Ecological and Human Health Hazards from Broadcast Application of 0.005% Diphacinone Rodenticide Baits in Native Hawaiian Ecosystems

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ABSTRACT: In the early 1990s, a coalition of federal and state agencies, NGOs, and private landowners in Hawaii agreed to pursue a Special Local Needs pesticide registration [24(c) FIFRA] for the aerial broadcast of a 0.005% diphacinone rodenticide for the control of rodents in native ecosystems. While there was recognition of the important role introduced rodents play in the decline and extinction of native species, there were concerns expressed about the potential non-target impacts of this technique. Over the next 10 years, numerous studies were undertaken to address specific non-target issues. This research, along with other published and unpublished research on diphacinone and its human pharmaceutical counterpart, Dipaxin, was compiled and analyzed in 4 hazard assessments (human dietary and drinking water consumption, aquatic and terrestrial non-target species) that comprise the foundation of Hawaii's registration application. Hazards to humans and other non-target terrestrial organisms were evaluated in terms of dietary intake of contaminated food or water required before lethal or sublethal effects might be anticipated. Hazard to aquatic organisms was assessed according to traditional risk quotient methods employed by the U.S. Environmental Protection Agency. These assessments indicate the greatest human health hazard is to pregnant women drinking untreated stream water; however, even this risk is low. With a few exceptions, such as the Hawaiian crow, the ecological assessments indicate the acute risks to terrestrial or aquatic non-target species are minimal, even under the most conservative risk scenarios. However, there could be detectable physiological effects in birds exposed at sublethal levels. We believe that under proper supervision, this technique can be safely used in Hawaii, and elsewhere, to protect native species from the impacts of introduced rodents.

KEY WORDS: anticoagulant, diphacinone, endangered species, eradication, Hawaii, risk assessment, rodenticide

Proc. 22nd Vertebr. Pest Conf. (R. M. Timm and J. M. O'Brien, Eds.)
Published at Univ. of Calif., Davis. 2006. Pp. 413-433.

INTRODUCTION

The geographic isolation of the Hawaiian archipelago has provided the stage for development of one of the world's most unique suites of ecosystems. Because of this isolation, endemic flora and fauna evolved from the relatively few colonists that arrived on the islands via air, drifted across the ocean, or evolved from marine organisms. As with so many other island ecosystems, these communities evolved in the absence of most terrestrial mammals, with the exception of 2 species of bats. With the arrival and colonization of Hawaii by the Polynesians came significant changes in island habitats for agriculture as well as the introduction of animals like the pig (*Sus scrofa*), dog (*Canis familiaris*), and Polynesian rat (*Rattus exulans*) (Kirch 1982, Olson and James 1982*a*). The introduction of mammals has had a devastating impact on Hawaiian flora and fauna. Fossil evidence (Olson and James 1982*a*,*b*) indicates that over 40 species of native birds became extinct between the time Polynesians arrived and when Captain Cook landed in 1776 (Scott *et al*. 1986).

Three species of rats, the roof rat (*Rattus rattus*), Norway rat (*Rattus norvegicus*), and the Polynesian rat, and the house mouse (*Mus musculus*) now inhabit the islands (Tomich 1986, Scott *et al*. 1986, Sugihara 1997, Cox 1999). Because of their omnivorous and sometimes predatory feeding habits, rats have been implicated as one cause of the significant population declines observed in many species of native invertebrates, plants, and birds in Hawaii (Baker and Allen 1976, Atkinson 1977, Scowcroft and Sakai 1984, Stone 1985, Scott *et al*. 1986, Hadfield *et al*. 1993).

New Zealand's successful use of broadscale rodent control in native ecosystems to restore rare species (Innes *et al*. 1995, Towns and Broome 2003) led a coalition of federal and state agencies, non-governmental organizations (NGOs), and private landowners in Hawaii to pursue regulatory approval of rodenticides for conservation purposes. After reviewing rodenticides commercially available in the U.S., the Hawaii Toxicant Registration Working Group elected to pursue registration of diphacinone rodenticides, because of their effectiveness against rats in Hawaii (Tobin 1992), favorable environmental track record (Kaukeinen 1982, Lund 1988), and the use of diphacinone as a human pharmaceutical (Willis *et al*. 1953, Katz *et al*. 1954). In 1994, the first 24c registration under provisions of the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) was granted for a diphacinone product in bait stations for the protection of native species in Hawaii. Currently, HACCO Inc.'s Ramik® Mini Bars All-Weather Rat and Mouse Killer (SLN HI-980005) is registered in Hawaii for bait station use in natural areas.

Rodenticide bait stations can be used in easily accessible areas such as nut orchards or commensal areas, but become operationally impossible in large, heavily

vegetated or relatively inaccessible areas where frequent maintenance visits to bait stations are impossible (Nelson *et al*. 2002). Furthermore, this baiting strategy has proven more effective on black and Norway rats than on Polynesian rats. It is believed that poor Polynesian rat control may be due to their reluctance to enter bait stations (Swift 1998). Field trials in Hawaii have demonstrated that broadcast application techniques overcome these limitations (Dunlevy *et al*. 2000, Dunlevy and Campbell 2002, Spurr *et al*. 2003*a*,*b*).

On behalf of the Hawaii Toxicant Registration Working Group, HACCO Inc. is therefore applying for a 24c registration to hand and aerially broadcast a specially formulated form of Ramik® Green (0.005% diphacinone) in Hawaii for conservation purposes. The proposed product is larger (6.5 g) than the commercially available Ramik® Green products to increase the probability pellets will penetrate the forest canopy and settle on the forest floor. Proposed application directions include the following restrictions. Bait can be either hand broadcast or aerially broadcast from a differential GIS-equipped helicopter using a suspended bucket/spreader attachment. The application rate will be limited to a maximum single application rate of 14 kg of bait per hectare (12.5 lbs/ac). Five to 7 days after the first application, a second broadcast application can be made at a rate no greater than 14 kg/hectare. A second series of applications could occur no sooner than 2 months after the first series. The maximum yearly cumulative application rate may not exceed 56 kg/hectare (50 lbs/ac).

OBJECTIVES AND SCOPE OF HAZARD ASSESSMENT

FIFRA mandates that pesticides be evaluated for "unreasonable adverse effects to the environment" (FIFRA §3(c)(5)) and registered only if use of the product will not cause unreasonable impacts. FIFRA defines "unreasonable adverse effects" to include risk to "man and the environment taking into account the economic, social and environmental cost and benefit(s)" (FIFRA §2(bb)). In this assessment for broadcast application of diphacinone rodenticide baits for conservation purposes in Hawaii, only risks to humans and the environment are assessed. No determination is made regarding the economic, social, and environmental benefits rodent control in native ecosystems may produce. Four hazard assessments have been conducted to attempt to quantify the potential for non-target effects to humans, native terrestrial and aquatic species, and other species. These assessments have been conducted to anticipate EPA registration concerns and to address concerns brought up by members of the public, communities adjacent to areas proposed for treatment, wildlife managers, and others.

CONCEPTUAL MODEL AND ASSESSMENT ENDPOINTS

The conceptual model used in these assessments identifies the major primary and secondary routes of exposure evaluated (Figure 1). Exposure routes assessed for human hazard include sublethal effects for acute and chronic exposure due to the consumption of bait pellets, diphacinone-contaminated game (pigs and birds), and water (treated and untreated) originating within a potential treatment site. Human exposure due to consumption of aquatic organisms was not addressed, due to lack of residue data. However, given the extremely low diphacinone concentrations predicted in the water column, residues in tissues of fish and invertebrates would likely be low. Furthermore, this is an unlikely route of exposure. Streams and reservoirs stocked with introduced game fish would not be directly treated, and diphacinone concentrations in water from treated sites potentially flowing into these areas would be diluted to very low levels. While people do take native stream organisms, it is unlikely that they would consume enough to reach levels of concern for humans.

Aquatic organisms are assessed for risk of primary exposure to diphacinone in solution and from ingesting bait particles, and through secondary routes for those species that are predatory on other fish. With the exception of the Hawaiian short-eared owl (*Asio flammeus sandwichensis*), Hawaiian hawk (*Buteo solitarius*), and the Hawaiian hoary bat (*Lasiurus cinereus semotus*), all of the species or groups of terrestrial organisms identified in the conceptual model could potentially consume bait pellets and therefore will be assessed for primary hazard. Secondary exposure hazard is addressed for all species that could become exposed through consuming the carcasses of or preying upon exposed rodents. Rodents (roof rats, Norway rats, Polynesian rats, and house mice) are proposed as allowable targets under this registration, so risk to those species is not addressed. In addition, Small Indian mongooses (*Herpestes auropunctatus*) are not addressed because they are considered pests in Hawaii and are currently targeted on the bait station label for conservation use.

Assessment endpoints used in the hazard assessments for species other than humans include both death and impacts from sublethal exposure, which might lead to death under field conditions. Risk is evaluated for both single and multiple-day exposure by using toxicological test data collected under laboratory conditions. Many ecological hazard assessments stop at the potential for acute lethal exposure. However, the proposed broadcast baiting will be conducted in the habitats of threatened and endangered species, and minor impacts to T&E or other species of concern could have great implications. Consequently, the effect of sublethal exposure to diphacinone, which has been shown to cause illness and bleeding in test organisms, is evaluated in this hazard assessment. The species and exposure routes evaluated in this assessment are specified in Table 1. Human risk is conservatively estimated by basing all risk evaluations on sublethal effects. The toxicity values used to evaluate risk to humans are the lowest reported value shown to have even minor effects on test organisms. In nearly all cases, this is in relation to changes in the blood's clotting ability. In the case of maternal or developmental effects it is in terms of complications of pregnancy.

HAZARD ASSESSMENT

Hazards to humans are evaluated for people consuming meat from animals living and foraging in baited areas, and for people drinking water originating in

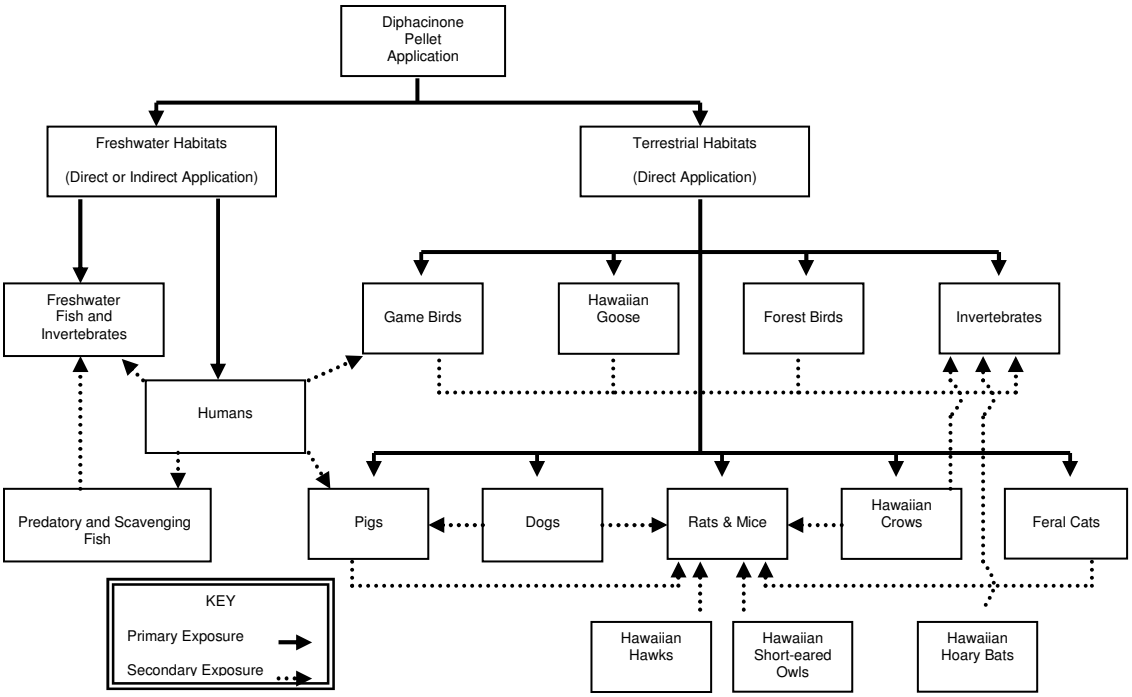

Figure 1. Conceptual model for assessing the risk posed by aerially broadcasting diphacinone rodenticide baits in native Hawaiian ecosystems.

Table 1. Species assessed and assessment endpoints in the human and ecological risk assessment of aerial broadcast application of a diphacinone rodenticide in native Hawaiian ecosystems.

Species	Primary Exposure	Secondary Exposure		Water
		Invertebrates	Rodent or Swine Tissue	
Humans	-	-	SL	SL
Freshwater fish	L	-	-	L
Freshwater Invertebrates	L	-	-	L
Terrestrial mollusks	L	-	-	-
Game birds	L, SL	L, SL	-	-
Non-game birds	L, SL	L, SL	-	-
Hawaiian goose (*Branta sandvicensis)*	L, SL	L, SL	-	-
Hawaiian hawk (*Buteo solitarius*)	-	-	L, SL	-
Hawaiian crow (*Corvus hawaiiensis)*	L, SL	L, SL	L, SL	-
Hawaiian short-eared owl (*Asio flammeus sandwichensis*)	-	-	L, SL	-
Swine (*Sus scrofa*)	L, SL	-	L, SL	-
Hawaiian hoary bat (*Lasiurus cinereus semotus)*	-	L, SL		-
Dog (*Canis familiaris*)*	L, SL	-	L, SL	-
Cat (*Felis catus*)*	L, SL	-	L, SL	-

L = Lethal acute exposure
SL = Sublethal chronic exposure
* Hazard calculations are presented in Table 6 but not discussed in the text.

baited areas. In these assessments, human health hazards were not estimated according to methods traditionally used by the U.S. Environmental Protection Agency. Instead, risks to humans are presented in terms of the minimum quantity of meat or water an adult female would have to consume to ingest amounts of diphacinone that have been shown to cause sublethal exposure effects in laboratory rats. These amounts are also compared with the dosages given to people in the early clinical trials for diphacinone's human pharmaceutical counterpart, Dipaxin. This methodology was used because it was felt the results would be more easily interpretable to a wider variety of audiences than traditional risk assessment outputs.

The ecological risk assessment is not exhaustive in terms of predicting risk for every organism in Hawaiian ecosystems. Instead, risk is evaluated according to traditional EPA risk assessment methodology, which employs

generic species as surrogates for species found in Hawaii. Evaluation of risk to terrestrial organisms goes beyond the risk quotient method employed by U.S. EPA in their 1998 assessment (U.S. EPA 1998). Like the human assessments, risk to terrestrial organisms is based on daily consumption estimates of bait or other contaminated dietary items required to ingest doses equivalent to published toxicity values for other species. Since toxicity data are somewhat limited, many of the estimates are driven by the organism's body weight. Traditional EPA methods were used to estimate risk to aquatic species. Risk quotients were calculated by dividing the highest expected diphacinone residue in food or water by the lowest toxicity value reported in the literature (RQ = exposure/toxicity). This applies the highest exposure scenario to the most vulnerable individuals. These risk quotients were then compared to EPA's established "Levels of Concern" (LOC). In all assessments, human or ecological, the most conservative parameters were chosen to estimate risk. Consequently, risk tends to be over-estimated.

Diphacinone Residues in Aquatic Environments

One of the habitat types proposed for aerial broadcast treatment is upper elevation rainforest. These forests often have streams that contain native aquatic organisms, and they can also be the watersheds that provide human drinking water. Therefore, it is critical to determine the diphacinone concentrations that could occur in streams and drinking water sources, and whether these levels would be of concern. The U.S. EPA, Office of Pesticide Programs' Water Quality Technical Team was consulted regarding the most appropriate method of determining residues in surface water. They indicated that standard surface water models (GENEEC and PRZM EXAMS) are not appropriate for modeling pesticide applications in forests and recommended modeling residues using a direct application to water model (Dr. Alex Clem, U.S. EPA, pers. commun.). This model incorporated the following assumptions: 1) the total volume of water should be based on actual stream flow data from a potential treatment site and utilize water volume estimates of the average minimum daily stream flow as well as the average mean daily stream flow; 2) the maximum application rate should be based on both 2-dimensional (2D) and 3-dimensional (3D) acreage estimates within drainages that might be impacted by future aerial applications; 3) the assessment should be based on making 2 applications at the maximum application rate (14 kg/ha or 12.5 lbs/acre) 5 days apart; and 4) it should be assumed that 100% of the diphacinone ends up in the water at the time of application.

We modeled the Hanawi watershed, located on the northeastern slope of Haleakala volcano in East Maui, one of the primary drainages in the Hanawi Natural Area Reserve. Hanawi NAR is a high priority area for rat control due to the large number of rare native species found there. At lower elevations, Hanawi Stream is diverted into the Koolau Irrigation Canal, which is a major source of irrigation and drinking water for the Upcountry area of Maui. The Hanawi drainage is typical of other potential application areas that receive high amounts of precipitation, in that stream channels are well defined and the vegetative community is characterized by an open forest canopy with dense understory vegetation. Soil surfaces are irregular and littered with decaying plant material, making active transport of baits into stream channels unlikely. However, due to the high volume of rain received in this drainage, standing water and saturated soils and sheet surface flows are common.

To ensure that steep slopes receive the correct bait application rate, a treatment area with a high degree of geographical relief should be quantified using 3 dimensions to incorporate these additional near-vertical surface areas. If the actual 3D acreage is considerably larger than the 2-dimensional area, a larger amount of bait would be applied within the treatment area. Two-dimensional and 3-dimensional acreage within the Hanawi watershed was determined by the U.S. Fish and Wildlife Service (unpubl. data, Ron Salz, Pacific Islands Fish and Wildlife Office, Honolulu, HI). Three-dimensional area was calculated from digital elevation models of the watershed using ArcGIS Spatial Analyst and 3D Analyst software. This analysis revealed that in the potential treatment region of the Hanawi watershed (>4,000 ft in elevation), 3D surface area is only 1.08 times greater than 2D area, so the 2D area above 4,000 ft (658 ha) was used to determine the total amount of bait that would be applied over the treatment area.

Surface water stream flow data obtained from the USGS (http://waterdata.usgs.gov) was used to determine the amount of water to which the bait would be applied. The USGS data included measurements of daily stream flow: peak daily stream flow, mean daily stream flow, and minimum daily stream flow. Examination of the data revealed enormous variation in daily stream flows over the 86 years records were kept. It is common for this area of Maui to experience extremely high rainfall in a very short period of time, resulting in short-duration flash floods. Additionally, flow could range from 1 cubic foot per second (CFS) to over 2,000 CFS over the course of a year. There also appeared to be a distinct dry season between early April and mid-September. Precipitation could exceed 635 cm (250 in) per year.

To calculate the worst case scenario of the highest possible diphacinone concentration in stream waters, several unlikely and inherently contradictory circumstances were used in the model: extremely low water levels in the stream and the transport of all of the applied bait from the forest floor and canopy into the main channel of the stream. Mass transport of bait would require tremendously heavy precipitation and sheet flow throughout the treatment area, which would immediately dilute the diphacinone concentration in the stream. To estimate the lowest stream flow level, the mean daily stream flow during the dry season minus 1 standard deviation was used because short-duration floods can significantly influence the average daily stream flow and tend to overestimate typical stream flows. Mean daily stream flows during the dry season averaged 45.5 million (±21.5 million) liters of water per day. Therefore, assuming all of the bait applied in both applications (28 kg/ha) ends up in this volume of surface water and all of the diphacinone goes into solution, the maximum dipha-

cinone concentration in the water of Hanawi Stream where it enters the Koolau Irrigation Canal would be approximately 0.020 ppm. If residues are based on minus one standard deviation of the mean flow, diphacinone concentration would be 0.038 ppm.

Hazard to Aquatic Organisms

Methodology used to conduct the aquatic organism risk assessment followed standard U.S. EPA guidelines (U.S. EPA 1998). This method involves comparing the diphacinone concentration predicted in the water column to the toxicity of the chemical to various aquatic organisms (Table 2). This comparison results in risk quotients (RQ = exposure/toxicity). RQs are then compared to EPA's established Levels of Concern (LOCs), which indicate potential risk to non-target organisms and the need to consider regulatory action. An RQ exceeding 0.5 indicates the compound and associated use pattern present an acute high risk to all aquatic organisms. An RQ exceeding 0.05 indicates that the use causes concern for endangered species.

The risk quotients calculated for the aerial application of diphacinone into Hanawi Stream are presented in Table 3. The highest risk quotient calculated for this application is for *Daphnia* (RQ = 0.021). None of the RQs for fish, crustaceans, or other aquatic invertebrates exceed 0.05, so no risk is predicted for any aquatic organism. Under actual use, the risk to aquatic organisms from diphacinone dissolved in the water column would be lower for a number of reasons. The modeled scenario assumes all of the treatment is made as a single application of 28 kg/ha (25 lbs/acre) and all of the bait goes directly into the water. It also ignores the fact that the diphacinone will not be applied as pure technical material, but will be incorporated into a grain bait product. Diphacinone has a high affinity for organic matter (U.S. EPA 1998) and will be released slowly from bait pellets. Furthermore, diphacinone has a low solubility in water (17 to 30 ppm) (WHO 1995, U.S. EPA 1998) and binds tightly to soil, so most of the diphacinone is expected to remain on the soil surface until it degrades. Surface water contamination would only occur via the movement of eroded bait or soil particles entering water bodies, and not by dissolution in runoff. In aquatic environments, diphacinone is expected to be partitioned in suspended and bottom sediments rather than in the water column (U.S. EPA 1998). All of these factors would significantly lower the concentrations in the water column and reduce the risk to aquatic organisms even more.

Aquatic organisms could also be exposed to diphacinone by consuming bait particles suspended in the water column or in bottom sediments. One study has attempted to evaluate the toxicity of diphacinone through this route. Ells' (1976) study with catfish included water column exposure to diphacinone leaching from contaminated sediment and, to a limited extent, consumption of contaminated sediment. The soil was treated at a nominal concentration of 0.32 mg diphacinone/kg soil. Water column concentrations of diphacinone peaked at 2 µg/L and averaged 1.4 µg/L, concentrations significantly lower than diphacinone's solubility or Hanawi Stream modeling estimates. Catfish were maintained in this system for a period of many months and were provided with supplemental feed. Ells' results were confounded by high mortality in the control group beginning on the third week of the study. However, the first mortality in the treatment group was not observed until day 36. The 96-hour LC_{50} for catfish was reported as 2.1 mg/L in Kosmin and Barlow (1976), but no information about the methods of this study is known. This value is slightly higher than the average concentration observed by Ells and, if valid, may explain why Ells' catfish were able to survive for 36 days. Additional exposure via consumption of contaminated sediment may have been minimized because of the supplemental feeding in Ells' study. These results are difficult to interpret, since no attempt was made to quantify how much diphacinone-contaminated material was eaten. Regardless, one would have to question whether the mortality was caused by diphacinone. No other studies on aquatic or terrestrial species report mortality as far out as 36 days from initial exposure. The studies available for assessing risk to fish do not provide information to draw strong conclusions. However, since Ells did report mortality, a conservative assumption is that bottom-feeding fish in streams in diphacinone-treated areas might ingest a lethal dose of bait particles.

Mortality due to secondary poisoning from predatory or scavenging fish eating contaminated fish has not been studied. U.S. EPA (1998) reports an octanol/water partition coefficient (P_{ow}) for diphacinone of Log P_{ow} = 4.27. Compounds with Log P_{ow} of greater than 4 are generally considered as being moderately lipophilic and having moderate bioaccumulation potential. This is reinforced by the fact that diphacinone has been detected in the tissues of many terrestrial organisms. It should be expected that if diphacinone enters the aquatic environment, fish and aquatic invertebrates are likely to accumulate residues. The degree of accumulation and therefore the risk to predators and scavengers is unknown. Many of the introduced fish species in Hawaii are predatory. Only one of the native Hawaiian freshwater fish, *Eleotris sandwicensis*, is highly predatory on other fish. Most of the other native fish species are scavengers, bottom feeders or drift feeders (Nishimoto and Kuamoo 1991).

Of the 3 pathways by which aquatic species may be exposed to diphacinone residues, adequate data are only available for assessing the risk posed by residues in solution in the water column. This risk appears to be minimal. In addition, insufficient data are available to address aquatic secondary hazards. The exposure route with the greatest risk potential is that caused by suspended bait particles in streams. However, there are a number of factors that reduce this risk. The broadcast application rate has been carefully determined to minimize the amount of bait used (Swift 1998, Dunlevy *et al*. 2000), and few pellets have remained uneaten by rodents for more than a few days during field trials (Dunlevy *et al*. 2000, Dunlevy and Campbell 2002, Spurr *et al*. 2003*a*,*b*). Additionally, vegetation is very dense in native rainforests where streams occur, and it will trap some of the bait in the tree canopy and understory. Bait that does reach the forest floor may become sufficiently lodged in vegetation and detritus on the soil surface to prevent it

Table 2. Acute oral toxicity of technical diphacinone to birds, mammals, and aquatic organisms.

Species	Toxicity (LD_{50} mg/kg & LC_{50} ppm)	Citation
Laboratory rat (*Rattus spp.*)	1.5 to 43.3	Correll *et al.* 1952, Bentley and Larthe 1959, Kusano 1974, Goldenthal *et al.* 1975, Kosmin and Barlow 1976, Shapiro 1990
Mouse (*Mus spp.*)	28.0 to 340	Correll *et al.* 1952, Kusano 1974, Kosmin and Barlow 1976
Pine vole (*Microtus pinetorum*)	67.7	Byers 1978
Meadow vole(*Microtus pennsylvanicus*)	11.7	Byers 1978
Rabbit (*Oryctolagus spp.)*	35.0	Correll *et al.* 1952
Dog (*Canis domesticus*)	2.0 to 3.0 45.0 (single dose - 4/6 died)	Evans and Ward 1967, Lisella *et al.* 1971, Mount and Feldman 1983, Travlos *et al.* 1984
Coyote (*Canis latrans*)	0.6; no mortality at 0.9	Savarie *et al.* 1979, Sterner 1979
Cat (*Felis catus*)	15.0	RTECS 2002
Mongoose (*Herpestes auropunctatus*)	0.2	Keith and Hirata 1987
Pig (*Sus sp.*)	> 150.0	Hazelton 1957
Ferret (*Mustela furo*)	21.4 (in fish paste)	Spurr *et al.* 2005
Cow (*Bovis spp.)*	5 (no ill effects at this dose)	Thompson *et al.* 1972
Vampire bat (*Desmodus rotundus)*	0.91	Thompson *et al.* 1972, Fernandez 1973
Brown treesnake (*Boiga irregularis*)	$20 < LD_{50} < 40$	Brooks *et al.* 1998
Mallard (*Anas platyrhynchos*)	3160	Erickson and Urban 2004
Northern bobwhite (*Colinus virginianus*)	$400<LD_{50}<2{,}000$	Campbell *et al.* 1991
Northern bobwhite (*Colinus virginianus*)	LC_{50} = 5,000 ppm	Long *et al.* 1992*a*
Mallard (*Anas platyrhynchos*)	LC_{50} >906 ppm	Long *et al.* 1992*b*
Rainbow trout (*Oncorhynchus mykiss*)	LC_{50} 2.6 to 2.8 ppm	Kosmin and Barlow 1976, Machado 1994*a*
Bluegill sunfish (*Lepomis machrochirus*)	LC_{50} = 7.5 to 7.6 ppm	Kosmin and Barlow 1976, Machado 1994*b*
Channel catfish (*Ictalurus punctatus*)	LC_{50} = 2.1 ppm	Kosmin and Barlow 1976
Water flea (*Daphnia magna*)	LC_{50} = 1.8 ppm	Putt 1992
Pink shrimp (*Penaeus duorarum)*	LC_{50} >10 ppm	Kosmin and Barlow 1976
Fiddler crab (*Uca pugilator)*	LC_{50} >10 ppm	Kosmin and Barlow 1976

Table 3. Risk quotient analysis for acute risk to aquatic species from exposure to theoretical diphacinone residues in Hanawi Stream.

Organism	EC_{50} (ppm)	Risk Quotients (Based on EEC = 0.038 ppm[1])
Rainbow trout[2]	2.6	0.016
Bluegill sunfish[3]	7.5	0.005
Channel catfish[4]	2.1	0.018
Daphnia[5]	1.8	0.021
Pink shrimp[4]	> 10	0.004
Fiddler crab[4]	> 10	0.004

[1] Based on treating the entire Hanawi drainage above 4000 feet in elevation at the maximum application rate (2 x 14 kg/ha) at one standard deviation below the dry season mean stream flow

[2] Machado 1994*a*

[3] Machado 1994*b*

[4] Kosmin and Barlow 1976

[5] Putt 1992

from being transported into nearby streams. If baiting occurs during the dry season, this possibility of bait being transported into streams by surface flow becomes even less likely. As demonstrated by Dunlevy *et al*. (2000), the bait itself is very durable and resistant to disintegration or crumbling, reducing the probability that small bait particles would be transported into streams. Thus, the actual risk to aquatic organisms will be low.

Diphacinone Residues in Drinking Water

Hanawi Stream is near the origin of the Koolau Irrigation Canal, which collects water from numerous streams and drainages between Hanawi Stream and the Kamaole Weir. The water is treated at the Kamole Weir and supplied as the primary drinking water source to approximately 6,000 residences in the Upcountry region of Maui (Dept. of Water, Maui County 2003). Average daily flow at the last gauge prior to the Kamaole Weir was approximately 106.1 million gallons per day and ranges from 90.3 to 121.8 million gallons per day, according to annual ditch flow records obtained for the years 1999 through 2002 (Hew 1999, 2000, 2001, 2002). Using this water flow estimate and assuming the entire Hanawi watershed above 4,000 ft is baited, the predicted maximum diphacinone concentration entering the Kamaole Weir is 0.003 ppm. The Kamaole Weir Water Treatment Facility employs a microfiltration technology that is designed to filter out particles greater than 0.2

micrometers (http://www.usfilter.com/Memcor/cmfbasics .htm). Since a diphacinone molecule is smaller than 0.2 microns, this system would not remove diphacinone in solution (Lisa Sorgini, U.S. Filter, pers. commun.). However, because diphacinone has a high affinity for organic material, it is reasonable to assume the diphacinone incorporated in suspended bait particles or adhered to other organic material would be removed by the filtration system.

Toxicity of Diphacinone to Humans

The effects of diphacinone are well-characterized due to its use as a human pharmaceutical. Numerous clinical human studies were conducted in the 1950s to establish a therapeutic diphacinone dose for use on patients requiring anti-clotting medication. Willis *et al*. (1953) administered diphacinone to 64 patients in an initial dose of 30 mg followed by 40 mg in 3 doses over the next 36 hours (13.3 mg every 12 hours). The average daily maintenance dose was 5 to 10 mg given either once or twice daily. The mean effective dose was reported as 63 mg. After collecting data on 43 patients, Duff *et al*. (1953) recommended an initial dose of 30 to 75 mg diphacinone followed by a maintenance dose of 5 to 30 mg or less per day. The most definitive study with diphacinone in humans was conducted by Katz *et al*. (1954), in which a single 30-mg dose was administered to 10 healthy individuals whose blood-clotting times were closely monitored. Dipaxin (diphacinone) had an effect on the prothrombin complex within 17 hours of administration, but clotting times did not reach therapeutically beneficial levels (defined as a prothrombin complex concentration below 30% of normal) in all individuals until 41 hours after dosing. The authors noted that a quicker response could be obtained by increasing the dose to 40 mg. They also used diphacinone to treat 60 patients with thromboembolic disease. Patients were initially administered 30 mg of diphacinone. Depending upon the individual's response to the first dose, subsequent daily doses ranged from 3 to 5 mg of diphacinone. No deaths have been reported for diphacinone during the time it was used as a human pharmaceutical and as a rodenticide.

Hazard to Humans from Drinking Water

Exposure estimates are based upon diphacinone concentrations calculated for water taken from 2 sources: Hanawi Stream at the point at which it is intercepted by the Koolau Irrigation Ditch (0.038 ppm) and at the Kamaole Weir Water Treatment Facility, where other streams entering the ditch have theoretically diluted the diphacinone concentration to 0.003 ppm. Toxicity estimates for humans are derived from a number of studies using laboratory rats (Table 4): single dose acute oral toxicity (Daniel 1993*a*, U.S. EPA 1998), multiple consecutive daily dose (Rogers 1994, U.S. EPA 1998), and developmental and maternal toxicity (Daniel 1993*b*, U.S. EPA 1998). The No Observed Effect Levels (NOELs) and Lowest Observed Effect Levels (LOELs) from these studies were used to predict the amount of water from the Hanawi Stream and at the Kamaole Weir WTF a 55-kg (121-lb) person would have to drink to ingest potentially toxic levels of diphacinone. This quantity was calculated by dividing the NOEL and LOEL by the estimated diphacinone concentrations at these 2 locations. The NOELs and LOELs were also compared with the amount of diphacinone recommended by Katz *et al*. (1954) for human patients (30 mg).

A 55-kg (121-lb) person would have to drink more than 188 liters of water from Hanawi Stream or more than 2,383 liters of water from the Kamaole Weir WTF in a single day to ingest an amount of diphacinone sufficient to trigger detectable changes in blood clotting as observed in laboratory rats (Table 4, Single Dose Acute Oral). If a 55-kg person were to drink water from these sources for multiple days, they would still have to consume more than 57 liters per day for multiple days from Hanawi Stream, or more than 733 liters of water per day for multiple days from the Kamaole Weir WTF, to ingest an amount of diphacinone sufficient to trigger detectable changes in blood clotting as observed in laboratory rats (Table 4, Multiple Consecutive Daily Dose). A 55-kg person would have to drink 789 liters from Hanawi Stream or 10,000 liters of water from the Kamaole Weir WTF in a single day to ingest a dose equivalent to that previously prescribed for human patients (30 mg) (Table 4, Human Dose).

The groups with the highest risk potential are pregnant women and infants. In a laboratory study that dosed pregnant rats for multiple consecutive days with diphacinone, the lowest dose tested, 0.01 mg/kg/day, caused vaginal bleeding (Daniel 1993*b*). A 55-kg woman would need to drink 14 liters of water a day from Hanawi Stream for multiple days and up to 183 liters from the Kamaole Wier for multiple days to ingest an equivalent dose (Table 4, Maternal Toxicity). Fetal resorptions were noted at 0.075 mg/kg/day, but not at 0.025 mg/kg/day (Daniel 1993*b*). To ingest the equivalent amounts, a pregnant woman would need to drink between 36 and 109 liters of water a day from Hanawi Stream for multiple days and between 460 and 1,377 liters of water a day from the Kamaole Wier for multiple days (Table 4, Developmental Toxicity). These consumption predictions for diphacinone-contaminated water can be compared to published U.S. drinking water surveys. The U.S. EPA (2000) assumes pregnant women ingest an average of 0.872 liters of liquids per day (95^{th} percentile: 2.59 liters per day). In the worst case scenario modeled in this assessment, a pregnant woman consuming 14 liters of Hanawi Stream water per day is consuming 5.4 times more water than women on the high end of the daily consumption estimates. In its analysis of the environmental health risks of anticoagulant rodenticides, the World Health Organization did not find evidence to conclude that any rodenticide other than warfarin caused birth defects (WHO 1995).

The hazard to children can be estimated in 2 ways. The most direct way is to simply perform the same calculations as used above, using an individual of lower body weight. A 10-kg (22-lb) child would have to drink more than 30.8 liters of Hanawi Stream water in one day or 9.5 liters per day for multiple consecutive days to ingest a dose equivalent to that shown to cause blood

Table 4. Assessment of human risk related to drinking water (liters) and food (kilograms) consumption; quantity required to equal the lowest toxicity endpoints reported for laboratory animals in single and multiple dose studies, and the recommended therapeutic dose when diphacinone was used medically for heart patients.

Exposure Type	Toxicity Endpoint	55 kg Person (mg/day)	Daily Consumption Requirements to Equal Toxic Endpoint				
			Untreated Surface Water (0.038 ppm[1])	Treated Surface Water (0.003 ppm[2])	Swine Muscle (0.251 ppm[3])	Swine Liver (3.07 ppm[4])	Pheasant Liver (0.56 ppm[5])
Single Dose Acute Oral[6]	NOEL (0.13 mg/kg)	7.15	188 L	2,383 L	28.49 kg	2.33 kg	12.77 kg
	LOEL (0.20 mg/kg)	11.0	289 L	3,667 L	43.83 kg	3.58 kg	19.64 kg
Multiple Consecutive Daily Dose[6]	NOEL (0.040 mg/kg/day)	2.20	57 L	733 L	8.77 kg	0.72 kg	3.93 kg
	LOEL (0.085 mg/kg/day)	4.67	122 L	1,557 L	18.64 kg	1.52 kg	8.35 kg
Developmental Toxicity[6]	NOEL (0.025 mg/kg)	1.38	36 L	460 L	5.50 kg	0.45 kg	2.46 kg
	LOEL (0.075 mg/kg)	4.13	109 L	1,377 L	16.45 kg	1.34 kg	7.37 kg
Maternal Toxicity[6]	NOEL (< 0.01 mg/kg/day)	<0.55	<14 L	<183 L	<2.19 kg	<0.17 kg	<0.98 kg
	LOEL (0.01 mg/kg/day)	0.55	14 L	183 L	2.19 kg	0.17 kg	0.98 kg
Human Dose[7]	Therapeutic Dose (30 mg)	30.0	789 L	10,000 L	119.52 kg	9.78 kg	53.57 kg

[1] Maximum residue predicted for untreated surface water calculated on mean surface flow (minus 1 Std. Dev.) from Hanawi Stream
[2] The 4-year average daily flow (minus 1 Std. Dev.) for the Kamaole Weir (90.28 Million Gallon/Day, 342.2 MG/Day)
[3] The highest diphacinone concentration detected in swine muscle (Pitt *et al.* 2005)
[4] The highest diphacinone concentration detected in swine liver (Pitt *et al.* 2005)
[5] The highest diphacinone concentration detected in pheasant liver (Hegdal 1985)
[6] NOEL and LOEL based on reported increased prothrombin time (PT) and activated partial thromboplastin time (aPTT) (Daniel 1993*a*, 1993*b*)
[7] Therapeutic dose based on 30 mg administered on the first day, subsequent doses of 2.5 to 5.0 mg/day (Katz *et al.* 1954)

clotting effects in laboratory rats. In areas where drinking water was obtained from the Kamaole Weir, a child would have to drink more than 390 liters of water in a single day or 120 liters of water per day for multiple days to ingest doses equivalent to that which caused clotting effects in laboratory rats. The U.S. EPA (2000) assumes a 10-kg child will drink one liter of water per day, so a child would have to drink more than 9.5 times the average daily water intake on multiple days to reach the lowest amount of water from Hanawi Stream estimated to affect blood clotting times.

One problem with the above calculations is that juveniles can be more sensitive to chemical exposure than adults; therefore, a simple ratio calculation, as presented above, underestimates the actual risk. The hazard to infants posed by exposure to diphacinone is greater than that for the general population because newborns are vitamin K deficient until approximately 12 weeks of life (American Academy of Pediatrics 1993). The 10-kg child modeled above would in all probability be closer to 1 year old and no longer vitamin K-deficient.

For younger nursing infants, risk can be evaluated if it is assumed that mammillary transfer of diphacinone is similar between cows and humans. Bullard *et al*. (1977) administered doses of 1.0 mg diphacinone/kg body weight and 2.75 mg/kg to lactating cows and measured diphacinone levels in their milk for 6 days. No residues were detected in milk from the lower-dosed cows. Diphacinone levels in the milk from the higher-dosed cows peaked at 0.021 ppm and no diphacinone was detected after 48 hours post-dosing. Their nursing calves showed no changes in blood clotting times. The U.S. EPA (2000) assumes lactating women ingest an average of 1.67 liters of liquids per day (95th percentile: 3.59 liters per day). If a 55-kg nursing mother drank 3.59 liters of water from Hanawi Stream (0.038 ppm diphacinone), she would ingest 0.136 mg of diphacinone or a dose of 0.0025 mg/kg. A dose of 0.0025 mg/kg is 400 times lower than the lowest dose (1.0 mg diphacinone/kg body weight) tested by Bullard *et al*. (1977) at which no diphacinone residues in cow's milk or negative effects in nursing calves were detected. Thus, the 3 exposure routes– direct consumption of contaminated stream water, consumption of formula mixed with diphacinone-contaminated water, or via mammillary transfer from an exposed mother– would not result in an infant ingesting amounts of diphacinone high enough to affect clotting times in surrogate animals.

Diphacinone Residues in Game Animals

Three studies have addressed acute dietary toxicity and the subsequent diphacinone residues in pig liver and muscle tissue under laboratory conditions (Keith *et al*. 1990, Fletcher 2002, Fisher 2006), and one study evaluated diphacinone residues in wild pigs in a Hawaiian forest treated with both an aerial application of 0.005% diphacinone bait and bait stations stocked with the same product (Pitt *et al*. 2005). Keith *et al*. (1990) dosed wild pigs captured on the island of Hawaii with technical diphacinone mixed in maintenance diet at an average dose of 0.6 mg/pig/day for 2 days (~0.007 mg/kg/day, low dose) and 1.5 mg/pig/day for 5 days (~0.018 mg/kg/day, high dose). Pigs were sacrificed either 2 or 10 days after the last exposure. The highest residues detected were 0.83 ppm in the livers of 2 high-dosed pigs, one sacrificed 2 days post exposure and the other at 10 days (Table 5). No quantifiable diphacinone was detected in muscle tissue. Fletcher (2002) dosed groups of 4 domestic pigs at rates of 0.133 and 0.333 mg diphacinone/kg body weight/day for 7 consecutive days Pigs were sacrificed either 2 or 10 days after the last exposure. The highest residues detected were 0.83 ppm in the livers of 2 high-dosed pigs, one sacrificed 2 days post exposure, and the other at 10 days (Table 5). No quantifiable diphacinone was detected in muscle tissue. Fletcher (2002) dosed groups of 4 domestic pigs at rates of 0.133 and 0.333 mg diphacinone/kg body weight/day for 7 consecutive days. In the low-dose group, only one

Table 5. Diphacinone concentrations in tissues of animals analyzed from laboratory or field studies conducted with 0.005% diphacinone rodenticide baits.

Species	Study Location	Method[1]	N	Mean (Range) Diphacinone Concentration (ppm)			Reference
				Liver	Muscle	Whole Body	
California ground squirrel	Rangeland	SB	7	-	-	1.4 (0.62 - 3.44)	Baroch 1994*a*[2]
		BS	10	-	-	0.91 (0.50 - 1.89)	Baroch 1994*b*[2]
		SB	8	-	-	0.23 (0.04 - 0.50)	Salmon *et al.* 2002
		HB	16	-	-	0.31 (0.04 - 0.81)	Salmon *et al.* 2002
Black rat	Hawaiian forest	HB	8	4.6 (1.5 - 12.0)	-	-	Spurr *et al.* 2003*b*
		AB	7	4.4 (<MLOD - 12.0)	-	-	Spurr *et al.* 2003*a*
House mouse	Hawaiian forest	HB	2	2.07 (1.75 - 2.39)	-	-	Spurr *et al.* 2003*b*
		AB	2	2.3 (2.1 - 2.4)	-	-	Spurr *et al.* 2003*a*
Mongoose	Hawaiian forest	HB	1	1.35	-	-	Spurr *et al.* 2003*b*
Peromyscus spp.	Orchard	HB	2	1.41 (0.92 - 1.9)	-	-	Hegdal 1985
Microtus spp.	Orchard	HB	1	0.77	-	-	Hegdal 1985
Coyote	Orchard	HB	1	1.2	-	-	Hegdal 1985
House cat	Orchard	HB	1	0.64	-	-	Hegdal 1985
Pig	Hawaiian forest	AB, BS	18	0.83 (<MLOD - 3.07)	0.06 (<MLOD - 0.25)	-	Pitt *et al.* 2005
	Laboratory	0.007 mg/kg/d	4	<MLOD	<MLOD	-	Keith *et al.* 1990
	Laboratory	0.018 mg/kg/d	4	0.42 (<MLOD - 0.83)	<MLOD	-	Keith *et al.* 1990
	Laboratory	0.13 mg/kg/d	4	0.05 (0.04 - 0.07)	<0.001 (<MLOD - 0.004)	-	Fletcher 2002[3]
	Laboratory	0.33 mg/kg/d	4	0.04 (0.03 - 0.06)	<MLOD	-	Fletcher 2002[3]
	Laboratory	12.5 mg/kg	4	2.83 (2.45 - 3.22)	0.31 (0.22 - 0.37)	-	Fisher 2006[4]
	Laboratory	12.5 mg/kg	3	0.33 (0.18 - 0.41)	<MLOD	-	Fisher 2006[5]
Kalij pheasent	Hawaiian forest	AB	2	0.15 (0.12 - 0.18)	-	-	Spurr *et al.* 2003*a*
		HB	5	0.02 (<MLOD - 0.09)	-	-	Spurr *et al.* 2003*b*
Red-billed leothrix	Hawaiian forest	AB	8	2.45 (0.74 - 4.90)	-	-	Spurr *et al.* 2003*a*
		HB	6	0.28 (<MLOD - 0.70)	-	-	Spurr *et al.* 2003*b*
Northern cardinal	Hawaiian forest	AB	2	0.11 (0.08 - 0.13)	-	-	Spurr *et al.* 2003*a*
		HB	6	0.07 (<MLOD - 0.39)	-	-	Spurr *et al.* 2003*b*
Japanese white-eye	Hawaiian forest	AB	5	<MLOD	-	-	Spurr *et al.* 2003*a*
		HB	10	<MLOD	-	-	Spurr *et al.* 2003*b*
Ring-necked pheasant	Orchard	MB	44	0.23 (<MLOD - 0.51)	-	-	Hegdal 1985
California quail	Orchard	MB	19	0.21 (<MLOD - 0.56)	-	-	Hegdal 1985
Chukar	Orchard	MB	15	0.28 (<MLOD - 4.2)	-	-	Hegdal 1985
Hawaiian owl	Hawaiian forest	AB	1	0.62	0.08	-	Pitt *et al.* 2005
Deroceras laeve (slug)	Laboratory	*Ad libitum*	37	-	-	2.64 (1.63 - 5.01)	Johnston *et al.* 2005
	Hawaiian forest	AB	3	-	-	0.23 (0.21 - 0.25)	Johnston *et al.* 2005
Limax maximus (slug)	Laboratory	*Ad libitum*	19	-	-	0.81 (<MLOD - 2.26)	Johnston *et al.* 2005
	Hawaiian forest	AB	3	-	-	0.61 (0.60 - 0.61)	Johnston *et al.* 2005
Oxychilus spp. (snail)	Laboratory	*Ad libitum*	15	-	-	1.77 (1.06 - 2.91)	Johnston *et al.* 2005
	Hawaiian forest	AB	3	-	-	0.69 (0.59 - 0.79)	Johnston *et al.* 2005
Coconut crab (*Birgus latro*)	Laboratory	*Ad libitum*	12	0.02 (<MLOD - 0.35)	0.048 (0.01 - 0.14)	-	Tanner *et al.* 2004

[1] SB – Spot Baiting, BS – Bait Stations, HB – Hand Broadcast, AB – Aerial Broadcast, MB – Mechanical Broadcast

[2] More detailed information from this study is found in Salmon *et al.* 2002

[3] 14 days post exposure

[4] 1 day post exposure

[5] 15 days post exposure

pig had detectable residues in the muscle (0.004 ppm), while all 4 pigs had detectable diphacinone residues in the liver, ranging from 0.04 ppm to 0.07 ppm with a mean concentration of 0.05 ppm. Residue analysis of pigs in the high-dose group showed no detectable residues in the muscle. All 4 pigs had detectable diphacinone residues in the liver, ranging from 0.03 ppm to 0.06 ppm with a mean concentration of 0.04 ppm (Table 5).

In 2003, an experimental aerial application, supplemented with bait stations in some areas, was made with a 0.005% diphacinone bait in a native forest on the island of Hawaii. Problems were encountered during aerial application that resulted in uneven bait distribution, in some instances resulting in extremely high densities of pellets (Tim Ohashi, Wildlife Services, Hilo, HI, pers. commun.). Pitt *et al*. (2005) evaluated tissue diphacinone residues in 18 wild pigs inhabiting the treatment areas. The mean diphacinone residue detected in the liver of the pigs was 0.83 ppm and ranged from 0 to 3.07 ppm. The mean diphacinone residue detected in the muscle of the pigs was 0.006 ppm and ranged from 0 to 0.25 ppm. Numerous problems with the trial resulted in pigs consuming amounts of bait considerably higher than would be expected to occur during a correctly conducted aerial broadcast, so this misapplication serves as a worst-case scenario for exposure of wild pigs.

Fisher (2006) conducted a study with 12 domestic pigs to evaluate the persistence of diphacinone residues following a single exposure to 12.5 mg/kg. Fisher reported a biphasic degradation curve in the liver with an initial phase half-life of 1.30 days (days 1 to 4) and a terminal phase half-life of 14.12 days (days 4 to 15). The overall diphacinone half-life in the liver was 5.43 days. The average diphacinone residue in the liver was below 1 ppm 4 days after exposure. Fisher also calculated diphacinone half-lives of 4.48 and 2.29 days for muscle and fat,

respectively. The highest residues detected were 3.22 mg/kg in the liver, 0.37 mg/kg in muscle, and 0.38 mg/kg in fat, all one day after dosing.

Two studies evaluated diphacinone residues in game birds captured from sites in Hawaii that had been treated by hand or aerial broadcasting 0.005% diphacinone bait (Table 5). The first study utilized hand broadcast techniques on a 10-acre treatment area (Spurr *et al*. 2003*b*). Five Kalij pheasants (*Lophura leucomelana*) were collected within the treatment area between 2 and 6 weeks after treatment. Of the 5, only one contained detectable diphacinone residues. The liver of this bird contained 0.09 ppm diphacinone. The second study was an aerial broadcast trial in support of the proposed aerial broadcast registration of Ramik® Green (Spurr *et al*. 2003*a*). Two Kalij pheasants were collected within the 112 acre treatment area one month after treatment. Diphacinone residues of 0.12 and 0.18 ppm were found in the livers of these birds.

Hegdal (1985) evaluated the impact of baiting voles (*Microtus* spp.) in orchards in Washington State with hand-broadcast 0.005% diphacinone bait (2 treatments of 12.9 kg/ha, or 11.5 lbs/acre, each, 20 - 30 days apart) on ring-necked pheasant (*Phasianus colchicus*), California quail (*Callipepla californica*), and chukar partridge (*Alectoris chukar*). All 3 species showed evidence of bait consumption (bait in the gastrointestinal tract or diphacinone residues in tissues), but the chukar was significantly less prone to exposure. Of the 15 chuckars analyzed, only one had detectable diphacinone residues. Five out of 44 ring-necked pheasants contained bait pellets in the crop. Ten pheasants contained diphacinone residues in their liver, with an average concentration of 0.23 ppm. Nineteen California quail were also collected, of which 10 contained bait pellets in the crop. Eleven of the 19 livers analyzed contained diphacinone residues. The average concentration was 0.21 ppm with a range from below the Method Limit of Detection (MLOD) to 0.56 ppm (Table 5). Three conditions in this study likely resulted in birds consuming amounts of bait considerably higher than what might be encountered during an aerial application in Hawaii using the proposed registration. First, adjoining landowners applied bait at the same time as the study, and Hegdal reported applications were likely as high as 45 kg/ha (40 lbs/acre). Second, being brown, Ramik® Brown (2 g) may closely resemble the diet the birds were fed in captivity and might be more likely foraged upon than the proposed bait because the proposed bait is large (6.5 g) and colored bright green. Finally, Hegdal utilized a combination of wild and pen-raised birds. The pen-raised birds may have been more accustomed to foraging on a pelleted diet, resulting in the pen-raised birds accumulating residues higher than wild birds. Therefore, the highest liver value reported from Hegdal, 0.56 ppm, will be used as a worst-case scenario for game bird exposure.

Hazard to Humans from Game Meat

Exposure estimates are calculated using the same toxicity studies and methods as described under the drinking water section. Residue values in pig muscle (0.251 ppm) and liver (3.07 ppm) are taken from Pitt *et al*. (2005). The residue values for game bird liver (0.56 ppm) are taken from Hegdal (1985). These are the highest residue values reported for these species; the exposures occurred under unusual circumstances and thus the values are representative of worst-case scenarios. Risk calculations are also based on the assumption that absorption efficiency of the gastrointestinal system is equivalent for both technical diphacinone administered orally by intubation, and diphacinone administered in association with food. Since it is not known if cooking reduces the anticoagulant effect of diphacinone (heating frequently causes the decomposition of compounds), the calculations assume that both liver and muscle are consumed raw.

At the maximum diphacinone concentrations detected in pig muscle, pig liver, and game bird liver, a 55-kg person would need to eat more than 28.49 kg (62.81 lbs), 2.33 kg (5.14 lbs), or 12.77 kg (28.15 lbs), respectively, in a single day to ingest a diphacinone dose shown to cause detectable changes in blood clotting in laboratory rats (Table 4, Single Dose Acute Oral). If these same tissues were consumed over multiple consecutive days, these quantities would drop to 8.77 kg (19.3 lbs), 0.72 kg (1.6 lbs), or 3.93 kg (8.6 lbs) per day, respectively (Table 4, Multiple Consecutive Daily Dose).

Based on the maximum residue concentrations detected in pig muscle, pig liver, and game bird liver, a 55-kg pregnant woman would have to eat more than 5.50 kg (12.13 lbs), 0.45 kg (0.99 lb), or 2.46 kg (5.42 lbs) a day, respectively, for multiple days to ingest an amount of diphacinone equivalent to the dose shown to cause fetal reabsorption in rats (Table 4, Developmental Toxicity). The lowest dose tested in rats caused maternal bleeding. This dose would be equivalent to a pregnant woman consuming less than 2.19 kg (4.83 lbs), 0.17 kg (0.37 lb), or 0.98 kg (2.16 lbs) of contaminated pig muscle, pig liver, or game bird liver, respectively, daily for multiple days (Table 4, Maternal Toxicity).

Similarly, at the maximum diphacinone concentrations detected in pig muscle, pig liver, and game bird liver, a 10-kg child would need to eat more than 5.18 kg (11.42 lbs), 0.42 kg (0.93 lb), or 2.32 kg (5.11 lbs), respectively, in a single day to ingest a diphacinone dose shown to cause detectable changes in blood clotting in laboratory rats. If these same tissues were consumed over multiple consecutive days, these quantities would drop to more than 1.59 kg (3.51 lbs), 0.13 kg (0.29 lb), or 0.62 kg (1.37 lbs), respectively.

For younger nursing infants, the indirect route of exposure to contaminated game meat through the mother's breast milk can be evaluated. If a 55-kg nursing mother ate an entire pound (0.454 kg) of pig liver with a concentration of 3.07 mg diphacinone/kg liver (the highest value reported in pig liver), her dose would be 0.025 mg/kg. This dose is 40 times lower than the lowest dose tested by Bullard *et al*. (1977) at which no diphacinone residues were detected in the cows' milk and no negative effects in nursing calves were observed.

Risk to people eating meat from birds and pigs harvested within treatment areas is very low. Of the categories evaluated, pregnant women appear to have the lowest threshold for symptoms of exposure, based on

laboratory studies with rats. Of the tissue types evaluated, liver had the highest residues, consistent with how diphacinone is metabolized. Thus, the exposure scenario presenting the highest risk for humans would be a pregnant woman eating less than 0.17 kg (0.37 lb) of pig liver per day for multiple days. A more likely exposure scenario would be for a pregnant woman to eat muscle tissue, for which the amount possibly causing symptoms such as vaginal bleeding would rise to less than 2.19 kg, or almost 5 pounds of pork meat per day for multiple days. Worst-case exposure scenarios for a non-pregnant woman require a daily dose of more than 0.72 kg (1.58 lbs) of pig liver or 8.77 kg (19.29 lbs) of pork meat for multiple days to possibly affect her blood clotting times, using laboratory rat data. Using the standard dosage prescribed to human patients (30 mg), these amounts increase to 9.78 kg (21.52 lbs) of pig liver or 119.52 kg (262.94 lbs) of pork meat to possibly affect the blood clotting times of a non-pregnant woman (Table 4, Human Dose). Because diphacinone is more toxic when consumed over a number of days, one-time doses are considerably higher. Although consumption rates of 0.17 kg (0.37 lb) and 0.72 kg (1.58 lbs) per day of pig liver for multiple days are possible, they are not likely. However, since the laboratory trial with pregnant rats did not test a dosage low enough for no effects to be observed, and since the effects of diphacinone on pregnant human females are not known, this evaluation only presents a rough estimate of risk.

Fisher's (2006) diphacinone metabolism data, Pitt *et al*.'s (2005) residue data, and bait disappearance data from Dunlevy *et al*. (2000) and Spurr *et al*. (2003*a*,*b*) provide some insight as to the length of time diphacinone might be in pig tissues at levels that would be of concern for human consumption. The dose Fisher used, 12.5 mg/kg, was based on a pig consuming the contents of 5 bait stations containing a total of 10 kg (22 lbs) of 50 ppm diphacinone bait in 1 day, and represents an extreme exposure scenario. The highest diphacinone residue reported in Fisher's study for liver 1 day after exposure was only slightly higher than the highest residue reported in free-ranging pigs by Pitt *et al*. (2005). Fisher's work showed that within 2 days of exposure, diphacinone residues in the liver declined by one-half. After 4 days, as residues neared 1 ppm, degradation slowed significantly to a half-life of approximately 2 weeks. Dunlevy *et al*. (2000) demonstrated that at an application rate of 22.5 kg/ha (20 lbs/acre), bait disappears rapidly, but can still be on the ground approximately 2 weeks after application. He reported 50% and 20% of the bait remained on days 6 and 12, respectively. Spurr *et al*. (2003*a*) reported wide variation in bait disappearance rates in hand broadcast trials (2 treatments of 11.2 kg/ha, or 10 lbs/acre, 4 - 6 days apart), ranging from no bait remaining after 7 days to 50% remaining after 14 days and 15% after 24 days. Twenty-five percent of monitored bait pellets remained 2 weeks after each aerial broadcast application (11.83 kg/ha, or 10.51 lbs/acre, and 10.56 kg/ha, or 9.39 lbs/acre 5 days apart) (Spurr *et al.* 2003*a*). Consequently, pigs could be repeatedly exposed to diphacinone during the weeks following an aerial application of bait, but the risk of exposure and level of exposure would decrease rapidly with time. This factor, combined with the rapid clearance of diphacinone residues from pig tissue, reduces the risk of human exposure to contaminated pig meat significantly. Only 2 of the 18 pigs collected by Pitt *et al*. (2005) after a misapplication of bait contained diphacinone residues greater than 1 ppm in the liver, and only 5 of the 18 pigs contained diphacinone residues in the muscle (0.10 - 0.25 ppm). A pig collected 18 days after the bait application began contained the highest diphacinone residues and had bait in its gastrointestinal tract, indicating it had recently eaten bait.

Thus, while exposure can occur as long as bait is present, the previously discussed field trials demonstrate that during a correctly conducted broadcast application, the amount of bait accessible to pigs should decline within a few weeks to levels insufficient to cause detectible residues in pig tissue. Once bait is gone or a pig stops foraging on bait, residues in all pig tissues drop by half within 2 to 4 days. Some pigs can have residue levels in the liver above 3 ppm immediately after exposure, but most pigs that have been analyzed have contained diphacinone residues less than 1 ppm, making the risk of exposure to levels high enough to cause deleterious effects in humans even less likely. The highest residue recorded in pig muscle was 0.37 ppm (Fisher 2006), a level that indicated little hazard to most individuals. With a half-life of 4.5 days, the risk from eating diphacinone-contaminated pig meat decreases even further.

Hazard to Wildlife

The acute oral toxicity of technical diphacinone has been tested in at least 12 species of mammals and 2 species of birds (Table 2). Different species exhibit wide variation in their susceptibility to diphacinone. The acute oral toxicity of technical diphacinone to rats (*Rattus* spp.) is generally reported as less than 7.0 mg/kg but has been reported as high as 43.3 mg/kg. Mice (*Mus* spp.) appear to be more tolerant of technical diphacinone, with LD_{50}s between 28.0 - 340 mg/kg. Mongoose (*Herpestes auropunctatus*) and vampire bats (*Desmodus rotundus*) are among the most sensitive species known, with LD_{50}s lower than 1 mg/kg. Cows (*Bovis* spp.) tolerated a dose of 5 mg/kg with no effects. The LD_{50} for the cat (*Felis catus*) is reported as 15 mg/kg. Canine species are sensitive to diphacinone, with the LD_{50}s for the domestic dog (*Canis domesticus*) around 2-3 mg/kg and coyotes (*Canis latrans*) reported as low as 0.6 mg/kg. One study with pigs (*Sus* sp.) reported an LD_{50} of greater than 150 mg/kg. Birds are much more tolerant of diphacinone, with the LD_{50} and the dietary LC_{50} for the mallard (*Anas platyrhynchos*) 3,160 and 906 mg/kg, respectively, and the LD_{50} and the dietary LC_{50} for the northern bobwhite (*Colinus virginianus*) greater than 400 mg/kg but less than 2,000 mg/kg, and >5,000 mg/kg, respectively.

The risks estimated for native and nonnative birds and mammals found in Hawaiian ecosystems are summarized in Table 6 for mammals and Table 7 for birds. Each table is divided into the risks from a single, acute exposure, and from multiple exposures producing sublethal effects. Acute exposure is presented as the amounts of 50 ppm diphacinone bait and invertebrate or animal tissue

Table 6. Acute and sublethal dietary risk to mammals from primary exposure to Ramik® Green pellets or through secondary exposure from eating invertebrates or rodents exposed to diphacinone bait. (Calculated values are the grams of food required to be ingested to equal the lowest reported mammalian LD_{50}, NOEL, and LOEL.)

Acute Exposure				
Species	**Weight (kg)**	**LD_{50} (mg/kg)**	**Primary Exposure (grams in one day)[1]**	**Secondary Exposure (grams in one day)[2]**
Dog	15.00	0.6[3]	180	2,932
Cat	3.60	15	1,080	17,590
Bat	0.017	0.91[4]	na	3.09[5]
Swine	50.0	>150	>150,000	>2,442,997

Sublethal Exposure							
Species	**Weight (kg)**	**Sublethal Toxicity (mg/kg/day)[6]**		**Primary Exposure (g/day)[1]**		**Secondary Exposure (g/day)[2]**	
		NOEL	**LOEL**	**NOEL**	**LOEL**	**NOEL**	**LOEL**
Dog	15.0	0.040	0.085	12.00	25.40	195.44	415.31
Cat	3.60	0.040	0.085	2.88	6.12	46.91	99.67
Bat	0.017	0.040	0.085	na	na	0.14[5]	0.29[5]
Swine	50.0	0.040	0.085	40	85.0	651.47	1384.36

[1] Based on a diphacinone concentration in Ramik® Green of 50 mg/kg
[2] Based on the highest diphacinone residue found in pig liver (3.07 mg/kg, Pitt *et al.* 2005)
[3] Based on the LD_{50} for the coyote (Savarie *et al.* 1979)
[4] Based on the LD_{50} for the vampire bat (Thompson *et al.* 1972)
[5] Based on the highest diphacinone residue detected in mollusks (5.01 mg/kg, Johnston *et al.* 2005)
[6] Based on the NOEL and LOEL observed in rats (Rogers 1994)

Table 7. Acute and sublethal dietary risk to birds from primary exposure to Ramik® Green pellets or through secondary exposure from eating contaminated invertebrates or rodents. (Calculated values are the grams of food required to be ingested to equal the lowest reported avian LD_{50}, Lowest Lethal Dose - LLD, and LOEL.)

Acute Exposure							
Species	**Weight (kg)**	**LD_{50} (mg/kg)[1]**	**LD_{50} Primary Exposure (g)[2]**	**LD_{50} Secondary Exposure (g)[3]**	**Lowest Lethal Dose (LLD) (mg/kg/day)[4]**	**LLD Primary Exposure (g)[2]**	**LLD Secondary Exposure (g)[3]**
Game bird	1.00	>400	>8,000	>79,840	0.6	12.00	119.76
Non-game bird	0.03	>400	>240	>2,395	0.6	0.36	3.59
Hawaiian goose	1.50	>400	>12,000	>119,760	0.6	18.0	179.64
Hawaiian hawk	0.45	>400	na	>58,632[5]	0.6	na	87.95[5]
Hawaiian crow	0.500	>400	>4,000	>39,920	0.6	6.00	59.88
Hawaiian owl	0.350	>400	na	>45,603[5]	0.6	na	68.40[5]

Sublethal Exposure				
Species	**Weight (kg)**	**LOEL Sublethal Toxicity (mg/kg/day)[6]**	**Primary Exposure (g/day)[2]**	**Secondary Exposure (g/day)[3]**
Game bird	1.00	0.11	2.20	21.96
Non-game bird	0.03	0.11	0.07	0.65
Hawaiian goose	1.50	0.11	3.30	32.93
Hawaiian hawk	0.45	0.11	na	16.12[5]
Hawaiian crow	0.500	0.11	1.10	10.98
Hawaiian owl	0.350	0.11	na	12.54[5]

[1] Based on the northern bobwhite LD_{50} (Campbell *et al.* 1991)
[2] Based on the diphacinone concentration in Ramik® Green (50 ppm)
[3] Based on the highest diphacinone residue found in mollusks (5.01 ppm, Johnston *et al.* 2005)
[4] Based on the Lowest Lethal Dose in the mallard acute dietary toxicity study (Long *et al.* 1992*b*)
[5] Based on the highest diphacinone residues found in pig liver (3.07 ppm, Pitt *et al.* 2005)
[6] Based on the LOEL observed in golden eagles (Savarie *et al.* 1979). An NOEL was not determined by Savarie *et al.*

equivalent to the LD_{50}s from the studies listed in Table 2. Thresholds for sublethal exposure are evaluated using the lowest values observed to cause clotting effects in laboratory rats (Rogers 1994, U.S. EPA 1998) for mammals, or in golden eagles (Savarie *et al*. 1979) for birds. Secondary poisoning hazards were evaluated using the highest diphacinone residue values found in vertebrate (3.07 ppm for pig liver, Pitt *et al*. 2005) and invertebrate (5.01 ppm for mollusks, Johnston *et al*. 2005) tissue from Hawaii-based studies.

Hazard to Wildlife - Mammals

Acute and Sublethal Hazards to Pigs

Feral pigs are found in many native ecosystems in Hawaii. Primary and secondary hazard calculations for acute toxicity presented in Table 6 show that a 50-kg (110-lb) pig would need to consume 150 kg (330 lbs) of diphacinone bait and 2,443 kg (5,375 lbs) of contaminated animal tissue at 3.07 mg/kg to ingest a dose equivalent to the LD_{50} for pigs. To potentially experience sublethal blood clotting effects as observed in laboratory rats, a 50-kg pig would need to eat between 40 g (~6 bait pellets) and 85 g (~13 bait pellets) of diphacinone bait, and between 651 g (1.43 lbs) and 1,384 g (3.05 lbs) of animal tissue containing 3.07 mg/kg diphacinone.

The pellets proposed for registration are bright green, fish-flavored, and contain a sweetener. If pigs found the flavor desirable, it is likely they would actively search for the bait. A pig could reasonably find and eat the sublethal dose of 6 to 13 pellets a day calculated above using the exposure thresholds observed for laboratory rats. This quantity is approximately 3.3× to 1.5× lower than the lowest dose tested (0.133 mg/kg/day) on domestic pigs in the laboratory by Fletcher (2002). At this dose, Fletcher reported no signs of illness or mortality in domestic pigs. At a dose of 0.333 mg/kg/day or 51 pellets per day, Fletcher reported anticoagulant-related symptoms, but no mortality. Fletcher did not assess blood clotting effects in his study. It is possible that in the wild, where environmental conditions are not as favorable and the risk of accidental injury is higher, the pigs may have died from this level of exposure. Pitt *et al*. (2005) reported pig mortality when Eaton's Bait Pellet Rodenticide with Fish Flavorizer® was aerially applied. Wild pigs in the area were able to find sufficient quantities of bait pellets to consume lethal doses of diphacinone. However, a number of unusual circumstances, including bait spillage and uneven broadcast rates, likely facilitated the consumption of large amounts of bait by individual pigs.

The aerial broadcast application rate proposed for registration, 14 kg/ha (12.5 lbs/acre), would result in a pellet density of one per 4.6 m^2 on the ground. At this density, 13 pellets would cover an area of 59.8 m^2. If a pig developed a search pattern for bait pellets, this is not an unreasonably large area. Under actual aerial application conditions, it is expected that some pellets will become lodged in the canopy of trees, tree ferns, shrubs, or in cracks in the lava soils, out of the reach of foraging pigs. This would increase the search area required for a pig to consume doses equivalent to estimated lethal and sublethal levels.

Acute and Sublethal Hazard to the Hawaiian Hoary Bat

The endangered Hawaiian hoary bat feeds on moths and other flying insects (Jacobs 1999). The probability that bats will consume diphacinone bait is very low and is not addressed in this assessment. However, invertebrates feeding on diphacinone pellets could pose a threat of secondary exposure to the hoary bat. Secondary risk can be evaluated using maximum residue data collected on exposed slugs and snails (Johnston *et al*. 2005) and toxicity data for vampire bats (Thompson *et al*. 1972). Consumption estimates presented in Table 6 indicate a 17-g bat would have to eat 3.09 g of invertebrates (18% of its body weight) containing 5.01 mg/kg of diphacinone to ingest a dose equivalent to the vampire bat LD_{50}. A dose potentially resulting in sublethal effects as observed in laboratory rats could occur if bats ate between 0.14 g and 0.29 g of exposed invertebrates.

A Hawaiian hoary bat could ingest sufficient quantities of contaminated flying insects in a single night's foraging to receive sublethal and lethal doses of diphacinone, according to these calculations. However, this assumes that the Hawaiian hoary bat is as sensitive to diphacinone as the vampire bat. The vampire bat feeds on blood and therefore may be physiologically more susceptible to an anticoagulant. This risk assessment also assumes that the larvae of flying insects would retain a high concentration of diphacinone in their tissues through metamorphosis. If even only one of these assumptions was invalid, the risk to the Hawaiian hoary bat would be reduced substantially. No bats were found dead during any of the broadcast trials conducted in Hawaii (Spurr *et al*. 2003*a*,*b*; Pitt *et al*. 2005).

Hazards to Wildlife - Birds

Unfortunately, the toxicity data available for birds is very limited compared to mammals. The lack of data makes accurate predictions of risk to birds difficult. Standard U.S. EPA methods of estimating risk show that there is low risk for avian species from diphacinone (U.S. EPA 1998). However, the extremely low numbers of some species of Hawaiian birds warrant a more detailed examination of the available data for specific groups and species. Thus, the first paragraph under each of the following subsections evaluating risk to particular groups or species of birds includes 3 sets of calculations from the least to most conservative:

1) **LD_{50}** – the amount that can be expected to cause death in half of the individuals exposed; the EPA's standard method, which uses the lowest LD_{50} value from an acute toxicity study for any avian species (northern bobwhite, >400 mg/kg);
2) **LLD** – the lowest lethal dose in an avian study (0.6 mg/kg/day in the mallard, Long *et al*. 1992*b*);
3) **LOEL** – the lowest observed effects level from Savarie *et al*.'s (1979) study in which muscle tissue from diphacinone-dosed sheep was fed to golden eagles (*Aquila chrysaetos*) for 5- or 10-day periods. The dose values at which increased clotting times occurred in the eagles (0.11 mg/kg/day) were then converted into the amount of 0.005% diphacinone bait, and invertebrate or

animal tissue that would be required per day to equal that dose. No mortality occurred in the eagles, so no lethal value for chronic exposure was estimated in this study.

As noted above, the least conservative method of assessing avian toxicity, the lowest LD_{50}, is used by the EPA to determine the risk to non-target avian species from pesticides. The additional 2 methods of estimating risk presented below allow a more careful evaluation of the risks to individuals, but may be overly conservative.

Acute and Sublethal Hazards to Game Birds

A variety of gallinaceous game birds have been introduced to Hawaii. Populations are well-established and many are hunted for sport. Primary and secondary hazard calculations for acute oral toxicity (LD_{50}) for game birds presented in Table 7 show that a 1-kg (2.2-lb) bird would have to consume 8.00 kg (17.64 lbs) of bait or more than 79.84 kg (176.02 lbs) of invertebrates at 5.01 mg/kg diphacinone in one day to ingest a dose equivalent to the LD_{50}. Since these quantities are well above the bird's body weight, this risk is extremely low. However, for a dose equivalent to the lowest dietary dose which caused mortality (LLD) in a bird (0.6 mg/kg/day, Long *et al*. 1992*b*), a 1-kg (2.2-lb) bird would need to consume 12.00 g (0.03 lb) of bait or 119.76 g (0.26 lb) of invertebrates per day for multiple days. Hazard calculations for sublethal exposure (LOEL) presented in Table 7 show that a 1-kg bird would only need to eat 2.20 g (<0.01 lb) of bait (⅓ of a pellet) and 21.96 g (0.05 lb) of invertebrates per day for multiple days to ingest a dose equivalent to that observed to cause blood clotting effects in the Savarie *et al*. (1979) study.

The secondary hazard calculations are based on the maximum residue (5.01 mg/kg) detected in slugs and snails intentionally exposed to diphacinone bait under laboratory conditions, and may substantially overestimate diphacinone levels for invertebrates feeding on bait in the field. Johnston *et al*. (2005) also collected snails from a Hawaiian forest that had been aerially baited. The highest and average residues detected in field-collected invertebrates were 0.79 mg/kg and 0.69 mg/kg, respectively. A 1-kg bird would have to consume 870 g (1.91 lbs) of invertebrates per day for multiple days containing the average residue of 0.69 mg/kg for a dose equivalent to the lowest dietary dose which caused mortality (LLD) in a bird (0.6 mg/kg/day, Long *et al*. 1992*b*), and 159 g (0.35 lb) of invertebrates (16% of its body weight) containing the average residue of 0.69 mg/kg a day for multiple days to ingest a dose which could cause blood clotting abnormalities (LOEL). Therefore, the risk is likely to be significantly lower under actual field conditions.

Field evidence supports the premise that gamebirds will be exposed to diphacinone bait following broadcast application. Spurr *et al*. (2003*a*,*b*) reported Kalij pheasants eating bait pellets. In addition, Kalij pheasants collected from broadcast baited forests contained diphacinone residues in their livers. Dunlevy and Campbell's (unpubl. data*)* study of non-target risk associated with broadcast baiting reported taking nearly 21,000 still pictures of vertebrates near placebo bait pellets using motion-triggered cameras. Despite the fact that some of Dunley and Campbell's study sites were the same as Spurr *et al*.'s (2003*a*,*b*), only one image was of a gallinaceous bird, the Erckel's francolin (*Francolinus erckelii*). In the picture, the francolin did not appear to be consuming the bait. However, Spurr *et al*.'s (2003*a*,*b*) data clearly show pheasants will consume either bait or other food items containing diphacinone residues. Yet despite the potential for exposure, no game bird carcasses have been discovered during carcass searches in any of the diphacinone field studies conducted in Hawaii. These data suggest game birds would be at risk of sublethal exposure following broadcast baiting of diphacinone bait. However, the rate at which exposure actually occurs may be low, given the data presented by Dunlevy and Campbell (unpubl. data).

Acute and Sublethal Hazards to Non-Game Birds

'Non-game birds' is meant to include both introduced and native avian species weighing 100 g (0.22 lb) or less. Hawaii has a unique assemblage of native forest passerine species, 19 of which are federally listed as endangered (U.S. FWS 2006). In addition, numerous introduced species have become naturalized in the Hawaiian Islands. This hazard assessment is based primarily on body weight. Consequently, the quantitative results can be applied to a myriad of different sized birds, both introduced and native.

Primary and secondary hazard calculations for acute oral toxicity (LD_{50}) for non-game birds presented in Table 7 show that a 30-g (0.07-lb) bird would have to consume at least 240 g (0.53 lb) of bait or 2,395 g (5.27 lbs) of invertebrates in one day to ingest a dose equivalent to the LD_{50} for the northern bobwhite. Since these quantities are well above the bird's body weight, the risk when calculated this way is extremely low. However, for a dose equivalent to the lowest dietary dose which caused mortality (LLD) in a bird (0.6 mg/kg/day, Long *et al*. 1992*b*), a 30-g bird would only need to consume 0.36 g of bait or 3.59 g of invertebrates; however, this dose would have to be consumed daily over multiple days. Hazard calculations for sublethal exposure (LOEL) presented in Table 7 show that a 30-g bird would only need to eat 0.07 g of bait (a 100^{th} of a bait pellet or 0.2% of its body weight) or 0.65 g of invertebrates per day for multiple days to ingest a dose that resulted in blood clotting effects in golden eagles. Both of these scenarios are well within the reasonable range of daily food consumption rates of a 30-g bird. Therefore, these birds could be at risk of suffering sublethal or even lethal effects through both primary and secondary exposure if they forage on bait or contaminated invertebrates.

Dunlevy and Campbell (unpubl. data) documented non-target species' use of bait pellets and species in the vicinity of bait pellets. During the 76,800 hours (21,217 pictures) of motion-activated still photography-monitored bait pellets, no native birds were photographed, despite the fact that avian surveys recorded 10 species of native birds in the study sites. Nonnative birds were documented in 100 photographs (0.47%) of vertebrates. In 28 pictures, nonnative species were photographed interacting with bait pellets. During this same time, avian surveys documented 12 species of nonnative birds in the study

sites. The red-billed leiothrix (*Leiothrix lutea*) was recorded in 98 of the photographs. The Japanese bush warbler (*Cettia diphone*) and the Erkel's francolin were each recorded in one picture. Ramik® Green bait was consumed in 9 of the 28 events. Only the red-billed leiothrix was observed eating bait. The diphacinone bait broadcast studies by Spurr *et al*. (2003*a*,*b*) were conducted in the same area as one of the sites used by Dunlevy and Campbell. Spurr *et al*. collected nonnative birds from the study sites and analyzed them for diphacinone residues. Eleven of 14 red-billed leiothrix and 3 of 8 northern cardinals (*Cardinalis cardinalis*) collected contained diphacinone residues. None of the 9 Japanese white-eyes (*Zosterops japonicus*) collected contained diphacinone residues (Table 4). Pitt *et al*. (2005) also used motion-activated cameras to monitor bait pellets during a trial aerial broadcast. Over a 7-day period, 250 images were taken. Of these, 183 (73.2%) were of bait only, 60 (24%) were of rodents, 5 (2.0%) were of mongooses, and 2 (0.8%) were of red-billed leiothrix. These studies indicate at least 2 species of introduced non-game birds (red-billed leiothrix and northern cardinal) are being exposed to diphacinone during broadcast baiting operations.

Of the 19 species of endangered forest birds, the po′ouli (*Melamprosops phaeosoma*) was singled out for a more detailed risk analysis due to its extremely low numbers and its foraging behavior. The last 2 known individuals were last seen in 2003 and 2004 in its only known range, the native forest habitats of the upper Hanawi watershed (U.S. FWS 2006). The po′ouli feeds primarily on insects and snails by searching through leaves, branches, and trunks of ohia (*Metrosideris polymorpha*) and other trees and shrubs (Baldwin and Casey 1983, Scott *et al*. 1986, Mountainspring *et al*. 1990). They most frequently forage approximately 13 to 23 feet above the ground on the bark of the ohia tree and will pull up lichen and moss in search of prey. They have rarely been observed foraging on the forest floor (Mountainspring *et al*. 1990).

Po′ouli weigh approximately 32 g (U.S. FWS 2006). Therefore, the daily consumption estimates for similarly-sized birds calculated above are directly applicable to this species. Diphacinone residue data for ground-dwelling invertebrates, slugs (*Deroceras laeve, Limax maximus*), and snails (*Oxychilus sp*.), were reported by Johnston *et al*. (2005) (Table 4). The species tested by Johnston *et al*. include those commonly seen on bait in rodenticide bait stations and are principally ground-dwelling (Severns 1984). *D. laeve, L. maximus*, and *Oxichilus* sp. weigh approximately 0.43 g, 5.24 g, and 0.35 g, respectively (Eisemann, unpubl. data). A po′ouli would have to eat 2,555 g or 5,942 *D. laeve* (at a mean of 2.64 mg/kg), the species accumulating the highest residues, or 80 times the bird's body weight in a day to ingest a dose equivalent to the lowest reported avian LD_{50}. Dr. Greg Massey, former Hawaii State Veterinarian, recommended providing captive po′ouli with meal worms at a rate of 1.5 g/hour to 18 g/day (pers. commun.). In comparison, to ingest the quantity equivalent to the lowest dietary dose which caused mortality (LLD) in mallards (0.6 mg/kg/day, Long *et al*. 1992*b*), a po′ouli would have to eat 29 snails from the lab study or 75 field-collected snails daily for multiple days. Sublethal effects (LOEL) could occur if the bird ate as little as 0.7 g of *D. laeve* (at the maximum residue of 5.01 mg/kg) a day for multiple days. Even if one considered the average diphacinone residue in *D. laeve* (2.64 mg/kg) instead of the maximum, sublethal effects could occur if the bird ate as little as 1.33 g. This is equivalent to slightly more than 3 slugs. Johnston *et al*. (2005) reported average diphacinone residues in garlic snails (*Oxychilus sp*.) of 1.77 mg/kg and 0.69 mg/kg from laboratory experiments and field collection, respectively. At these diphacinone concentrations, a po′ouli would need to eat approximately 6 snails from the lab study or 15 field-collected snails per day for multiple days to ingest the dose that caused increased blood-clotting times in golden eagles. All of these calculations may dramatically overstate the risk to the po′ouli, because ground-dwelling mollusks were not observed in their diets (Baldwin and Casey 1983, Mountainspring *et al*. 1990).

The results of this simple stochastic assessment are supported by a more detailed probabilistic assessment conducted by Johnston *et al*. (2005). Johnston *et al*. determined that there was a 0% chance of mortality if a po′ouli were only exposed for a single day. However, when a 5-day exposure period was considered, the risk of mortality increased to 3% for adults and 8% for juveniles. For po′ouli that consume snails containing diphacinone residues for 14 days, Johnston *et al*. predicted clotting abnormalities for 0.42% and 11% of adult and juvenile birds, respectively.

The above assessments indicate a po′ouli could be at risk of lethal and sublethal effects from diphacinone even at small quantities of dietary intake. However, these assessments are based on multiple assumptions, each of which could impact the accuracy of the assessments. These assumptions include: 1) Hawaiian birds, specifically the po′ouli, are as sensitive as the most sensitive tested species, 2) diphacinone absorption rates are similar regardless of the matrix or gut in which it occurs, 3) mollusk species preyed upon by the po′ouli will be exposed to diphacinone pellets, and 4) all species of mollusks accumulate diphacinone residues at the same rate. Because the 6.5-g Ramik® Green bait pellet is specifically designed to be large enough to penetrate the forest canopy and come to rest on the forest floor, only a small proportion of broadcast pellets will be accessible to arboreal native mollusks. Therefore, the likelihood of po′ouli encountering diphacinone-exposed invertebrates as they forage in the canopy and on tree trunks is extremely low.

Acute and Sublethal Hazards to the Hawaiian Goose (Nene)

Primary and secondary hazard calculations for acute oral toxicity (LD_{50}) for the Hawaiian goose, or nene (*Branta sandwicensis*), presented in Table 7, show that a 1.5-kg (3.3-lb) bird would have to consume at least 12 kg (26.46 lbs) of bait or 120 kg (265 lbs) of invertebrates in one day to ingest a dose equivalent to the LD_{50} for the northern bobwhite. Since these quantities are well above the bird's body weight, the risk when calculated this way is extremely low. However, for a dose equivalent to the

lowest dietary dose which caused mortality (LLD) in a bird (0.6 mg/kg/day, Long *et al*. 1992*b*), a 1.5-kg bird would need to consume 18.00 g (0.04 lb) of bait or 179.64 g (0.40 lb) of contaminated invertebrates a day for multiple days. Hazard calculations for sublethal exposure (LOEL) presented in Table 7 show that a 1.5-kg bird would only need to eat 3.3 g of bait (~1/2 of a bait pellet) or 32.9 g of invertebrates per day for multiple days to ingest a dose that resulted in blood clotting effects in golden eagles. Although some of these conservative scenarios are within the reasonable range of daily food consumption rates of a 1.5-kg bird, their foraging behavior indicates that the risk of suffering lethal or sublethal effects through both primary and secondary exposure is still quite low.

Hawaiian geese are generalist vegetarians. Fecal analysis studies have documented consumption of at least 50 species of plants (Banko *et al*. 1999). Consumption of invertebrates may occur incidentally with the ingestion of plant material, but neither adults nor goslings appear to seek out or actively hunt invertebrates (Rojek 1994, Banko *et al*. 1999).

Two studies have attempted to characterize dietary preferences and diphacinone toxicity to geese. Massey *et al*. (no date) conducted a series of 2-choice palatability trials with the Hawaiian goose to evaluate consumption rates of nontoxic rodenticide baits. Massey *et al*.'s study showed that goose pairs consumed an average of 6.55 g (0.01 lb) of bait per day over the 5-day test period, a significantly smaller quantity than maintenance diet. Witmer (2001) used Canada geese (*Branta canadensis*) as a surrogate for the Hawaiian goose in a series of multiple-day, single-choice toxicity trials with both Ramik® Green and Eaton's Bait Pellet Rodenticide with Fish Flavorizer. On average, geese consumed approximately 11.0 g/day whole Eaton's bait, 14.1 g/day crushed Eaton's bait, 13 g/day whole Ramik® Green bait, and 4.2 g/day crushed Ramik® Green bait. No geese died during these tests and there was no reported effect on blood packed cell volume.

In both studies, Massey *et al*. (no date) and Witmer (2001), geese were shown to consume small quantities of rodenticide bait, quantities far smaller than what are predicted to cause mortality from acute exposure. These results can be used to further characterize sublethal exposure effects. Using Massey *et al*.'s reported bait consumption for pairs for a single bird (6.55 g) instead, if a wild Hawaiian goose consumed equivalent quantities of 0.005% diphacinone bait, a 1.5-kg Hawaiian goose would consume approximately 0.22 mg/kg of diphacinone per day. This is approximately 2 times higher than the lowest dose Savarie *et al*. (1979) demonstrated caused clotting disorders in eagles. The maximum amount of bait consumed by Canada geese in Witmer's (2001) study was 14.1 g/day/bird. At this rate, a 3-kg (6.61-lb) Canada goose eating bait would ingest 0.235 mg diphacinone/kg/day, a rate 2.1 times higher than Savarie *et al*.'s lowest dose. Yet, the Canada geese showed no effects, indicating that geese may be less susceptible to diphacinone than eagles. Depending upon which study the assessment is based on, there is either some or no potential for effects.

Under actual field conditions, the hazard may be significantly lower. As Witmer's (2001) data show, after eating only a few whole or crushed pellets, the birds found the bait unpalatable. The geese in Witmer's study fed Ramik® Green ate only 13 g/day (0.03 lb/day). Under field conditions, a 3-kg (6.61-lb) Canada goose would typically eat 90 g (0.20 lb) of food per day (U.S. EPA 1993). The geese in Witmer's study chose starvation over eating bait. Neophobia to new food items may have played a role in these results, but it was obvious the geese declined to eat the bait after trying it. Hawaiian geese feed almost exclusively on vegetation, including leafy material, fruits, and seeds. Ramik® Green is a grain-based bait but it is very hard and large (2 cm in diameter). Under natural conditions, it would be very difficult for a goose to eat the bait unless it had shattered upon application or had undergone significant weathering. Even then it is unlikely that they would eat enough to cause sublethal effects that could jeopardize their survival.

Acute and Sublethal Hazards to the Hawaiian Hawk ('Io)

Because of its carnivorous diet, the probability that a Hawaiian hawk will eat a Ramik® Green bait pellet is low. Therefore, a primary hazard assessment was not conducted. There is a secondary risk to the Hawaiian hawk from consuming live rodents or carcasses of rodents exposed to diphacinone baits. Secondary hazard calculations for acute oral toxicity (LD_{50}) for the Hawaiian hawk presented in Table 7 show that a 450-g (~1-lb) bird would have to consume at least 58.63 kg (129.26 lbs) of rodent tissue containing 3.07 ppm diphacinone in 1 day to ingest a dose equivalent to the LD_{50}. Since this is well above the bird's body weight, the risk when calculated this way is extremely low. For a dose equivalent to the lowest dietary dose which caused mortality (LLD) in mallards (0.6 mg/kg/day, Long *et al*. 1992*b*), a 450-g hawk would need to consume 87.95 g (0.19 lb) of contaminated rodents a day for multiple days. This is 20% of its body weight, also an unlikely scenario. Hazard calculations for sublethal exposure (LOEL) presented in Table 7 show that a 450-g bird would only need to eat 16.12 g (0.04 lb) of rodent tissue per day, less than a single mouse or rat, for multiple days to ingest a dose that caused blood clotting effects in golden eagles. Under these conservative scenarios, the Hawaiian hawk may be at risk of suffering sublethal but not lethal effects through secondary exposure, if their foraging habitat was treated with diphacinone bait.

Low accessibility of poisoned rodents could reduce the actual risk. Field studies have demonstrated that most rats exposed to diphacinone bait died in locations inaccessible to avian predators and scavengers (Lindsey and Mosher 1994, Spurr *et al*. 2003*a*,*b*). In addition, Lindsey and Mosher reported that avian predators did not take any rodent carcasses intentionally placed on the forest floor. They also reported that most radio-collared rats did not move during the day, before and after consuming diphacinone bait, and remained under cover in or adjacent to their nest sites, minimizing their exposure to diurnal avian predators. Spurr *et al*. (2003*b*) found a

Hawaiian hawk carcass 6 months after a forested study site had been aerially treated with Ramik® Green, but the cause of death could not be determined because of the decomposed state of the carcass. The Hawaiian hawk is widespread on the island of Hawaii and forages in most of the agricultural and conservation areas where diphacinone in bait stations is used. The hawk reported by Spurr *et al*. is the only dead native Hawaiian bird that has been found in any area treated with diphacinone.

Acute and Sublethal Hazards to the Hawaiian Crow ('Alala)

Primary and secondary hazard calculations for acute oral toxicity (LD_{50}) for the Hawaiian crow or ′Alala (*Corvus hawaiiensis*) presented in Table 7 show that a 500-g (1.10-lb) bird would have to consume at least 4 kg (8.82 lbs) of bait, 39.92 kg (88.01 lbs) of contaminated invertebrates containing 5.01 mg diphacinone/kg, or 65.3 kg (143.7 lbs) of rodents containing 3.07 ppm diphacinone in one day to ingest a dose equivalent to the LD_{50} for the northern bobwhite. Since these quantities are well above the bird's body weight, the risk when calculated this way is extremely low. For a dose equivalent to the lowest dietary dose (LLD) which caused mortality in mallards (0.6 mg/kg/day; Long *et al*. 1992*b*), a 500-g crow would need to consume 6 g of bait pellets (approximately one pellet), 59.88 g (0.13 lb) of contaminated invertebrates, or 97.7 g (0.22 lb) of rodents a day for multiple days. Mortality due to direct consumption of bait pellets under this conservative scenario is quite possible. Secondary poisoning due to consumption of contaminated invertebrates or rodents would require consumption of large amounts relative to the crow's body weight (20% of its weight for rodents). Hazard calculations for sublethal exposure (LOEL) presented in Table 7 show that a 500-g bird would only need to eat 1.10 g of bait (1/6 of a bait pellet), 10.98 g (0.02 lb) of contaminated invertebrates, or 17.92 g (0.04 lb) of rodents a day for multiple days to ingest a dose that caused blood clotting effects in golden eagles. Under these conservative scenarios, the Hawaiian crow may be at risk of suffering lethal effects from primary exposure, and sublethal effects through both primary and secondary exposure if its foraging habitat included treatment sites.

The generalist/opportunist diet of corvids (Banko *et al*. 2002) indicates the probability of a Hawaiian crow consuming either bait or diphacinone-exposed rodents is a concern if their home range includes baited areas. Mortality in wild corvids from primary or secondary consumption of Ramik® Green pellets has occurred, albeit under very different circumstances from those of the proposed broadcast technique. Dunlevy (unpubl. data) observed common ravens (*Corvus corax*) removing the 2-g (0.5 cm in diameter) Ramik® Green bait pellets from burrow placements and bait stations during rat eradications in the Aleutian Islands. A bird found dead had both Ramik® Green in its gastrointestinal tract and showed extensive signs of anticoagulant poisoning. Two live individuals were collected and both had diphacinone residues in their livers (0.17 ppm and 2.8 ppm). One individual had Ramik® Green and rat remains in its stomach. However, while this demonstrates that wild corvids are capable of consuming Ramik® Green bait, the consumption was facilitated by several factors. These birds had habituated to feeding on bait at fixed locations, rather than on widely scattered pellets, and likely a large quantity of bait over a period of weeks. Even under these circumstances, only one bird died (Dunlevy, pers. commun.). Furthermore, the 6.5-g (2 cm in diameter) Ramik® Green pellets that will be used for aerial broadcast are considerably larger and would be difficult to swallow.

Massey *et al*. (no date) attempted to characterize the risk of secondary exposure in a study on American crows (*Corvus brachyrhynchos*) using rats fed lethal amounts of 0.005% diphacinone Ramik® Green. One group of crows was fed one rat carcass on the first day of the study (low-dose group). Another group was fed one rat carcass each morning for 5 consecutive days and 2 rats on day 6 (high-dose group). One bird in the high-dose group showed signs of diphacinone poisoning (bleeding from the nares, pinpoint red spots in the cloacal mucosa). A significant increase in blood clotting times was noted in the high-dose group 8 days after exposure started. Only slight increases in prothrombin times were noted in the low-dose group 5 days after exposure. Based on the results of this study, the authors concluded that if American crows are appropriate surrogates for the endangered Hawaiian crow, rodent control programs using diphacinone bait would present minimal risk from secondary poisoning to the Hawaiian crow. They based this conclusion on the fact that under field conditions, most rats die underground (Lindsey and Mosher 1994, Spurr *et al*. 2003*a*,*b*) and it would be unlikely a crow would be able to find poisoned rats on a daily basis.

The Hawaiian crow is a critically endangered species, currently existing only in captivity, although reintroduction to the wild is being planned. Because the limited data suggest that exposure to diphacinone could produce both sublethal and lethal effects in corvids, further laboratory and field studies are needed to better characterize the risk to the Hawaiian crow. Laboratory studies with closely-related surrogate species should use sufficient sample sizes and carefully measured doses to quantify the toxicity of diphacinone to corvids. The potential for Hawaiian crows to actually ingest lethal and sublethal doses of broadcast diphacinone bait pellets should be evaluated based on the results of field studies using placebo bait and rodent carcasses.

Acute and Sublethal Hazards to the Hawaiian Owl (Pueo)

A primary hazard assessment for the Hawaiian owl was not conducted because of the low probability a Hawaiian owl would consume a Ramik® Green bait pellet. However, a Hawaiian owl living in a baited area could consume rodents exposed to diphacinone. Secondary hazard calculations for acute oral toxicity (LD_{50}) for the Hawaiian owl presented in Table 7 show that a 350-g (0.77-lb) bird would have to consume at least 45.60 kg (100.53 lbs) of rodents containing 3.07 ppm diphacinone (the highest residue found in feral pig liver) in one day to ingest a dose equivalent to the LD_{50} for the northern bobwhite. Since this is well above the bird's body

weight, this risk is extremely low. For a dose equivalent to the lowest dietary dose which caused mortality (LLD) in mallards (0.6 mg/kg/day, Long *et al*. 1992*b*), a 350-g owl would need to consume 68.40 g (0.15 lb) of rodents containing 3.07 ppm diphacinone for multiple days. Secondary poisoning from contaminated rodents would require consumption of large amounts relative to the owl's body weight (20% of its weight). Hazard calculations for sublethal exposure (LOEL) presented in Table 7 show that a 350-g owl would only need to eat 12.54 g (0.03 lb) of rodent tissue containing 3.07 ppm diphacinone per day (3.6% of its body weight) for multiple days to ingest a dose that caused blood clotting effects in golden eagles. This amount is less than one rodent per day, so using the most conservative measure the Hawaiian owl could be at risk of suffering sublethal effects through secondary exposure to rodents if its foraging habitat included treatment sites.

Pitt *et al*. (2005) reported finding a dead barn owl (*Tyto alba*) in a study site following aerial application of Eaton's Bait Pellet Rodenticide with Fish Flavorizer. The owl contained 0.081 ppm and 0.620 ppm diphacinone in the muscle and liver, respectively. These results can be compared to those of Mendenhall and Pank (1980) to further characterize this risk. In this study, 2 out of 3 great horned owls (*Bubo virginianus*) died 9 days after consuming an average maximum daily diphacinone dose of 0.78 mg/kg/day for 5 days. The survivor exhibited signs of anticoagulant poisoning (not specified by the authors). The only saw-whet owl (*Aegolius acadicus*) tested died 2 days after consuming a maximum daily diphacinone dose of 11.1 mg/kg/day for 5 days. No barn owls died or exhibited signs of anticoagulant poisoning when they consumed a maximum daily diphacinone dose of up to 1.71 mg/kg/day for 10 days. If the Hawaiian owl (350 g) was as sensitive as the most sensitive owl tested by Mendenhall and Pank (great horned owl, maximum daily dose of 0.67 mg/kg/day), a dose of 0.23 mg/day for 5 days could possibly cause mortality. This is roughly the equivalent of 75 g of rodent tissue containing 3.07 ppm diphacinone, or 3 average-sized house mice per day. This scenario is well within reason. Consequently, Hawaiian owls foraging within treated areas could be at risk of lethal exposure if they are as sensitive to diphacinone as the great horned owl. However, the Hawaiian owl is widespread in the Hawaiian Islands and forages in most of the agricultural and conservation areas where diphacinone in bait stations is used. No Hawaiian owl deaths have been linked to these field uses of diphacinone.

CONCLUSION

The ecological benefits of removing rodents from island ecosystems have been widely demonstrated in New Zealand, the U.S., and other countries. Rodent eradication projects conducted on islands in the U.S. have been done under the emergency use provision of FIFRA. In order for the broadcast application of rodenticides to become a viable tool for natural resource management in the U.S., a product must be registered and fully reviewed for human and ecological safety by the U.S. Environmental Protection Agency and state regulatory authorities where use is proposed.

The assessments in this manuscript are based on very conservative assumptions, and as a result are assumed to overestimate the actual hazard an aerial broadcast of a diphacinone rodenticide would pose. As such, these assessments should be used to identify those areas where additional consideration should be given during planning aerial rodenticide applications in Hawaii.

With the exception of the bat, at the median lethal dose level (the LD_{50}), the terrestrial and aquatic risk assessments indicate little risk of mortality from one-time exposure to diphacinone bait or from a single secondary exposure to other organisms containing diphacinone residues. However, since the proposed application sites include habitat for endangered species, it is prudent to evaluate the hazards based on the lowest doses known to result in mortality for individuals, and in an even more conservative approach, to evaluate the risk of animals suffering from slight physiological effects which could compromise their survival. In that respect, the calculations in Tables 6 and 7 indicate there could be effects of concern for all of the species evaluated.

However, we strongly caution against placing too much emphasis on the quantitative analyses for each species. Laboratory studies with surrogate species and field trials within the Hawaiian species' habitats have provided important information confirming or invalidating the theoretical calculations. Species in Hawaiian ecosystems may have physiological and/or behavioral traits which would either increase or decrease their risk of exposure and/or susceptibility to diphacinone. For example, despite the relatively low risk indicated for pigs based on laboratory toxicity studies, wild pigs died as a result of a misapplication of 0.005% rodenticide bait. In contrast, the Hawaiian goose's foraging behavior and dietary preferences substantially reduce its risk from that indicated by the toxicity calculations, and a number of assumptions used for the Hawaiian hoary bat likely substantially overestimate its risk.

The potential for lethal human exposure was not addressed in this assessment. Instead, we based the human assessment on subtle physiological effects that would occur at doses much lower than those resulting in death. The location of potential sites primarily in uninhabited areas limits human exposure. Most of the sites considered for aerial application are either remote and inaccessible or could have access restricted during the time bait may be on the ground. While people could consume bait particles or diphacinone in solution in their drinking water if the source were from a stream draining a watershed, the quantities of water required to even reach levels that might cause changes in blood clotting are beyond human ability. The one exception might be if a pregnant woman drank directly from a stream after an aerial broadcast, since a dose was not determined that did not cause maternal bleeding in laboratory rats. However, this is an unlikely scenario, given the remoteness of these streams.

This manuscript highlights some areas where more consideration should be given prior to making broadcast applications of a diphacinone rodenticide in native Hawaiian ecosystems. The planning process for future broadcast applications will include carefully designed

monitoring programs to better characterize the actual risk to the species in each proposed location. All of this information will help natural resource managers and others weigh the potential negative impacts from diphacinone against the benefits of reduced rodent depredation on Hawaii's native flora and fauna.

Aerial rodenticide applications conducted in insular areas elsewhere in the world have produced significant net benefits to native species and ecosystems. The short term loss of non-target individuals in a treatment site are typically offset by substantial increases in the abundance of species negatively impacted by rodent depredation. The direct and indirect impacts that introduced rodents have on Hawaii's ecosystems may be so great that the use of aerial broadcast of rodenticides in conservation areas could mean the difference between extinction and survival for some native species.

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