toxic bait for the lethal control of invasive wild pigs (Snow et al. 2017a). HOGGONE® placebo has been found to be highly palatable to invasive wild pigs in both pen and free range studies (Snow et al. 2016, Snow et al. 2017b). The toxic bait has also shown to be highly lethal to groups of invasive wild pigs in a pen setting (Snow et al 2017b). The use of toxicants in the United States is strictly regulated by the United States Environmental Protection Agency (EPA) and at the time of this study, the efficacy of toxic HOGGONE® bait was yet to be tested in a free range setting in the US. Following completion of pen trials in 2016 and prior to initiation of free range toxic trials under an Experimental Use Permit from the EPA in 2018, a simulated free range toxic deployment was scheduled to be conducted using HOGGONE® placebo containing a biomarker (N. P. Snow, USDA-APHIS National Wildlife Research Center, unpubl. data). The addition of a biomarker to HOGGONE® placebo in a free range deployment could simulate consumption and associated mortality rates of both invasive wild pigs and non-target species during a toxic deployment, and provide
METHODS

Our study took place in April 2017 at the Texas Parks and Wildlife Department’s (TPWD) Feral Swine Research Facility located on the Kerr Wildlife Management Area (KWMA), Hunt, TX. Private landowners and USDA/APHIS/Wildlife Services personnel trapped free-ranging invasive wild pigs throughout nearby counties and TPWD employees transported them to KWMA via cattle trailer for housing and testing. While at the research facility, invasive wild pigs were fed Bluebonnet® Sow Ration Pellets (AC Nutrition, LP, Ardmore, OK) at 3-5% of group body mass, daily. Water was provided ad libitum from self-maintaining water troughs. All invasive wild pigs were group-housed in a 2.02 ha holding pen with naturally-growing vegetation and were provided supplemental shade structures as well as a small pond for wallowing. All experimental methods were approved by the TPWD-KWMA Institutional Animal Care and Use Committee (protocol 211072020151).

Prior to the trial, invasive wild pigs were moved through a chute system into a handling facility and three individuals were randomly selected to be placed in one of five trial pens (N = 15). Random assignment to groups was conducted under the condition that each animal’s weight was ≤50 kg due to staff safety while handling the animals. Upon selection, invasive wild pigs were moved into their respective pens for the trial (pens 23, 24, 25, QP 1, and QP 2). Invasive wild pigs were allowed one day to acclimate to their new environment and were provided water ad libitum as well as the same daily diet ration provided in the 2.02 ha holding pen.

Each trial pen was approximately 15 × 15 m and were setup following the methods outlined in Blass et al. (2016) with the exception that we placed four 58-l rubber feeding tubs (Marshalltown Company, Fayetteville, AR) in each pen rather than two. We placed two RECONYX PC800 remote cameras (RECONYX, Inc., Holmen, WI) above each feeding station with two tubs in the frame of each camera. We placed an additional camera at the back of each pen approximately 10 m from the feeding stations to observe group feeding behavior. Cameras were set to time-lapse mode and were scheduled to take a single photo every 15 seconds.

We conducted five simultaneous 2-choice tests to compare the consumption of standard placebo and RB placebo containing 0.5% Rb. Each bait was offered at 4% of group body weight calculated individually for each pen. Bait was evenly distributed across the four feeding tubs in an alternating pattern from left to right starting with RB bait (i.e. RB placebo, standard placebo, RB placebo, standard placebo). Baits were removed from each pen and weighed 15 hours after the start of the trial.

We calculated a conservative estimate of the RB concentration required in placebo HOGGONE to effectively mark the facial vibrissae of invasive wild pigs at which time the individual consumed a dose equivalent to the known LD₅₀ of 400 mg/kg for toxic HOGGONE® (Cowled et al. 2008). Given average single night consumption of placebo by free ranging invasive wild pigs of 300 grams (Snow et al. 2016), and the minimum dose for long term marking with RB 15-30 mg/kg (Webster et al. 2017), we estimated that a 75 kg pig would be effectively marked by 300 g of HOGGONE® with a 0.5% RB concentration and could be assumed dead had toxic HOGGONE® been consumed.

We compared relative preferences between RB placebo and standard placebo using two metrics: 1) the amount of each bait consumed and 2) the amount of time spent at each bait. We compared total bait consumption between RB placebo and standard placebo by measuring bait remaining after the 15 hour trial. We analyzed these data to determine if there were statistically significant differences in consumption using a Gosset’s-student’s- T-test. We compared time spent at each type of bait in each pen with camera data indexed using 15 second time lapse photos from remote cameras. These data were analyzed to determine if there were statistically significant differences in time spent at each bait using a Gosset’s student’s t-test. All statistics were performed in Microsoft Excel (Windows 2007-2010) and JMP (JMP®, Version 12. SAS Institute Inc., Cary, NC).

RESULTS

Statistical analyses suggest that there were no significant differences in consumption between RB placebo and standard placebo (P = 0.90, DF = 8). Of the 6.71 kg of total bait consumed in all pens, 3.51 kg was RB placebo and 3.20 kg was standard placebo (Figure 1).

Statistical analyses of camera data illustrate that total time spent at feed varied significantly among pens (P = 0.03, DF = 8), however there was no significant differences between time spent feeding at either bait type across all pens (P = 0.89, DF = 8) (Figure 2).
Results of this study indicate that the addition of RB at 0.5% of total bait composition did not significantly affect consumption of HOGGONE® placebo. Proportions of baits consumed in this study were similar to consumption results reported by Blass et al. (2016). Though there was no significant statistical difference in consumption between RB and standard placebo, more RB placebo was consumed across all pens and feed tubs containing RB placebo were the first tubs visited in four out of five pens. Both standard HOGGONE® placebo and the feed tubs used in this trial were black. The color contrast between the reddish-purple RB placebo and the black feed tubs may have introduced bias in this study even though domestic swine are believed to be dichromatic and invasive wild pigs have been shown to only consistently distinguish blue from other colors (Neitz and Jacobs 1989, Eguchi et al. 1997).

ACKNOWLEDGEMENTS

We would like to thank the Kerr Wildlife Management Area staff; B. Palm, F. Gutierrez, J. McCoy, L. Wolle, R. Reitz, and D. Frels, for all their hard work and dedication to this project and a special thank you goes out to J. Bustamante for the time he spent sorting photos. We would also like to thank anonymous reviewers for their comments on this manuscript. Mention of commercial products or companies does not represent an endorsement by the Texas or U.S. government.

LITERATURE CITED

Baruzzi, C., J. Coats, R. Callaby, D. P. Cowan, and G. Massei. 2017. Rhodamine B as a long-term semi-quantitative bait marker for wild boar. Wildlife Society Bulletin 41:271-277.

Beasley, J. C., S. Ditchkoff, J. Mayer, M. Smith, and K. C. VerCauteren. 2018. Research priorities for managing invasive wild pigs in North America. Journal of Wildlife Management 82:674-681.

Beasley, J., S. C. Webster, O. E. Rhodes, and F. L. Cunningham. 2015. Evaluation of Rhodamine B as a biomarker for assessing bait acceptance in wild pigs. Wildlife Society Bulletin 39:188-192.

Blass, C. R., N. P. Snow, J. C. Kinsey, J. A. Foster, and K. C. VerCauteren. 2016. Evaluation of potential food items as challenge diets in 2-choice tests with feral swine. Proceedings of the Vertebrate Pest Conference 27:174-177.

Campbell, T. A., D. B. Long, and B. R. Leland. 2010. Feral swine behavior relative to aerial gunning in southern Texas. Journal of Wildlife Management 74(2):337-341.

Choquenot, D., R. J. Kilgour, and B. S. Lukins. 1993. An evaluation of feral pig trapping. Wildlife Research 20(1):15-22.

Cowled, B. D., P. Elsworth and S. J. Lapidge. 2008. Additional toxins for feral pig (Sus scrofa) control: identifying and testing Achilles’ heels. Wildlife Research 35(7):651-662.

Dickson, J. G., J. J. Mayer, and J. D. Dickson. 2001. Wild hogs. Pages 191-208 in J. G. Dickson, editor. Wildlife of southern forests: habitat and management. Hancock House Publishers, Blaine, WA.

Evans, J., and R. Griffith. 1973. A fluorescent tracer and marker for animal studies. Journal of Wildlife Management 37(1):73-81.
Farry, S. C., S. E. Henke, A. M. Anderson, and F. M. Gayne. 1998. Responses of captive and free-ranging coyotes to simulated oral rabies vaccine baits. Journal of Wildlife Diseases 34(1):13-22.

Fleming, J. S., D. Choquenot, and R. J. Mason. 2000. Aerial baiting of feral pigs (Sus scrofa) for the control of exotic disease in the semi-arid rangelands of New South Wales. Wildlife Research 27:531-537.

Johns, B. E., and H. P. Pan. 1981. Analytical techniques for fluorescent chemicals used as systemic or external wildlife markers. Pages 86-93 in E. W. Schafer Jr. and C. R. Walker, editors. Vertebrate pest control and management materials. STP 752, American Society for Testing and Materials, Philadelphia PA.

Lindsey, G. D. 1983. Rhodamine B: a systemic fluorescent marker for studying mountain beavers (Aplodontia rufa) and other animals. Northwest Science 57(1):16-21.

Lowe, S., M. Browne, S. Boudjelas, and M. De Poorter. 2000. 100 of the world’s worst invasive alien species: a selection from the global invasive species database. Published by The Invasive Species Specialist Group (ISSG) a specialist group of the Species Survival Commission (SSC) of the World Conservation Union (IUCN), Auckland, New Zealand.

Mascari, T. M., and L. D. Foi. 2009. Evaluation of Rhodamine B as an orally delivered biomarker for rodents and a feed-through transstadial biomarker for phlebotomine sand flies (Diptera: Psychodidae). Journal of Medical Entomology 46(5):1131-1137.

Mayer, J. J., and I. L. Brisbin, Jr. editors. 2009. Wild pigs: biology, damage, control techniques and management. SRNL-RP-2009-00869. Savannah River National Laboratory, Aiken, SC.

Morgan, D. R. 1981. Monitoring bait acceptance in brush-tailed possum populations: development of a tracer technique. New Zealand Journal of Forest Science 11(3):271-277.

Neitz, J., and G. Jacobs. 1989. Spectral sensitivity of cones in an ungulate. Visual Neuroscience 2(2):97-100.

Pimentel, D. 2007. Environmental and economic costs of vertebrate species invasions into the United States. Pages 2-8 in G. W. Witmer, W. C. Pitt, and K. A. Fagerstone, editors. Managing vertebrate invasive species: proceedings of an international symposium. USDA/APHIS Wildlife Services, National Wildlife Research Center, Fort Collins, CO.