Risks of Hexazinone Use to Federally Threatened California Red-legged Frog
(*Rana aurora draytonii*)

Pesticide Effects Determination

Environmental Fate and Effects Division
Office of Pesticide Programs
Washington, D.C. 20460

February 20, 2008

Primary Authors:

Ronald Parker, PhD
Senior Environmental Engineer
Environmental Fate and Effects Division

Valerie Woodard, PhD
Biologist
Environmental Fate and Effects Division

Secondary Review:

Jean Holmes, D.V.M., M.P.H.
Mohammed Ruhman, Ph.D, Agronomist

Branch Chief, Environmental Risk Assessment Branch #:

Mah Shamim

Table of Contents

Appendices

1. Executive Summary

The purpose of this assessment is to evaluate potential direct and indirect effects on the California red-legged frog *(Rana aurora draytonii*) (CRLF) arising from FIFRA regulatory actions regarding use of hexazinone on agricultural and non-agricultural sites. In addition, this assessment evaluates whether these actions can be expected to result in modification of the species' designated critical habitat. This assessment was completed in accordance with the U.S. Fish and Wildlife Service (USFWS) and National Marine Fisheries Service (NMFS) *Endangered Species Consultation Handbook* (USFWS/NMFS, 1998 and procedures outlined in the Agency's Overview Document (U.S. EPA, 2004).

The CRLF was listed as a threatened species by USFWS in 1996. The species is endemic to California and Baja California (Mexico) and inhabits both coastal and interior mountain ranges. A total of 243 streams or drainages are believed to be currently occupied by the species, with the greatest numbers in Monterey, San Luis Obispo, and Santa Barbara counties (USFWS, 1996) in California.

Hexazinone is a triazinone herbicide used to control a broad spectrum of weeds including many annual, biennial and perennial weeds, as well as some woody plants in alfalfa, rangeland and pasture, woodland, pineapples, sugarcane and blueberries. It is also used for forest trees and other non-crop areas. Hexazinone is registered for pre-emergent, post-emergence, layby, directed spray and basal soil applications. It is used as a non-selective herbicide in noncropland areas and as a selective herbicide in reforestation practices. Current labels also include drainage areas, industrial sites, sewage disposal areas and airports. Of these uses, only a single application to one industrial site is recorded in the California PUR data reviewed for this assessment.
Although the maximum number of applications from the label for pasture, alfalfa and Christmas tree uses is one, other uses do not specify a maximum rate or maximum number of applications per year. For this assessment, one application per year for those other uses is based on additional information from the label. Blueberry uses are modeled at a maximum of one application per year due to no application within 450 days of harvest. Pineapple is modeled as one application per year due to the growing season, which is nearly a year in length. Noncrop uses and all granular uses, non-agricultural rights-of-way (ROW), forest site preparation and rangeland, are modeled as one application per year due to the requirement for rain to activate hexazinone and because maximum effects are achieved after 12-24 months.

The environmental fate properties of hexazinone, along with available monitoring data identifying its presence in surface water, air, and in precipitation in California, indicate that runoff, spray drift, volatilization, atmospheric transport and subsequent deposition represent potential transport mechanisms of hexazinone to the aquatic and terrestrial habitats of the CRLF. In this assessment transport of hexazinone from initial application sites through runoff and spray drift are considered in deriving quantitative estimates of hexazinone exposure to CRLF, its prey and its habitats. Although volatilization of hexazinone from treated areas resulting in atmospheric transport and eventual deposition represent relevant transport pathways leading to exposure of the CRLF and its habitats, it is expected that detected hexazinone concentrations in atmospheric monitoring data are reflective of near field spray drift and not long range transport, given hexazinone's low volatility and a lack of detections at higher elevations. In addition, adequate tools are not available at this time to quantify exposures through these pathways.

Therefore, volatilization, and potential atmospheric transport are discussed only qualitatively in this assessment.

Based on the available information, hexazinone appears to be persistent and mobile in soil and aquatic environments. The mobility of hexazinone was demonstrated in batch equilibrium data. The field and forestry dissipation data also confirm that hexazinone is persistent and mobile. Furthermore, the batch equilibrium data also suggests that degradates are very mobile. Based on the environmental fate properties of hexazinone, it can be concluded they may be of concern for surface water and groundwater contamination. This assessment is based on the parent because hexazinone is so persistent in the field, therefore consideration of toxicity of degradates will not change the result of this assessment.

Since CRLFs exist within aquatic and terrestrial habitats, exposure of the CRLF, its prey and its habitats to hexazinone are assessed separately for the two habitats. Tier-II aquatic exposure models are used to estimate high-end exposures of hexazinone in aquatic habitats resulting from runoff and spray drift from different uses. Peak model-estimated environmental concentrations resulting from different hexazinone uses range from 10.6 to156.6 μg/L. These estimates are supplemented with analysis of available California surface water monitoring data from the California Department of Pesticide Regulation (CDPR). Hexazinone is not on the list of pesticides for which the NAQWA program samples, therefore the NAQWA database will not be used.. There are three measured hexazinone concentrations above the limit of quantitation in the CDPR data. All three CDPR measured concentrations were recorded at the River Road sampling station on Orestimba Creek, tributary to the San Joaquin River in Stanislaus County. The sample with the highest of these three concentration values (0.5 μg/l) was collected on February 14, 2001. This maximum concentration of hexazinone reported by the California Department of Pesticide Regulation surface water database from 2000-2005 (x μg/L) is roughly 300 times lower than the highest peak model-estimated environmental concentration.

To estimate hexazinone exposures to the terrestrial-phase CRLF and its potential prey resulting from uses involving hexazinone applications, the T-REX model is used. For spray application uses AgDRIFT and AGDISP models are also used to estimate deposition of hexazinone on terrestrial and aquatic habitats from spray drift. The TerrPlant model is used to estimate hexazinone exposures to terrestrial-phase CRLF habitat, including plants inhabiting semi-aquatic and dry areas, resulting from uses involving foliar hexazinone applications. The T-HERPS model is used to allow for further characterization of dietary exposures of terrestrial-phase CRLFs relative to birds as a surrogate to the CRLF.

The assessment endpoints for the CRLF include direct toxic effects on the survival, reproduction, and growth of the CRLF itself, as well as indirect effects, such as reduction of the prey base or modification of its habitat. Direct effects to the CRLF in the aquatic habitat are based on toxicity information for freshwater fish, which are generally used as a surrogate for aquatic-phase amphibians. In the terrestrial habitat, direct effects are based on toxicity information for birds, which are used as a surrogate for terrestrial-phase amphibians. Given that the CRLF's prey items and designated critical habitat requirements in the aquatic habitat are dependant on the availability of freshwater aquatic invertebrates and aquatic plants, toxicity information for these taxonomic groups is also discussed. In the terrestrial habitat, indirect effects due to depletion of prey are assessed by considering effects to terrestrial insects, small terrestrial mammals, and frogs. Indirect effects due to modification of the terrestrial habitat are characterized by available data for terrestrial monocots and dicots.

Risk quotients (RQs) are derived as quantitative estimates of potential high-end risk. Acute and chronic RQs are compared to the Agency's levels of concern (LOCs) to identify instances where Hexazinone use within the action area has the potential to adversely affect the CRLF and its designated critical habitat via direct toxicity or indirectly based on direct effects to its food supply (i.e., freshwater invertebrates, algae, fish, frogs, terrestrial invertebrates, and mammals) or habitat (i.e., aquatic plants and terrestrial upland and riparian vegetation). When RQs for a particular type of effect are below LOCs, the pesticide is determined to have "no effect" on the subject species. Where RQs exceed LOCs, a potential to cause adverse effects is identified, leading to a conclusion of "may affect." If a determination is made that use of hexazinone use within the action area "may affect" the CRLF and its designated critical habitat, additional information is considered to refine the potential for exposure and effects, and the best available information is used to distinguish those actions that "may affect, but are not likely to adversely affect" (NLAA) from those actions that are "likely to adversely affect" (LAA) the CRLF and its critical habitat.

There is an overall "LAA" determination for hexazione exposure to the CRLF based on direct and indirect effects to the aquatic and terrestrial-phase CRLF. There is expected to be an effect of hexazinone exposure to the aquatic-phase CRLF. There is a "No Effect" determination for direct effects for the aquatic-phase CRLF using the fish as a surrogate due to no LOC exceedence.

There is an "LAA" determination for indirect prey reduction to the aquatic-phase CRLF. Indirect effects to the aquatic-phase CRLF via direct effects to food supply are determined using freshwater invertebrates, non-vascular plants, fish and frogs. There was no LOC exceedence for aquatic invertebrates resulting in a "no effect' determination for indirect effects to aquatic-phase CRLF via direct effects to food supply for all uses of hexazinone. There was an "LAA" determination for indirect dietary effects to the aquatic-phase CRLF consuming aquatic invertebrates. There was no LOC exceedence for fish resulting in a "no effect' determination for indirect effects to aquatic-phase CRLF via direct effects to food supply for all uses of hexazinone. There was no LOC exceedence for aquatic frogs using the fish as a surrogate, resulting in a "no effect' determination for indirect effects to aquatic-phase CRLF via direct effects to food supply for all uses of hexazinone.

There is an indirect "habitat modification" determination for the aquatic-phase CRLF due to LOC exceedences for non-vascular and vascular plants and the mode-of-action for hexazinone. A "likely to adversely affect" determination is made for some uses of hexazinone for indirect effects on habitat (i.e., aquatic and terrestrial plants) for the aquatic-phase CRLF.

There is expected to be an effect of hexazinone exposure to the terrestrial-phase CRLF. There is an "LAA" determination for direct effects for the terrestrial-phase CRLF using the birds as a surrogate due to endangered species LOC exceedence for (8 lb/A).

There is an "LAA" determination for indirect prey reduction to the terrestrial-phase CRLF due to the weight of evidence from results from T-REX and probit analysis, no relevant incident reports and no available open literature. Indirect effects to the terretrial-phase CRLF via direct effects to food supply are determined for terrestrial invertebrates, mammals, and.terrestrial amphibians using the bird as a surrogate. There was an "LAA" determination for terrestrial invertebrates due to the uncertainty due to no mortality at the highest concentration tested for all uses. There is an "LAA" determination for the CRLF consuming mammals for noncrop uses (8 lb/A) due to the RQ exceeding the non-listed LOC. There is a "LAA" determination for the CRLF consuming terrestrial amphibians using the bird as a surrogate for (8 lb/A) and (3 lb/A).There was an "LAA" determination for indirect dietary effects to the terrestrial-phase CRLF consuming aquatic invertebrates. There was an LOC exceedence for amphibians using birds resulting in a "no effect' determination for indirect effects to aquatic-phase CRLF via direct effects to food supply for all uses of hexazinone. There was no LOC exceedence for aquatic frogs using the fish as a surrogate, resulting in a "no effect' determination for indirect effects to aquatic-phase CRLF via direct effects to food supply for all uses of hexazinone.

There is an indirect "habitat modification" determination for all uses for the terrestrial-phase CRLF due to LOC exceedences for monocots and dicots and the mode-of-action for hexazinone.

Table 1.1 and 1.2 summarize hexazinone determinations.

Table 1.1 Effects Determination Summary for Direct and Indirect Effects of Hexazinone on the CRLF

Assessment Endpoint	Effects Determination[1]	Basis for Determination
Aquatic-Phase CRLF (Eggs, Larvae, and Adults)		
Direct Effect s of Hexazinone on the Aquatic-Phase CRLF		
Survival, growth, and reproduction of CRLF individuals via direct effects on aquatic phases	Using fish as a surrogate: No Effect	Using freshwater fish as a surrogate, no acute and chronic LOCs are exceeded for applications of Non-crop (agricultural Rights–of-Way) (12 lb/A-granular), there is no expectation for adverse effects for lower rates: noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas Tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
Indirect Effect s of Hexazinone on the Aquatic –Phase CRLF		
Survival, gth, and reproduction of CRLF individuals via effects to food supply (*i.e.,* freshwater invertebrates, non-vascular plants, fish, and frogs)	Freshwater invertebrates:	

	No effect	Using freshwater invertebrates, no acute and chronic LOCs are exceeded for applications of Non-agricultural Rights–of-way (12 lb/A-granular). Due to no exceedence for the 12 lb/A rate, it is assumed that there would also be no exceedence for the lower rates: Noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
	<u>Indirect Effects of Prey Reduction for Non-vascular aquatic plants for all uses</u>: May Affect <u>LAA</u> Non-vascular aquatic plants: No Effect	The May Affect for hexazinone uses related to applications for non-crop (non-agricultural ROW) (12 lb/A)non-crop uses (8 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), rangeland (3 lbs/A), Christmas trees (2.0 lb ai/A), alfalfa (1.5 lb ai/A) and pasture (1.1lb ai/A), exceed LOCs; therefore, indirect effects to tadpoles that feed on algae are possible. RQs range from 22.37 to 1.52. The LAA determination is due to mode of action Blueberry uses (RQ = 0.81) resulted in no LOC exceedence.
	Indirect Effects of Prey Reduction for Fish as surrogate for Frogs for all uses: No effect	 Using freshwater fish as a surrogate, no acute and chronic LOCs are exceeded for applications of non-crop (12 lb/A-granular), Noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A). .

Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat, cover, and/or primary productivity (*i.e.,* aquatic plant community)	Non-vascular aquatic plants: May Affect LAA No Effect	Hexazinone uses related to applications on Non-crop (12 lb/A-granular), Noncrop uses (8 lb/A), forest site preparation (5 lb/A), pineapple (3.6 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas Tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) exceed LOCs. Indirect effects to tadpoles that feed on algae are possible due to the MOA, which interferes with photosynthesis and RNA. There is no LOC exceedence for blueberry (3 lb/A) (RQ=0.81).
	Indirect Effects on habita for Vascular aquatic plants for all uses: May Affect LAA	The "May Affect" is based on the LOC exceedence for vascular aquatic plants for liquid applications of hexazinone to non-crop (12 lb/A), non-crop uses (8 lbs/A), Forest Site Preparation (5 lb/A), Conifer Release (3 lb/A).RQs range from 4.19 to 1.25. The "LAA" determination is based on the Mode of Action.
	No Effect	No LOC exceedence resulted for blueberry (3 lb/A), rangeland (3 lb/A) Christmas tree (2 lb/A), alfalfa (1.5 lb/A) or pasture (1.1 lb/A).
Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation, required to maintain acceptable water quality and habitat in ponds and streams comprising the species' current range.	Fforested and grassy/herbaceous riparian vegetation: May Affect	Riparian vegetation may be affected because terrestrial plant RQs exceed LOCs. RQs for semi-aquatic areas range from 87.66 to 937.50 Hexazinone effects on shading, bank stabilization, and structural diversity of riparian areas in the action area are expected. Aquatic-phase CRLFs may be indirectly affected by adverse effects to sensitive herbaceous vegetation (based on all hexazinone uses), which provides habitat and cover for the CRLF and attachment sites for its egg masses.

	LAA < 3366 ft (ground)	The LAA determination is based on MOA for ground applications of hexazinone within a drift buffer of 3366 ft based on the AGDISP results.
	NLAA ≥ 3366 ft (ground)	There is an "NLAA' determination fro animals outside the drift buffer of 3589 ft resulting from the AGDISP model.
	LAA <3589 ft (aerial);	The LAA determination is based on MOA for aerial applications of hexazinone within a drift buffer of 3589 ft based on the AGDISP results.
	NLAA ≥ 3589 ft (aerial)	There is an "NLAA" determination for animals outside the drift buffer of 3589 ft resulting from the AGDISP model.
Terrestrial-Phase CRLF (Juveniles and adults)		
Direct Effect s of Hexazinone on the Terrestrial-Phase CRLF		
Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	<u>Nongranular acute for medium size frog using the bird as a surrogate</u>: May affect	An adverse effect is expected based on weight of evidence for acute avian toxicity. The T-Rex analysis resulted in an endangered species exceedence for all crops.
	LAA for the medium size frog	Further refinement using the T-HERPS analysis resulted in an endangered species exceedence for the CRLF consuming small herbivore mammals with RQs ranging from 0.53 for (8 lb/A) resulting from T-Rex analysis.
	NLAA for the medium size frog	Further refinement using a T-HERPS analysis resulted in RQS falling between the endangered species (0.1) and acute risk (0.5) LOCs exceedence for the CRLF consuming small herbivore mammals for pineapple, blueberry, conifer release and Christmas tree uses for a 37 g animal. The RQs from the T-HERPS analysis were analyzed using probit analysis and resulted in discountable effects for prey reduction.
	No Effect for medium sized frog	A "No effect"determination for pasture (1.1 lb/A) is due to no LOC exceedence using the bird as a surrogate for the CRLF for the 37 g animal resulting from the T-REX analysis.

	Nongranular Acute Small Frog May Affect: NLAA	The May Affect resulted from LOC exceedence using the bird as a surrogate from the T-Rex analysis for all crops. An "NLAA" determination is due to no LOC exceedence for any use for the small CRLF (1.4 g) from the T-Herps analysis for consumption of small insect, large insect small, small herbivore mammal, insectivore mammals or small amphibian prey..
	Nongranular Acute Large Frog: May Affect: NLAA	The May Affect resulted from LOC exceedence using the bird as a surrogate in the T-Rex analysis for all crops. An "NLAA" determination is due to no LOC exceedence for any use for the juvenile CRLF (238 g) from the T-Herps analysis for consumption of small insect, large insect small, small herbivore mammal, insectivore mammals or small amphibian prey.
	Acute granular direct effects on the CRLF: No Effect	 The no effect determination is due to no LOC exceedence using the bird as a surrogate from the LDft[2] results for noncrop (non-agricultural ROW), forest site preparation (5 lb/A) and rangeland (3 lb/A).
	Nongranular chronic direct effects using the bird as a surrogate: May Affect: Direct chronic effects Using the bird as a surrogate: LAA for small herbivore mammal prey Direct chronic effects using the bird as a surrogate: NLAA for small herbivore mammal prey	 The May affect is based on LOC exceedence using the bird as a surrogate from te T-REX analysis. T-Herps was used to refine the May Affect from T-REX to either an "LAA" or "NLAA" determination. Chronic reproductive effects are possible, based on non-granular uses of hexazinone. T-Herps was used to refine the May Affect from T-Rex. The LAA determination resulted from LOC exceedence based on T-Herps for noncrop (8 lb/A) to Christmas tree (2 lb/A). RQs range from 4.22 to 1.05. The NLAA determination for alfalfa (1.5 lb/A) and pasture (1.1 lb/A) resulted from no LOC exceedence. RQs range from 0.79-0.58.

	Direct chronic effects using the bird as a surrogate: LAA for CRLF consuming small insect prey Direct chronic effects using the bird as a surrogate: NLAA for CRLF consuming small insect prey	T-Herps was used to refine the May Affect from T-REX to an "LAA" determination. All uses except Christmas tree, alfalfa and pasture resulted in LOC exceedences for broadleaf. RQs range from 3.6 (8 lb/A) to 1.35 (3 lb/A). Christmas tree (2 lb/), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) did not result in LOC exceedences for small small insect prey resulting from T-Herps modeling. RQs for small herbivore mammals range from 0.90 (1.5 lb/A) to 0.50 (1.1 lb/A).
	Direct chronic effects using the bird as a surrogate: NLAA consuming large insect prey Direct chronic effects uaing the bird as a surrogate: NLAA consuming small insectivore mammals Direct chronic effects using the bird as a surrogate: NLAA consuming small insectivore mammals	Non-crop (8 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A) Christmas tree (2 lb/A), alfalfa (1.1 lb/A) and pasture (1.1 lb/A) did not result in LOC exceedences for large insect prey resulting from T-Herps modeling. RQs for broadleaf range from 0.90 (2 lb/A) to 0.5 (1.1 lb/A) based on T-Herps analysis. An NLAA determination resulted from no LOC exceedence for small insectivore mammal,. RQs for small insectivore mammals range from 0.26 (8 lb/A) to 0.04 (1.1 lb/A). RQs for small amphibians range from for 0.12 (8 lb/A) to 0.02 (1.1 lb/A) based on T-Herps analysis.
	Chronic direct effects using the bird as a surrogate (Granular): May Affect LAA	Granular formulations for noncrop(12 lb/A), forest site preparation (5 lb/A) and rangeland (3 lb/A) resulted in LOC exceedences resulting from the T-REX analysis. RQs range from 1.35 for rangeland to 5.4 for noncrop.
Indirect Effect s of Hexazinone on the Terrestrial –Phase CRLF		

Survival, growth, and reproduction of CRLF individuals via effects on prey (*i.e.,* terrestrial invertebrates, small terrestrial vertebrates, including mammals and terrestrial phase amphibians)	Nongranualr Acute Indirect prey reduction for Terrestrial Invertebrates: May Affect	
	LAA	An LAA determination for small insect prey is based on the uncertainty regarding the effects of hexazinone on terrestrial invertebrates due to related toxicity values reporting no mortality at the highest concentration tested for all uses.
	Nongranular Acute Indirect prey reduction for terrestrial invertebrates (Large Insect Prey): May Affect	
	LAA	An LAA determination for large insect prey is based on the uncertainty regarding the effects of hexazinone on terrestrial invertebrates due to related toxicity values reporting no mortality at the highest concentration tested for noncrop (8 lb/A), pineapple (3.6 lb/A) and blueberry/conifer release (3 lb/A).
	Nongranular Acute No Effect	A "No effect" determination for large insect prey for Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) uses was based on no LOC exceedence from the T-REX analysis.
	Indirect mammal prey reduction:	
	Non-granular Acute May Affect	The May Affect is based on T-Rex results for 15 g mammal with a diet of shortgrass for non-crop (8 lb/A) through pasture (1.1 lb/A) uses.
	LAA	The LAA determination is due to noncrop (8 lb/A) and blueberry (3 lb/A) LOC exceedence from the T-REX analysis.

	Indirect mammal prey reduction Nongranular Acute: May Affect NLAA	The May Effect is based on RQs falling between the endangered species and acute risk LOC. Due to RQs falling between the endangered species and acute risk LOC further refinement used the the probit analysis. The potential reduction in abundance of terrestrial mammals as food for Christmas tree, alfalfa and pasture uses would be < 1%; therefore a "not likely to adversely affect" determination can be made.
	Granular Acute Indirect mammal prey reduction: No Effect (granular uses)	A No effect determination is based on the $LDft^2$ results from the T-REX analysis for non-crop(12 lb/A), forest site preparation (5 lb/A) and rangeland (3 lb/A).
	Indirect prey reduction for mammal Nongranular Chronic: May Affect:	Chronic reproductive effects are possible, based on non-granular uses of hexazinone. The May affect is due to LOC exceedences for all nongranular uses.

	LAA	All uses resulted in LOC exceedences for short grass. RQs range from 9.60 (8 lb/A) to 1.32 (1.1 lb/A) based on T-Rex.
	Indirect prey reduction for amphibian using the bird as a surrogate Nongranular Acute May Affect LAA	The May affect is due to LOC exceedence using the bird as a surrogate for all crops resulting from the T-Rex analysis. An "LAA" determination using the bird as a surrogate is based on LOC exceedence for noncrop (8 lb/A) uses. RQ for noncrop = 0.76 resulted from the T-REX analysis.
	Indirect prey reduction for amphibian using the bird as a surrogate NLAA	Conifer release, blueberry, Christmas tree, alfalfa and pasture uses resulted in RQs falling between the listed and non-listed LOCs from the T-REX analysis. Based on this assessment, the probit analysis was used as a further refinement for the determination. The potential reduction in abundance of amphibians as food for these uses would be < 1% at most; therefore a "not likely to adversely affect" determination can be made.
	Indirect prey reduction for amphibian using the bird as a surrogate Nongranular Chronic May Affect:	Chronic reproductive effects are possible, based on non-granular uses of hexazinone resulting from the T-REX analysis using the bird as a surrogate.

	LAA	All nongranular uses except Christmas tree, alfalfa and pasture resulted in LOC exceedences for broadleaf. RQs for broadleaf range from 1.35 (3 lb/A) to 3.60 (8 lb/A).
	<u>Amphibian using the bird as a surrogate</u> Nongranular Chronic No Effect	Christmas tree, alfalfa and pasture did not result in LOC exceedences broadleaf. RQs for broadleaf range from 0.5 (3 lb/A) to 0.90 (1.5 lb/A) resulting from the T-Rex analysis. .
Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.,* riparian vegetation)	<u>Woody and grassy/herbaceous riparian vegetation:</u> May Affect LAA < 184 ft (ground)	Riparian woody and herbaceous vegetation may be affected because terrestrial plant RQs are above LOCs. Due to MOA, which interferes with photosynthesis and RNA, terrestrial-phase CRLFs may be indirectly affected by adverse effects to sensitive woody and herbaceous vegetation which provide habitat and cover for the CRLF and its prey.
	NLAA ≥ 184 ft (ground)	There was an "NLAA" determination for animals outside the aerial drift buffer of 184 ft resulting from the AGDISP model..

	LAA < 850 ft (aerial);	Due to MOA, which interferes with photosynthesis and RNA, terrestrial-phase CRLFs may be indirectly affected by adverse effects to sensitive woody and herbaceous vegetation which provide habitat and cover for the CRLF and its prey.
	NLAA ≥ 850 ft (aerial)	There was an "NLAA" determination for animals outside the aerial drift buffer of 850 ft resulting from the AGDISP model.

Table 1.2 Effects Determination Summary for the Critical Habitat Impact Analysis		
Assessment Endpoint	**Effects Determination**	**Basis for Determination**
Aquatic-Phase PCEs (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)		
Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.	Habitat modification	Due to the MOA which interferes with photosynthesis and RNA ,sensitive herbaceous riparian vegetation may be affected based on all modeled uses of hexazinone; therefore, critical habitat may be modified by an increase in sediment deposition and reduction in herbaceous riparian vegetation that provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult aquatic-phase CRLFs.
Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.[1]	Habitat modification	Sensitive non-vascular aquatic plants may be affected; therefore, critical habitat may be modified via turbidity and reduction in oxygen content necessary for normal growth and viability of juvenile and adult aquatic-phase CRLFs.
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	No effect to growth and viability Habitat modification based on alteration of food source	Direct effects to the aquatic-phase CRLF, via mortality, growth, and/or fecundity, are not expected. However, critical habitat of the CRLF may be modified via hexazinone-related impacts to non-vascular aquatic plants as food items for tadpoles. LOCs are exceeded for non-vascular uses for non-crop (non-agricultural ROW) (12 lb/A), noncrop (8 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas Trees (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
	No Habitat Modification	There was no LOC exceedence for blueberry uses for nonvascular plants.
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.,* algae)	Habitat modification	Based on the results of the effects determinations for aquatic plants, critical habitat of the CRLF may

[1] Physico-chemical water quality parameters such as salinity, pH, and hardness are not evaluated because these processes are not biologically mediated and, therefore, are not relevant to the endpoints included in this assessment.

		be modified via hexazinone-related impacts to non-vascular aquatic plants as food items for tadpoles. LOCs are exceeded for modeled uses for (12 lb/A), noncrop uses (8 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas trees (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
	No Habitat Modification	Blueberry use resulted in no LOC exceedence.
Terrestrial-Phase PCEs (Upland Habitat and Dispersal Habitat)		
Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance	Habitat modification	Based on MOA, modification to critical habitat may occur via hexazinone-related impacts to sensitive woody and herbaceous vegetation, which provide habitat and cover for the terrestrial-phase CRLF and its prey, based on all assessed uses of hexazinone**.** Terrestial incident reports support a "habitat modification" determination.
Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal	Habitat modification	Based on the MOA for hexazione modification to dispersal habitat may occur via hexazinone-related impacts to sensitive woody and herbaceous vegetation, which provide habitat and cover for the terrestrial-phase CRLF and its prey, based on all assessed uses of hexazinone**.** Terrestrial incident reports support the "habitat modification' determination.
Reduction and/or modification of food sources for terrestrial phase juveniles and adults	Habitat modification	Based on the characterization of indirect effects to terrestrial-phase CRLFs via reduction in the prey base, critical habitat may be modified via a reduction in mammals and terrestrial-phase amphibians as food items.
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	Habitat modification	Direct acute effects, via mortality, are expected for the terrestrial-phase CRLF. Chronic reproductive effects are also possible. Therefore, hexazinone may modify critical habitat by altering chemical characteristics necessary for normal growth and viability of terrestrial-phase CRLFs and their mammalian and amphibian food sources.

When evaluating the significance of this risk assessment's direct/indirect and habitat modification effects determinations, it is important to note that pesticide exposures and predicted risks to the species and its resources (i.e., food and habitat) are not expected to be uniform across the action area. In fact, given the assumptions of drift and downstream transport (i.e., attenuation with distance), pesticide exposure and associated risks to the species and its resources are expected to decrease with increasing distance away from the treated field or site of application. Evaluation of the implication of this non-uniform distribution of risk to the species would require information and assessment techniques that are not currently available. Examples of such information and methodology required for this type of analysis would include the following:

- Enhanced information on the density and distribution of CRLF life stages within specific recovery units and/or designated critical habitat within the action area. This information would allow for quantitative extrapolation of the present risk assessment's predictions of individual effects to the proportion of the population extant within geographical areas where those effects are predicted. Furthermore, such population information would allow for a more comprehensive evaluation of the significance of potential resource impairment to individuals of the species.
-
- Quantitative information on prey base requirements for individual aquatic- and terrestrial-phase frogs. While existing information provides a preliminary picture of the types of food sources utilized by the frog, it does not establish minimal requirements to sustain healthy individuals at varying life stages. Such information could be used to establish biologically relevant thresholds of effects on the prey base, and ultimately establish geographical limits to those effects. This information could be used together with the density data discussed above to characterize the likelihood of adverse effects to individuals.
-
- Information on population responses of prey base organisms to the pesticide. Currently, methodologies are limited to predicting exposures and likely levels of direct mortality, growth or reproductive impairment immediately following exposure to the pesticide. The degree to which repeated exposure events and the inherent demographic characteristics of the prey population play into the extent to which prey resources may recover is not predictable. An enhanced understanding of long-term prey responses to pesticide exposure would allow for a more refined determination of the magnitude and duration of resource impairment, and together with the information described above, a more complete prediction of effects to individual frogs and potential modification to critical habitat.

2. Problem Formulation

Problem formulation provides a strategic framework for the risk assessment. By identifying the important components of the problem, it focuses the assessment on the most relevant life history stages, habitat components, chemical properties, exposure routes, and endpoints. The structure of this risk assessment is based on guidance contained in U.S. EPA's *Guidance for Ecological Risk Assessment* (U.S. EPA 1998), the Services' *Endangered Species Consultation Handbook* (USFWS/NMFS 1998) and is consistent with procedures and methodology outlined in the Overview Document (U.S. EPA 2004) and reviewed by the U.S. Fish and Wildlife Service and National Marine Fisheries Service (USFWS/NMFS 2004).

2.1 Purpose

The purpose of this endangered species assessment is to evaluate potential direct and indirect effects on individuals of the federally threatened California red-legged frog *(Rana aurora draytonii*) (CRLF) arising from FIFRA regulatory actions regarding use of hexazinone on both agricultural and non-agricultural sites. These includes: agricultural rights-of-way, fencerows and hedgerows; agricultural uncultivated areas; airports and landing fields; alfalfa; blueberries; Christmas tree plantations; conifer release applications; drainage systems; forest plantings in reforestation programs; tree farms and plantations; forest trees; outdoor industrial areas; non-crop/nonagricultural rights-of-way; nonagricultural uncultivated areas and soils; pastures; pineapples; rangeland; sewage disposal areas and sugarcane.

In addition, this assessment evaluates whether use on these crop sites is expected to result in modification of the species' designated critical habitat. This ecological risk assessment has been prepared consistent with a settlement agreement in the case *Center for Biological Diversity (CBD) vs. EPA et al*. (Case No. 02-1580-JSW (JL)) settlement entered in Federal District Court for the Northern District of California on October 20, 2006.

In this assessment, direct and indirect effects to the CRLF and potential modification to its designated critical habitat are evaluated in accordance with the methods described in the Agency's Overview Document (U.S. EPA 2004). Screening level methods include use of standard models such as PRZM-EXAMS, T-REX, TerrPlant, AgDRIFT, and AGDISP, all of which are described at length in the Overview Document. Use of such information is consistent with the methodology described in the Overview Document (U.S. EPA 2004), which specifies that "the assessment process may, on a case-by-case basis, incorporate additional methods, models, and lines of evidence that EPA finds technically appropriate for risk management objectives" (Section V, page 31 of U.S. EPA 2004).

In accordance with the Overview Document, provisions of the ESA, and the Services' *Endangered Species Consultation Handbook,* the assessment of effects associated with registrations of hexazinone is based on an action area. The action area is the area directly or indirectly affected by the federal action, as indicated by the exceedence of the Agency's Levels of Concern (LOCs). It is acknowledged that the action area for a national-level FIFRA regulatory decision associated with a use of hexazinone may potentially involve numerous areas throughout the United States and its Territories. However, for the purposes of this assessment, attention will be focused on relevant sections of the action area including those geographic areas associated with locations of the CRLF and its designated critical habitat within the state of California.

As part of the "effects determination," one of the following three conclusions will be reached regarding the potential use of hexazinone in accordance with current labels:

- "No effect";
- "May affect, but not likely to adversely affect"; or
- "May affect and likely to adversely affect".

Designated critical habitat identifies specific areas that have the physical and biological features, (known as primary constituent elements or PCEs) essential to the conservation of the listed species. The PCEs for CRLFs are aquatic and upland areas where suitable breeding and non-breeding aquatic habitat is located, interspersed with upland foraging and dispersal habitat.

If the results of initial screening-level assessment methods show no direct or indirect effects (no LOC exceedances) upon individual CRLFs or upon the PCEs of the species' designated critical habitat, a "no effect" determination is made for use of hexazinone as it relates to this species and its designated critical habitat. If, however, potential direct or indirect effects to individual CRLFs are anticipated or effects may impact the PCEs of the CRLF's designated critical habitat, a preliminary "may affect" determination is made for the FIFRA regulatory action regarding hexazinone.

If a determination is made that use of hexazinone within the action area(s) associated with the CRLF "may affect" this species or its designated critical habitat, additional information is considered to refine the potential for exposure and for effects to the CRLF and other taxonomic groups upon which these species depend (e.g., aquatic and terrestrial vertebrates and invertebrates, aquatic plants, riparian vegetation, etc.). Additional information, including spatial analysis (to determine the geographical proximity of CRLF habitat and hexazinone use sites) and further evaluation of the potential impact of hexazinone on the PCEs is also used to determine whether modification of designated critical habitat may occur. Based on the refined information, the Agency uses the best available information to distinguish those actions that "may affect, but are not likely to adversely affect" from those actions that "may affect and are likely to adversely affect" the CRLF or the PCEs of its designated critical habitat. This information is presented as part of the Risk Characterization in Section 5 of this document.

The Agency believes that the analysis of direct and indirect effects to listed species provides the basis for an analysis of potential effects on the designated critical habitat. Because hexazinone is expected to directly impact living organisms within the action area (defined in Section 2.7), critical habitat analysis for hexazinone is limited in a practical sense to those PCEs of critical habitat that are biological or that can be reasonably linked to biologically mediated processes (i.e., the biological resource requirements for the listed species associated with the critical habitat or important physical aspects of the habitat that may be reasonably influenced through biological processes). Activities that may modify critical habitat are those that alter the PCEs and appreciably diminish the value of the habitat. Evaluation of actions related to use of hexazinone that may alter the PCEs of the CRLF's critical habitat form the basis of the critical habitat impact analysis. Actions that may affect the CRLF's designated critical habitat have been identified by the Services and are discussed further in Section 2.6.

2.2 Scope

Hexazinone is an herbicide registered nationally for control of weeds on alfalfa, forestry (conifer release, forest tree planting), rights-of-ways, pastures, rangeland, Christmas trees, crops (sugarcane, pineapple and blueberries) and uncultivated areas, (non-crop/non-agricultural ROW, agricultural ROW, outdoor industrial, fencerows, airports, sewage, drainage) .

The end result of the EPA pesticide registration process (the FIFRA regulatory action) is an approved product label. The label is a legal document that stipulates how and where a given pesticide may be used. Product labels (also known as end-use labels) describe the formulation type (e.g., liquid or granular), acceptable methods of application, approved use sites, and any restrictions on how applications may be conducted. Thus, the use or potential use of hexazinone in accordance with the approved product labels for California is “the action” being assessed.

Although current registrations of hexazinone allow for use nationwide, this ecological risk assessment and effects determination addresses currently registered uses of hexazinone in portions of the action area that are reasonably assumed to be biologically relevant to the CRLF and its designated critical habitat. Further discussion of the action area for the CRLF and its critical habitat is provided in Section 2.7.

The Agency does not routinely include, in its risk assessments, an evaluation of mixtures of active ingredients, either those mixtures of multiple active ingredients in product formulations or those in the applicator’s tank. In the case of the product formulations of active ingredients (that is, a registered product containing more than one active ingredient), each active ingredient is subject to an individual risk assessment for regulatory decision regarding the active ingredient on a particular use site. If effects data are available for a formulated product containing more than one active ingredient, they may be used qualitatively or quantitatively in accordance with the Agency’s Overview Document and the Services’ Evaluation Memorandum (U.S., EPA 2004; USFWS/NMFS 2004).

Hexazinone has seven registered products that contain multiple active ingredients. Analysis of the available acute oral mammalian LD50 data for multiple active ingredient products relative to the single active ingredient is provided in Appendix A. The results of this analysis show that an assessment based on the toxicity of the single active ingredient of hexazinone is appropriate.

As discussed in USEPA (2000) a quantitative component-based evaluation of mixture toxicity requires data of appropriate quality for each component of a mixture. In this mixture evaluation LD_{50}s, with associated 95% confidence intervals, are needed for the formulated product. The same quality of data is also required for each component of the mixture. Given that many of the formulated products do not have LD_{50} values of the required quality, and since LD_{50} values are not available for all the components of these formulations, a quantitative analysis of potential interactive effects is not possible.

Hexazione has seven registered products that contain multiple active ingredients. Analysis of the available open literature data and acute oral mammalian LD_{50} data for multiple active ingredient products relative to the single active ingredient is provided in Appendix A. The result of this analysis show that an assessment based on the toxicity of the single active ingredient of hexazinone is appropriate.

In the case of hexazinone, only two products (EPA Reg. Nos. 352-603 and 352-618) have definitive product oral LD50 values. Although there are no confidence intervals associated with these products, the LD50 values for the products (1421 and 2073 mg/kg, respectively) are greater than the LD50 value for hexazinone (1200 mg/kg) and therefore do not indicate that the formulated products exhibit interactive effects. No further analysis is possible when LD50 values with associated confidence intervals are not available.

One open literature article (Ludy 2004) reported the use of hexazinone and diuron in mixtures for toxicity tests with *Chironomus tentans.* The resulting EC_{50}>1000 µg/L for the aquatic midge for both chemicals for individual toxicity tests. However, this study was not used in this assessment because the hexazinone and diuron were mixed with organo-phosphates, not other herbicides.

Because the active ingredients are not expected to have similar mechanisms of action, metabolites, or toxicokinetic behavior, it is reasonable to conclude that an assumption of dose-addition would be inappropriate. Consequently, an assessment based on the toxicity of hexazinone is the only reasonable approach that employs the available data to address the potential acute risks of the formulated products

2.3 Previous Assessments

A Reregistration Eligibility Decision (RED) that included an ecological risk assessment for aquatic fish, invertebrates, and plants, was issued in September 1994. The data were sufficient to allow the Agency to assess the registered uses of hexazinone and to determine that hexazinone could be used without resulting in unreasonable adverse effects to humans and the environment. Due to the risk to non-target plants, reduction of the maximum application rate from 13.5 lb a.i. per acre to 8 lb a.i. per acre was required. The 13.5 lb a.i. per acre rate is no longer in use, but there are still rates of 10.0 and 12.0 lbs a.i. per acre included on the label for selected non-agricultural uses. These rates are included in this assessment.

That assessment also determined that exposure of non-target organisms to hexazinone could result from direct application, spray drift from treated areas, and runoff from treated areas. Such exposure would be chronic as well as acute. Hexazinone exceeded the levels of concern (LOC) for terrestrial and aquatic plants, at all application rates, using aerial and ground equipment. Contamination of aquatic sites adjacent to treated areas could be of great ecological significance and may be exacerbated by the persistence and mobility of hexazinone. Effects to aquatic plants expected from the use of hexazinone may alter aquatic ecosystems, the severity of which is dependent on the frequency of application and the nature of the receiving body of water. Hexazinone also exceeded the LOC for small mammals at several of the higher application rates. Hexazinone exceeded the endangered species LOCs for grass- and insect-eating mammals at use rates of 3.6 pounds active ingredient per acre (lb ai/acre) or greater. It also exceeded the LOCs for both aquatic and terrestrial plants at all use rates.

2.4 Stressor Source and Distribution

2.4.1 Environmental Fate Properties

Hexazinone is a systemic herbicide used to control weeds and woody plants. Its potency is a function of soil texture, soil organic matter and post-application weather as it is activated by moisture. It is toxic to some evergreen trees and most broadleaf trees. The labels suggest field testing by the user for possible toxicity to evergreens for which the toxicity is uncertain. Based on the available information, hexazinone appears to be persistent and mobile in soil and aquatic environments and is considered as the stressor in this assessment (Table 2.1). The mobility of hexazinone was demonstrated in batch equilibrium as well as field and forestry dissipation data. It is stable to hydrolysis and photolysis and degrades only slowly by aerobic metabolism. Aerobic aquatic and aerobic soil half-life values are 60 and 216 days, respectively, and field dissipation half-lives were 123 to 154 days in bare ground silt loam soil. Laboratory adsorption data show low water/soil partitioning for hexazinone (K_{OC} = 37). These data indicate that hexazinone is highly mobile, thus having strong potential to leach to ground water systems, especially in soil systems such as loam and sand soils that are poor in organic matter.

Hexazinone is very soluble in water at 20°C with a solubility of 33,000 mg/L. Based on its low vapor pressure (2.0 x 10^{-7} mm Hg at 20°C) and Henry's Law Constant (2.0 x 10^{-12} atm·m^3/mol at 25°C), volatilization loss of hexazinone from soil and water systems is expected to be insignificant. Degradates and transformation products are formed slowly and are also mobile and persistent. Based on laboratory bioaccumulation in rainbow trout, hexazinone is not expected to bioaccumulate in fish, which is in concurrence with the low K_{ow} value of 15 (log K_{ow} = 1.12). Hexazinone has been shown to accumulate in rotational crops.

Table 2.1 Summary of Hexazione Chemical and Environmental Fate Properties			
Study Type/Hexazinone		**Value**	**Source (MRID)**
Water solubility	ppm (25 C)	33,000	Product Chemistry
Vapor pressure	mm Hg	2x10-7	Product Chemistry
Kow		15	Product Chemistry
Henry's Law Const	atm-m3/mol	2x10–12	Product Chemistry
Hydrolysis	pH 5	Stable	41587301 (acceptable)
	pH 7	Stable	41587301 (acceptable)
	pH 9	Stable	41587301 (acceptable)
Aqueous Photolysis (pH 7)		Stable	41300801 (acceptable)
Soil Photolysis		82 days	41300802 (acceptable)
Aerobic Soil		216 days	41807401; 42635001 (acceptable)
Anaerobic Aquatic		230 days	41807402 (acceptable)
Aerobic Aquatic		60 days	41811801 (acceptable)
Adsorption K_d		0.45 0.18	41528101 (acceptable) 43621501 (supplemental)
Adsorption Koc		37 50	41528101 (acceptable) 43621501 (supplemental)

2.4.2 Environmental Transport Mechanisms

Potential transport mechanisms include pesticide surface water runoff and spray drift. The magnitude of pesticide transport via secondary drift depends on the pesticide's ability to be mobilized into air and its eventual removal through wet and dry deposition of gases/particles and photochemical reactions in the atmosphere. A number of studies have documented atmospheric transport and redeposition of pesticides from the Central Valley to the Sierra Nevada Mountains (Fellers et al., 2004, Sparling et al., 2001, LeNoir et al., 1999, and McConnell et al., 1998). Prevailing winds blow across the Central Valley eastward to the Sierra Nevada Mountains, transporting airborne industrial and agricultural pollutants into Sierra Nevada ecosystems (Fellers et al., 2004, LeNoir et al., 1999, and McConnell et al., 1998). Therefore, physicochemical properties of the pesticide that describe its potential to enter the air from water or soil (e.g., Henry's Law constant and vapor pressure), pesticide use, modeled estimated concentrations in water and air, and available air monitoring data from the Central Valley and the Sierra Nevadas are considered in evaluating the potential for atmospheric transport of hexazinone to habitat for the CRLF**.**

In general, deposition of drifting or volatilized pesticides is expected to be greatest close to the site of application. Computer models of spray drift (AgDRIFT or AGDISP) are used to determine if the exposures to aquatic and terrestrial organisms are below the Agency's Levels of Concern (LOCs). If the limit of exposure that is below the LOC can be determined using AgDRIFT or AGDISP, longer-range transport is not considered in defining the action area. For example, if a buffer zone <1,000 feet (the optimal range for AgDRIFT and AGDISP models) results in terrestrial and aquatic exposures that are below LOCs, no further drift analysis is required. If exposures exceeding LOCs are expected beyond the standard modeling range of AgDRIFT or AGDISP, the Gaussian extension feature of AGDISP may be used. In addition to the use of spray drift models to determine potential off-site transport of pesticides, other factors such as available air monitoring data and the physicochemical properties of the chemical are also considered. Vegetative vigor and seedling emergence toxicity studies show that hexazinone is toxic to monocot and dicot terrestrial plants, thus the distance of potential impact away from the use sites (action area) is determined by the distance required to fall below the LOC for these non-target plants.

2.4.3 Mechanism of Action

Hexazinone is a systemic insecticide activated by irrigation water or rainfall in the period after application. It inhibits photosynthesis and, at higher levels of exposure, inhibits the synthesis of RNA, proteins, and lipids in plants.

2.4.4 Use Characterization

Both liquid and granular formulations of hexazinone may be applied by aircraftas well as ground sprayer.While the specific equipment for ground application varies between the liquid and granular formulations, both types of formulations are applied such that the herbicide sprayer for liquid or suspended granules or other equipment for granules is carried by backpack or some other appropriate container. Usually, hexazinone is applied directly to the soil rather than sprayed on the vegetation; however, sometimes, directed foliar applications are used. In soil applications, the hexazinone is applied in spots using a defined pattern.

Analysis of labeled use information is the critical first step in evaluating the federal action. The current label for hexazinone represents the FIFRA regulatory action; therefore, labeled use and application rates specified on the label form the basis of this assessment. The assessment of use information is critical to the development of the action area and selection of appropriate modeling scenarios and inputs.

Nationally, hexazinone is an herbicide used to control a broad spectrum of weeds including undesirable woody plants in alfalfa, rangeland and pasture, woodland, pineapples, sugarcane and blueberries. It is also used on ornamental plants, forest trees and other non-crop areas. Hexazinone is registered for pre-emergent, post-emergence, layby, directed spray and basal soil applications. It is used as a non-selective herbicide in noncropland areas and as a selective herbicide in reforestation practices. Hexazinone products are formulated as granulars, pellets/tablets, emulsifiable concentrates, ready-to-use liquids, soluble concentrates/solids and a technical grade active ingredient. Products are applied using aerial or ground equipment or by hand, or using a hand-held, boom, knapsack or power sprayer. Use practice limitations prohibit application of hexazinone through any type of irrigation system. There are no indoor uses.

National (Section 3) uses for hexazinone are presented in Table 2.2 with label maximum application rates. Although the maximum number of applications from the label for pasture, alfalfa and Christmas tree uses is one, other uses do not specify a maximum rate or maximum number of applications per year. For this assessment, one application per year for those other uses is based on additional information from the label. Blueberry uses are modeled at a maximum of one application per year due to no application within 450 days of harvest. Pineapple is modeled as one application per year due to the growing season, which is nearly a year in length. Noncrop uses and all granular uses, non-crop, forest site preparation and rangeland, are modeled as one application per year due to the requirement for rain to activate hexazinone and because maximum effects are achieved after 12-24 months.

Table 2.2 Hexazinone Uses[1] and Application Information

Crop	Max Appl Rate (lbs a.i./A)
NON-Crop (granular)	12
SEWAGE DISPOSAL AREAS (Non-granular)	8.
AGRICULTURAL RIGHTS-OF-WAY/FENCEROWS/HEDGEROWS (Non-granular)	8.
AGRICULTURAL UNCULTIVATED AREAS (Non-granular)	8.
AIRPORTS/LANDING FIELDS (granular)	8.
CONIFER RELEASE (non-granular)	3

Sugarcane (non-granular)	3.6
Pineapple (non-granular)	3.6
BLUEBERRY (non-granular)	3
RANGELAND (granular)	3
CHRISTMAS TREE PLANTATIONS (non-granular)	2.
ALFALFA (non-granular)	1.5
PASTURES (non-granular)	1.1
2 Uses assessed based on memorandum from SRRD dated 07/31/2007	

The Agency's Biological and Economic Analysis Division (BEAD) provides an analysis of both national- and county-level usage information (Hexazinone LUIS Report, 2007) using state-level usage data obtained from USDA-NASS[2], Doane (www.doane.com; the full dataset is not provided due to its proprietary nature), and the California Department of Pesticide Regulation Pesticide Use Reporting (CDPR PUR) database[3]. CDPR PUR is considered a more comprehensive source of usage data than USDA-NASS or EPA proprietary databases, and thus the usage data reported for hexazinone by county in this California-specific assessment were generated using CDPR PUR data. Usage data are averaged together over the years 2002 to 2005 to calculate average annual usage statistics by county and crop for hexazinone, including pounds of active ingredient applied and base acres treated. California State law requires that every commercial pesticide application be reported to the state and made available to the public. The summary of hexazinone usage for all use sites, including both agricultural and non-agricultural, is provided below in Table 2.3.

The two main hexazinone uses in the usage summary are the alfalfa and forest/timberland categories. Other uses include one to a few applications over the four-year period. There are several applications recorded in the rights-of-way category but only one single application is recorded on a per-acre basis and the data is categorized as questionable. There is no recorded usage on any of the three food crops on the label (blueberry, sugar cane, pineapple) in the 2002-2005 time periods.

Table 2.3 Summary of CALPUR Usage Data for Hexazinone (2002-2005)

Active Ingredient	Site Name	Range of Average Application Rates	Range of 95th Percentile Application Rates	Range of 99th Percentile Application Rates	Range of Maximum Application Rates
HEXAZINONE	ALFALFA	0.31-0.81	0.50-2.25	0.50-2.25	0.50-2.25
HEXAZINON E	FOREST, TIMBERLAND	0.56-3.14	0.71-16.39	0.71-16.74	0.71-29.77
HEXAZINONE	CHRISTMAS TREES	1.00-1.43	1.00-4.13	1.00-4.13	1.00-4.13
HEXAZINONE	RIGHTS OF WAY	0.30	0.30	0.30	0.30

[21] United States Department of Agriculture (USDA), National Agricultural Statistics Service (NASS) Chemical Use Reports provide summary pesticide usage statistics for select agricultural use sites by chemical, crop and state. See http://www.usda.gov/nass/pubs/estindx1.htm#agchem.
[23] The California Department of Pesticide Regulation's Pesticide Use Reporting database provides a census of pesticide applications in the state. See http://www.cdpr.ca.gov/docs/pur/purmain.htm.

HEXAZINONE	FORAGE HAY/ SILAGE	0.75	0.75	0.75	0.75
HEXAZINONE	UNCULTIVATED NONAG	1.19	1.53	1.53	1.53
HEXAZINONE	PASTURELAND	2.5	2.5	2.5	2.5
HEXAZINONE	RANGELAND	0.8	0.8	0.8	0.8

Some of the reported maximum rates are higher than labeled maximum rates for alfalfa, forest, Christmas trees and pasture. This may be due mis-reporting, misuse, uses no longer permitted by the label or to applications made on a less than per-acre basis.

2.5 Assessed Species

The CRLF was federally listed as a threatened species by USFWS effective June 24, 1996 (USFWS 1996). It is one of two subspecies of the red-legged frog and is the largest native frog in the western United States (USFWS 2002). A brief summary of information regarding CRLF distribution, reproduction, diet, and habitat requirements is provided in Sections 2.5.1 through 2.5.4, respectively. Further information on the status, distribution, and life history of and specific threats to the CRLF is provided in Attachment 1.

Final critical habitat for the CRLF was designated by USFWS on April 13, 2006 (USFWS 2006; 71 FR 19244-19346). Further information on designated critical habitat for the CRLF is provided in Section 2.6.

2.5.1 Distribution

The CRLF is endemic to California and Baja California (Mexico) and historically inhabited 46 counties in California including the Central Valley and both coastal and interior mountain ranges (USFWS 1996). Its range has been reduced by about 70%, and the species currently resides in 22 counties in California (USFWS 1996). The species has an elevational range of near sea level to 1,500 meters (5,200 feet) (Jennings and Hayes 1994); however, nearly all of the known CRLF populations have been documented below 1,050 meters (3,500 feet) (USFWS 2002).

Populations currently exist along the northern California coast, northern Transverse Ranges (USFWS 2002), foothills of the Sierra Nevada (5-6 populations), and in southern California south of Santa Barbara (two populations) (Fellers 2005a). Relatively larger numbers of CRLFs are located between Marin and Santa Barbara Counties (Jennings and Hayes 1994). A total of 243 streams or drainages are believed to be currently occupied by the species, with the greatest numbers in Monterey, San Luis Obispo, and Santa Barbara counties (USFWS 1996). Occupied drainages or watersheds include all bodies of water that support CRLFs (i.e., streams, creeks, tributaries, associated natural and artificial ponds, and adjacent drainages), and habitats through which CRLFs can move (i.e., riparian vegetation, uplands) (USFWS 2002).

The distribution of CRLFs within California is addressed in this assessment using four categories of location including recovery units, core areas, designated critical habitat, and known occurrences of the CRLF reported in the California Natural Diversity Database (CNDDB) that are not included within core areas and/or designated critical habitat (see Figure 2.1). Recovery units, core areas, and other known occurrences of the CRLF from the CNDDB are described in further detail in this section, and designated critical habitat is addressed in Section 2.6. Recovery units are large areas defined at the watershed level that have similar conservation needs and management strategies. The recovery unit is primarily an administrative designation, and land area within the recovery unit boundary is not exclusively CRLF habitat. Core areas are smaller areas within the recovery units that comprise portions of the species' historic and current range and have been determined by USFWS to be important in the preservation of the species. Designated critical habitat is generally contained within the core areas, although a number of critical habitat units are outside the boundaries of core areas, but within the boundaries of the recovery units. Additional information on CRLF occurrences from the CNDDB is used to cover the current range of the species not included in core areas and/or designated critical habitat, but within the recovery units.

Recovery Units

Eight recovery units have been established by USFWS for the CRLF. These areas are considered essential to the recovery of the species, and the status of the CRLF "may be considered within the smaller scale of the recovery units, as opposed to the statewide range" (USFWS 2002). Recovery units reflect areas with similar conservation needs and population statuses, and therefore, similar recovery goals. The eight units described for the CRLF are delineated by watershed boundaries defined by US Geological Survey hydrologic units and are limited to the elevational maximum for the species of 1,500 m above sea level. The eight recovery units for the CRLF are listed in Table 2.4 and shown in Figure 2.1.

Core Areas

USFWS has designated 35 core areas across the eight recovery units to focus their recovery efforts for the CRLF (Figure 2.1). Table 2.5 summarizes the geographical relationship among recovery units, core areas, and designated critical habitat. The core areas, which are distributed throughout portions of the historic and current range of the species, represent areas that allow for long-term viability of existing populations and reestablishment of populations within historic range. These areas were selected because they: 1) contain existing viable populations; or 2) they contribute to the connectivity of other habitat areas (USFWS 2002). Core area protection and enhancement are vital for maintenance and expansion of the CRLF's distribution and population throughout its range.

For purposes of this assessment, designated critical habitat, currently occupied (post-1985) core areas, and additional known occurrences of the CRLF from the CNDDB are considered. Each type of locational information is evaluated within the broader context of recovery units. For example, if no labeled uses of hexazinone occur (or if labeled uses occur at predicted exposures less than the Agency's LOCs) within an entire recovery unit, a "no effect" determination would be made for all designated critical habitat, currently occupied core areas, and other known CNDDB occurrences within that recovery unit. Historically occupied sections of the core areas are not evaluated as part of this assessment because the USFWS Recovery Plan (USFWS 2002) indicates that CRLFs are extirpated from these areas. A summary of currently and historically occupied core areas is provided in Table 2.4 (currently occupied core areas are bolded). While core areas are considered essential for recovery of the CRLF, core areas are not federally-designated critical habitat, although designated critical habitat is generally contained within these core recovery areas. It should be noted, however, that several critical habitat units are located outside of the core areas, but within the recovery units. The focus of this assessment is currently occupied core areas, designated critical habitat, and other known CNDDB CRLF occurrences within the recovery units. Federally-designated critical habitat for the CRLF is further explained in Section 2.6.

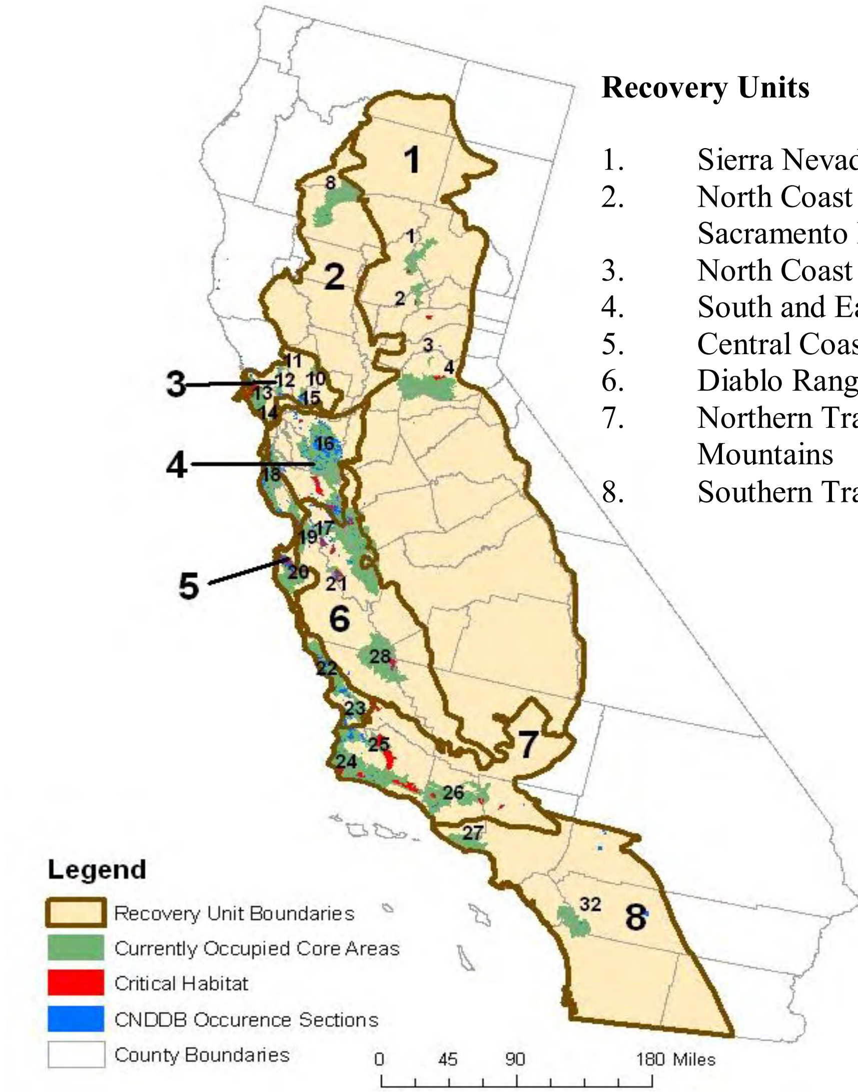
Table 2.4. California Red-legged Frog Recovery Units with Overlapping Core Areas and Designated Critical Habitat

Recovery Unit [1] (Figure 2.a)	**Core Areas [2,7] (Figure 2.1)**	**Critical Habitat Units [3]**	**Currently Occupied (post-1985) [4]**	**Historically Occupied [4]**
Sierra Nevada Foothills and Central Valley (1) (eastern boundary is the 1,500m elevation line)	**Cottonwood Creek (partial) (8)**	--	✓	
	Feather River (1)	BUT-1A-B	✓	
	Yuba River-S. Fork Feather River (2)	YUB-1	✓	
	--	NEV-1[6]		
	Traverse Creek/Middle Fork American River/Rubicon (3)	--	✓	
	Consumnes River (4)	ELD-1	✓	
	S. Fork Calaveras River (5)	--		✓
	Tuolumne River (6)	--		✓
	Piney Creek (7)	--		✓
	East San Francisco Bay (partial)(16)	--	✓	
North Coast Range Foothills and Western Sacramento River Valley (2)	**Cottonwood Creek (8)**	--	✓	
	Putah Creek-Cache Creek (9)	--		✓
	Jameson Canyon – Lower Napa Valley (partial) (15)	--	✓	
	Belvedere Lagoon (partial) (14)	-	✓	
	Pt. Reyes Peninsula (partial) (13)	--	✓	
North Coast and North San Francisco Bay (3)	Putah Creek-Cache Creek (partial) (9)			✓
	Lake Berryessa Tributaries (10)	NAP-1	✓	
	Upper Sonoma Creek (11)	--	✓	
	Petaluma Creek-Sonoma Creek (12)	--	✓	

	Pt. Reyes Peninsula (13)	MRN-1, MRN-2	✓	
	Belvedere Lagoon (14)	-	✓	
	Jameson Canyon-Lower Napa River (15)	SOL-1	✓	
South and East San Francisco Bay (4)	--	CCS-1A[6]		
	East San Francisco Bay (partial) (16)	ALA-1A, ALA-1B, STC-1B	✓	
	--	STC-1A[6]		
	South San Francisco Bay (partial) (18)	SNM-1A	✓	
Central Coast (5)	**South San Francisco Bay (partial) (18)**	SNM-1A, SNM-2C, SCZ-1	✓	
	Watsonville Slough- Elkhorn Slough (partial) (19)	SCZ-2 [5]	✓	
	Carmel River-Santa Lucia (20)	MNT-2	✓	
	Estero Bay (22)	--	✓	
	--	SLO-8[6]		
	Arroyo Grande Creek (23)	-	✓	
	Santa Maria River-Santa Ynez River (24)	--	✓	
Diablo Range and Salinas Valley (6)	**East San Francisco Bay (partial) (16)**	MER-1A-B, STC-1B	✓	
	--	SNB-1[6], SNB-2[6]		
	Santa Clara Valley (17)	--	✓	
	Watsonville Slough- Elkhorn Slough (partial)(19)	MNT-1	✓	
	Carmel River-Santa Lucia (partial)(20)	--	✓	
	Gablan Range (21)	SNB-3	✓	
	Estrella River (28)	SLO-1A-B	✓	
Northern Transverse Ranges and Tehachapi Mountains (7)	--	SLO-8[6]		
	Santa Maria River-Santa Ynez River (24)	STB-4, STB-5, STB-7	✓	
	Sisquoc River (25)	STB-1, STB-3	✓	
	Ventura River-Santa Clara River (26)	VEN-1, VEN-2, VEN-3	✓	
	--	LOS-1[6]		
Southern Transverse and Peninsular Ranges (8)	**Santa Monica Bay-Ventura Coastal Streams (27)**	--	✓	
	San Gabriel Mountain (29)	--		✓
	Forks of the Mojave (30)	--		✓
	Santa Ana Mountain (31)	--		✓
	Santa Rosa Plateau (32)	--	✓	
	San Luis Rey (33)	--		✓
	Sweetwater (34)	--		✓
	Laguna Mountain (35)	--		✓

[1] Recovery units designated by the USFWS (USFWS 2000, pg 49).
Core areas designated by the USFWS (USFWS 2000, pg 51).
Critical habitat units designated by the USFWS on April 13, 2006 (USFWS 2006, 71 FR 19244-19346).
Currently occupied (post-1985) and historically occupied core areas as designated by the USFWS (USFWS 2002, pg 54).

Critical habitat unit where identified threats specifically included pesticides or agricultural runoff (USFWS 2002).
Critical habitat units that are outside of core areas, but within recovery units.
[7] Currently occupied core areas that are included in this effects determination are bolded.



Recovery Units

1. Sierra Nevada Foothills and Central Valley
2. North Coast Range Foothills and Western Sacramento River Valley
3. North Coast and North San Francisco Bay
4. South and East San Francisco Bay
5. Central Coast
6. Diablo Range and Salinas Valley
7. Northern Transverse Ranges and Tehachapi Mountains
8. Southern Transverse and Peninsular Ranges

Core Areas

1Feather River
2Yuba River- S. Fork Feather River
3Traverse Creek/ Middle Fork/ American R. Rubicon
4Cosumnes River
5South Fork Calaveras River*
6Tuolumne River*
Piney Creek*
8Cottonwood Creek
9Putah Creek – Cache Creek*
10Lake Berryessa Tributaries
11Upper Sonoma Creek
12Petaluma Creek – Sonoma Creek
13Pt. Reyes Peninsula
14Belvedere Lagoon
15Jameson Canyon – Lower Napa River
16East San Francisco Bay
17Santa Clara Valley
18South San Francisco Bay
19Watsonville Slough-Elkhorn Slough
20Carmel River – Santa Lucia
2Gablan Range
2Estero Bay
2Arroyo Grange River
2Santa Maria River – Santa Ynez River
2Sisquoc River
26Ventura River – Santa Clara River
27Santa Monica Bay – Venura Coastal Streams
28Estrella River
29San Gabriel Mountain*
30Forks of the Mojave*
31Santa Ana Mountain*
32Santa Rosa Plateau
33San Luis Ray*
34Sweetwater*
35Laguna Mountain*

Core areas that were historically occupied by the California red-legged frog are not included in the map

Figure 2.1. Recovery Unit, Core Area, Critical Habitat, and Occurrence Designations for CRLF

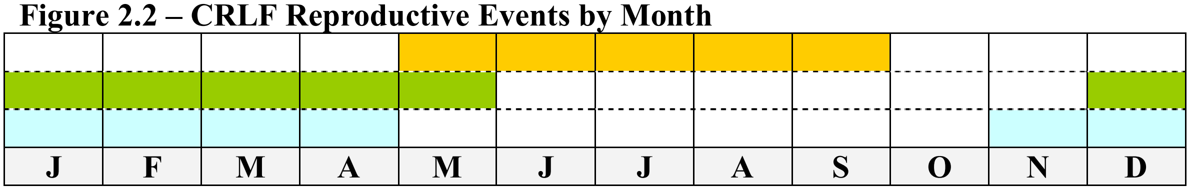
Other Known Occurrences from the CNDBB

The CNDDB provides location and natural history information on species found in California. The CNDDB serves as a repository for historical and current species location sightings. Information regarding known occurrences of CRLFs outside of the currently occupied core areas and designated critical habitat is considered in defining the current range of the CRLF. See: http://www.dfg.ca.gov/bdb/html/cnddb_info.html for additional information on the CNDDB.

2.5.2 Reproduction

CRLFs breed primarily in ponds; however, they may also breed in quiescent streams, marshes, and lagoons (Fellers 2005a). According to the Recovery Plan (USFWS 2002), CRLFs breed from November through late April. Peaks in spawning activity vary geographically; Fellers (2005b) reports peak spawning as early as January in parts of coastal central California. Eggs are fertilized as they are being laid. Egg masses are typically attached to emergent vegetation, such as bulrushes (*Scirpus* spp.) and cattails (*Typha* spp.) or roots and twigs, and float on or near the surface of the water (Hayes and Miyamoto 1984). Egg masses contain approximately 2000 to 6000 eggs ranging in size between 2 and 2.8 mm (Jennings and Hayes 1994). Embryos hatch 10 to 14 days after fertilization (Fellers 2005a) depending on water temperature. Egg predation is reported to be infrequent and most mortality is associated with the larval stage (particularly through predation by fish); however, predation on eggs by newts has also been reported (Rathburn 1998). Tadpoles require 11 to 28 weeks to metamorphose into juveniles (terrestrial-phase), typically between May and September (Jennings and Hayes 1994, USFWS 2002); tadpoles have been observed to over-winter (delay metamorphosis until the following year) (Fellers 2005b, USFWS 2002). Males reach sexual maturity at 2 years, and females reach sexual maturity at 3 years of age; adults have been reported to live 8 to 10 years (USFWS 2002). Figure 2.2 depicts CRLF annual reproductive timing.

Figure 2.2 – CRLF Reproductive Events by Month



Light Blue = **Breeding/Egg Masses**
Green = **Tadpoles (except those that over-winter)**
Orange = **Young Juveniles**
Adults and juveniles can be present all year

2.5.3 Diet

Although the diet of CRLF aquatic-phase larvae (tadpoles) has not been studied specifically, it is assumed that their diet is similar to that of other frog species, with the aquatic phase feeding exclusively in water and consuming diatoms, algae, and detritus (USFWS 2002). Tadpoles filter and entrap suspended algae (Seale and Beckvar, 1980) via mouthparts designed for effective grazing of periphyton (Wassersug, 1984, Kupferberg *et al.*; 1994; Kupferberg, 1997; Altig and McDiarmid, 1999).

Juvenile and adult CRLFs forage in aquatic and terrestrial habitats, and their diet differs greatly from that of larvae. The main food source for juvenile aquatic- and terrestrial-phase CRLFs is thought to be aquatic and terrestrial invertebrates found along the shoreline and on the water surface. Hayes and Tennant (1985) report, based on a study examining the gut content of 35 juvenile and adult CRLFs, that the species feeds on as many as 42 different invertebrate taxa, including Arachnida, Amphipoda, Isopoda, Insecta, and Mollusca. The most commonly observed prey species were larval alderflies (*Sialis* cf. *californica*), pillbugs (*Armadilliadrium vulgare*), and water striders (*Gerris* sp). The preferred prey species, however, was the sowbug (Hayes and Tennant, 1985). This study suggests that CRLFs forage primarily above water, although the authors note other data reporting that adults also feed under water, are cannibalistic, and consume fish. For larger CRLFs, over 50% of the prey mass may consists of vertebrates such as mice, frogs, and fish, although aquatic and terrestrial invertebrates were the most numerous food items (Hayes and Tennant 1985). For adults, feeding activity takes place primarily at night; for juveniles feeding occurs during the day and at night (Hayes and Tennant 1985).

2.5.4 Habitat

CRLFs require aquatic habitat for breeding, but also use other habitat types including riparian and upland areas throughout their life cycle. CRLF use of their environment varies; they may complete their entire life cycle in a particular habitat or they may utilize multiple habitat types. Overall, populations are most likely to exist where multiple breeding areas are embedded within varying habitats used for dispersal (USFWS 2002). Generally, CRLFs utilize habitat with perennial or near-perennial water (Jennings et al. 1997). Dense vegetation close to water, shading, and water of moderate depth are habitat features that appear especially important for CRLF (Hayes and Jennings 1988).

Breeding sites include streams, deep pools, backwaters within streams and creeks, ponds, marshes, sag ponds (land depressions between fault zones that have filled with water), dune ponds, and lagoons. Breeding adults have been found near deep (0.7 m) still or slow moving water surrounded by dense vegetation (USFWS 2002); however, the largest number of tadpoles have been found in shallower pools (0.26 – 0.5 m) (Reis, 1999). Data indicate that CRLFs do not frequently inhabit vernal pools, as conditions in these habitats generally are not suitable (Hayes and Jennings 1988).

CRLFs also frequently breed in artificial impoundments such as stock ponds, although additional research is needed to identify habitat requirements within artificial ponds (USFWS 2002). Adult CRLFs use dense, shrubby, or emergent vegetation closely associated with deep-water pools bordered with cattails and dense stands of overhanging vegetation (http://www.fws.gov/endangered/features/rl_frog/rlfrog.html#where).

In general, dispersal and habitat use depends on climatic conditions, habitat suitability, and life stage. Adults rely on riparian vegetation for resting, feeding, and dispersal. The foraging quality of the riparian habitat depends on moisture, composition of the plant community, and presence of pools and backwater aquatic areas for breeding. CRLFs can be found living within streams at distances up to 3 km (2 miles) from their breeding site and have been found up to 30 m (100 feet) from water in dense riparian vegetation for up to 77 days (USFWS 2002).

During dry periods, the CRLF is rarely found far from water, although it will sometimes disperse from its breeding habitat to forage and seek other suitable habitat under downed trees or logs, industrial debris, and agricultural features (UWFWS 2002). According to Jennings and Hayes (1994), CRLFs also use small mammal burrows and moist leaf litter as habitat. In addition, CRLFs may also use large cracks in the bottom of dried ponds as refugia; these cracks may provide moisture for individuals avoiding predation and solar exposure (Alvarez 2000).

2.6 Designated Critical Habitat

In a final rule published on April 13, 2006, 34 separate units of critical habitat were designated for the CRLF by USFWS (USFWS 2006; FR 51 19244-19346). A summary of the 34 critical habitat units relative to USFWS-designated recovery units and core areas (previously discussed in Section 2.5.1) is provided in Table 2.5.

'Critical habitat' is defined in the ESA as the geographic area occupied by the species at the time of the listing where the physical and biological features necessary for the conservation of the species exist, and there is a need for special management to protect the listed species. It may also include areas outside the occupied area at the time of listing if such areas are 'essential to the conservation of the species.' All designated critical habitat for the CRLF was occupied at the time of listing. Critical habitat receives protection under Section 7 of the ESA through prohibition against destruction or adverse modification with regard to actions carried out, funded, or authorized by a federal Agency. Section 7 requires consultation on federal actions that are likely to result in the destruction or modification of critical habitat.

To be included in a critical habitat designation, the habitat must be 'essential to the conservation of the species.' Critical habitat designations identify, to the extent known using the best scientific and commercial data available, habitat areas that provide essential life cycle needs of the species or areas that contain certain primary constituent elements (PCEs) (as defined in 50 CFR 414.12(b)). PCEs include, but are not limited to, space for individual and population growth and for normal behavior; food, water, air, light, minerals, or other nutritional or physiological requirements; cover or shelter; sites for breeding, reproduction, rearing (or development) of offspring; and habitats that are protected from disturbance or are representative of the historic geographical and ecological distributions of a species. The designated critical habitat areas for the CRLF are considered to have the following PCEs that justify critical habitat designation:

- Breeding aquatic habitat;
- Non-breeding aquatic habitat;
- Upland habitat; and
- Dispersal habitat.

Please note that a more complete description of these habitat types is provided in Attachment 1.

Occupied habitat may be included in the critical habitat only if essential features within the habitat may require special management or protection. Therefore, USFWS does not include areas where existing management is sufficient to conserve the species. Critical habitat is designated outside the geographic area presently occupied by the species only when a designation limited to its present range would be inadequate to ensure the conservation of the species. For the CRLF, all designated critical habitat units contain all four of the PCEs, and were occupied by the CRLF at the time of FR listing notice in April 2006. The FR notice designating critical habitat for the CRLF includes a special rule exempting routine ranching activities associated with livestock ranching from incidental take prohibitions. The purpose of this exemption is to promote the conservation of rangelands, which could be beneficial to the CRLF, and to reduce the rate of conversion to other land uses that are incompatible with CRLF conservation. Please see Attachment 1 for a full explanation on this special rule.

USFWS has established modification standards for designated critical habitat (USFWS 2006). Activities that may destroy or modify critical habitat are those that alter the PCEs and jeopardize the continued existence of the species. Evaluation of actions related to use of hexazinone that may alter the PCEs of the CRLF's critical habitat form the basis of the critical habitat impact analysis. According to USFWS (2006), activities that may affect critical habitat and therefore result in adverse effects to the CRLF include, but are not limited to the following:

(1) Significant alteration of water chemistry or temperature to levels beyond the tolerances of the CRLF that result in direct or cumulative adverse effects to individuals and their life-cycles.

(2) Significant increase in sediment deposition within the stream channel or pond or disturbance of upland foraging and dispersal habitat that could result in elimination or reduction of habitat necessary for the growth and reproduction of the CRLF by increasing the sediment deposition to levels that would adversely affect their ability to complete their life cycles.

(3) Significant alteration of channel/pond morphology or geometry that may lead to changes to the hydrologic functioning of the stream or pond and alter the timing, duration, water flows, and levels that would degrade or eliminate the CRLF and/or its habitat. Such an effect could also lead to increased sedimentation and degradation in water quality to levels that are beyond the CRLF's tolerances.

(4) Elimination of upland foraging and/or aestivating habitat or dispersal habitat.

(5) Introduction, spread, or augmentation of non-native aquatic species in stream segments or ponds used by the CRLF.

(6) Alteration or elimination of the CRLF's food sources or prey base (also evaluated as indirect effects to the CRLF).

As previously noted in Section 2.1, the Agency believes that the analysis of direct and indirect effects to listed species provides the basis for an analysis of potential effects on the designated critical habitat. Because hexazinone is expected to directly impact living organisms within the action area, critical habitat analysis for hexazinone is limited in a practical sense to those PCEs of critical habitat that are biological or that can be reasonably linked to biologically mediated processes.

2.7 Action Area

For listed species assessment purposes, the action area is considered to be the area affected directly or indirectly by the federal action and not merely the immediate area involved in the action (50 CFR 402.02). It is recognized that the overall action area for the national registration of hexazinone is likely to encompass considerable portions of the United States based on the large array of agricultural and non-agricultural uses. However, the scope of this assessment limits consideration of the overall action area to those portions that may be applicable to the protection of the CRLF and its designated critical habitat within the state of California. Deriving the geographical extent of this portion of the action area is the product of consideration of the types of effects that hexazinone may be expected to have on the environment, the exposure levels to hexazinone that are associated with those effects, and the best available information concerning the use of hexazinone and its fate and transport within the state of California.

The definition of action area requires a stepwise approach that begins with an understanding of the federal action. The federal action is defined by the currently labeled uses for hexazinone. An analysis of labeled uses and review of available product labels was completed. This analysis indicates that, for hexazinone, the following uses are considered as part of the federal action evaluated in this assessment:

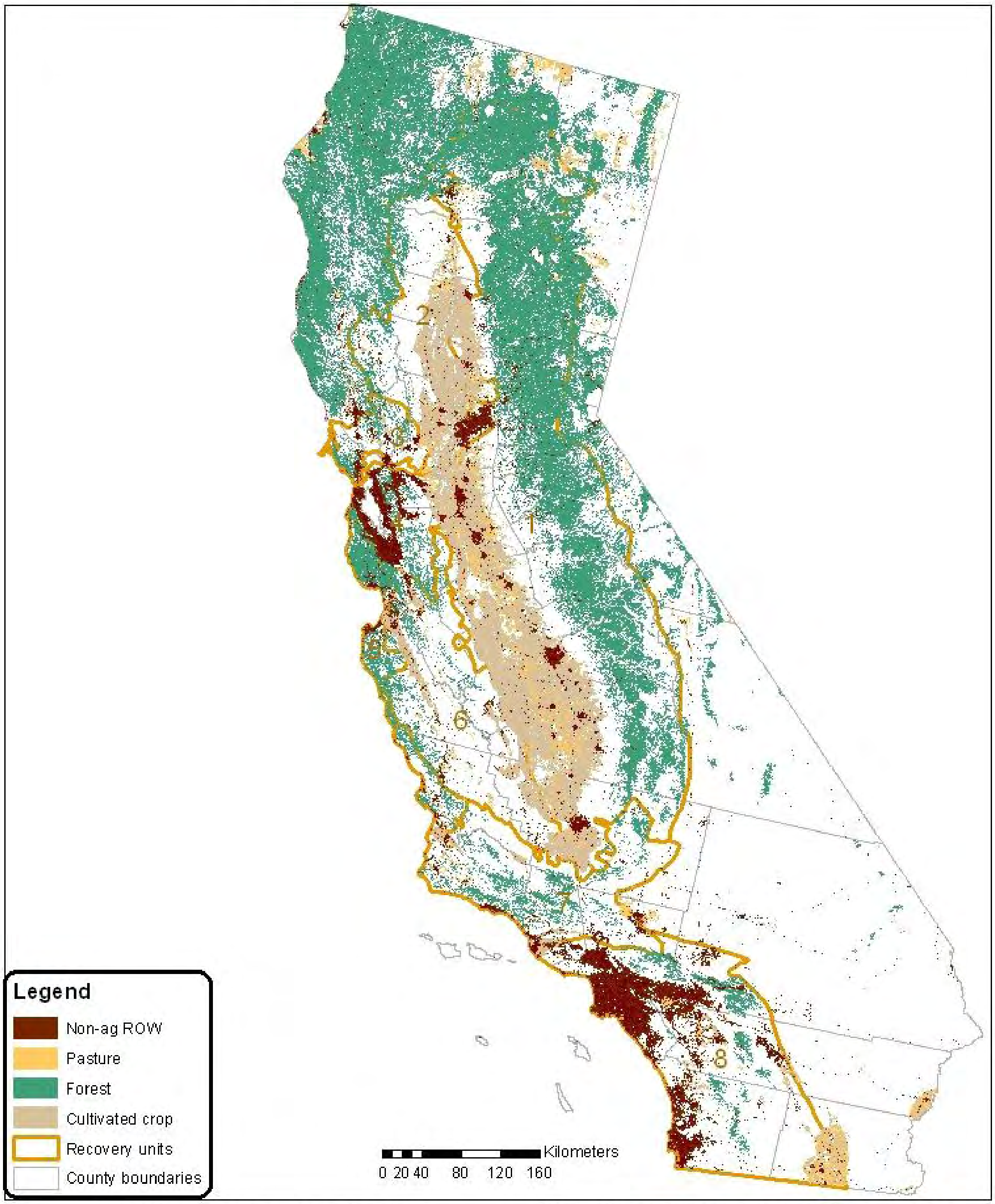
- Airports
- Industrial (outdoor)

- Sewage disposal
- Drainage
- Alfalfa
- Blueberries
- Pineapple
- Christmas Tree Plantations
- Forestry
- Rangeland
- Agricultural Rights-of-Way
- Non-Agricultural Rights-of-Way
- Pastures
- Rangeland
- Agricultural Uncultivated Areas
- Non- Agricultural Uncultivated Areas

After a determination of which uses will be assessed, an evaluation of the potential "footprint" of the use pattern should be determined. This "footprint" represents the initial area of concern and is typically based on available land cover data. Local land cover data available for the state of California were analyzed to refine the understanding of potential hexazinone use. The initial area of concern is defined as all land cover types that represent the labeled uses described above. A map representing all the land cover types that make up the initial area of concern is presented in Fig 2.3.Sugar cane use patterns were excluded from our assessment because itis not grown in California. No areas are excluded from the final action area based on usage and land cover data.

Fig. 2.3 Hexazinone Initial Area of Concern

Hexazinone Initial Area of Concern



Compiled from California County boundaries (ESRI, 2002), USDA National Agriculture Statistical Service (NASS, 2002) Gap Analysis Program Orchard/ Vineyard Landcover (GAP) National Land Cover Database (NLCD) (MRLC, 2001)

Map created by US Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983).

Produced: 1/23/2008

Once the initial area of concern is defined, the next step is to compare the extent of that area with the results of the screening level risk assessment. The screening level risk assessment will define which taxa, if any, are predicted to be exposed at concentrations above the Agency's Levels of Concern (LOC). The screening level assessment includes an evaluation of the environmental fate properties of hexazinone to determine which routes of transport are likely to have an impact on the CRLF.

For hexazinone, the principal routes of transport away from the application site are expected to be runoff and spray drift due to its mobility and moderate persistence. Furthermore, the vapor pressure of hexazinone suggests that volatilization leading to long-range transport is unlikely.

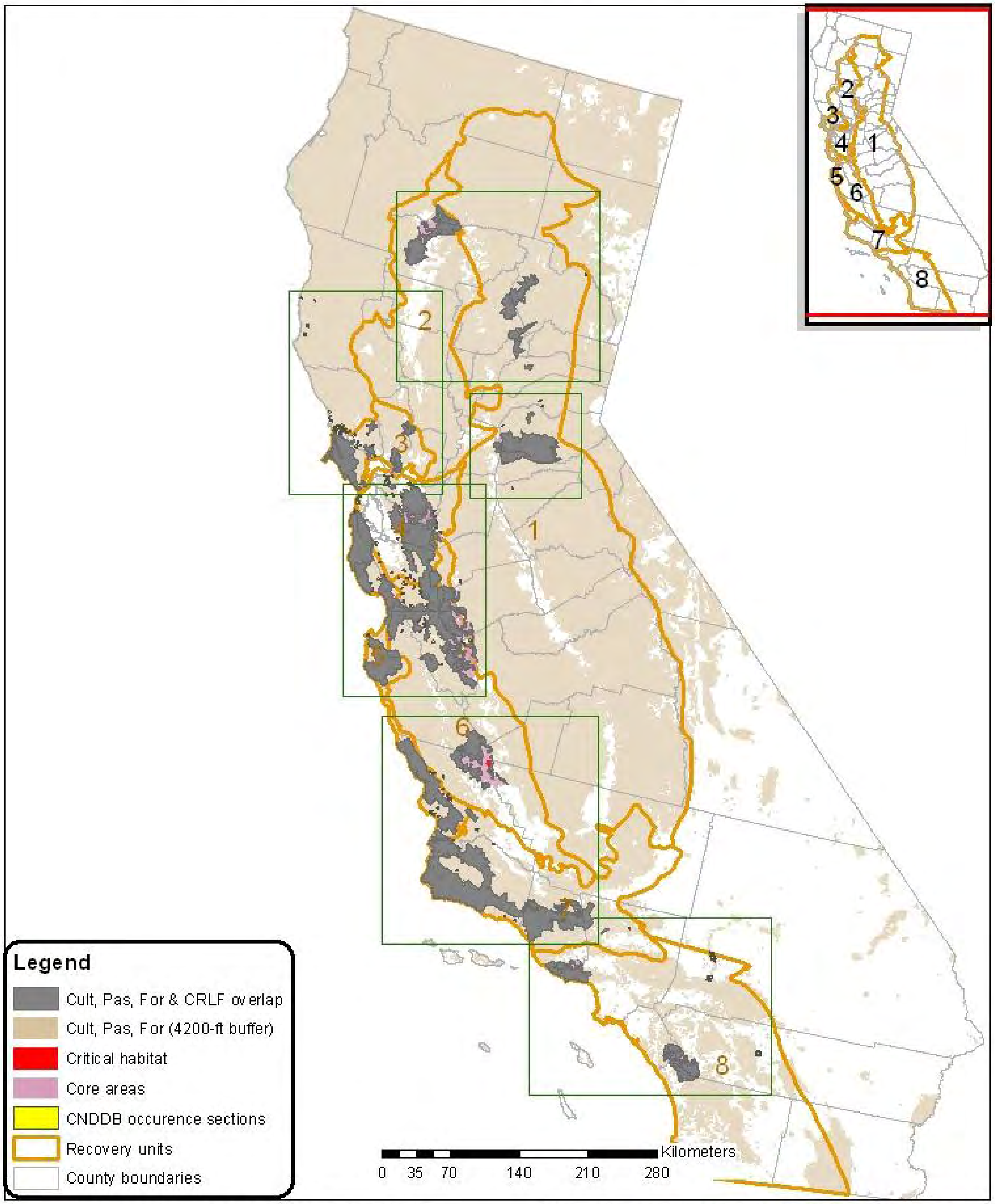
LOC exceedances are used to describe how far effects may be seen from the initial area of concern. Factors considered include: spray drift, downstream run-off, atmospheric transport, etc. This information is incorporated into GIS and a map of the action area is created.

The AgDRIFT model (Version 2.01) is used to define how far from the initial area of concern an effect to a given terrestrial species may be expected. The spray drift analysis for hexazinone using the most sensitive terrestrial toxicity endpoint, terrestrial plants, suggests that the distance for potential effects from the treated area of concern is beyond the range of the AgDRIFT model (*i.e.*, 1000 feet). Subsequently, the AgDISP model (Version 8.15) with the Gaussian extension (used for longer range transport because the limits of the regular AgDISP model were exceeded) was used to define this distance. The AgDISP model was run in ground mode using default settings (except for wind speed at 10 mph and release height at 4 feet). Using the Gaussian extension, a maximum spray drift distance of 4,501 feet for the listed species LOC was derived. The maximum spray drift distance of 3,366 feet for relevant portion of the action area is based on the non-listed species LOC for ground applications. A maximum spray drift distance of 4,200 feet for the listed species LOC was derived for the aerial application. The maximum spray drift distance of 3,589 feet for relevant portion of the action area is based on the non-listed species LOC for aerial applications. Further detail on the spray drift analysis is provided in Section 3.2.5.

In addition to the buffered area from the spray drift analysis, the final action area also considers the downstream extent of hexazinone that exceeds the LOC (discussed in Section 3.2.6). It should be noted that the action area for hexazinone is based on the endangered species LOCs for aquatic and terrestrial plants. However, the portion of the action area that is relevant to the CRLF is based on the non-listed species LOCs for aquatic and terrestrial plants because the CRLF does not have an obligate relationship with plants.

The action area including the overlap between the CRLF and cultivated crop, pasture and forest uses (Fig. 2.4) and non-crop non-agriacultural ROW (Fig. 2.5) uses is depicted in two separate maps to increase the clarity of the maps.

Hexazinone Cultivated crop, Pasture, Forest Action Area & CRLF overlap

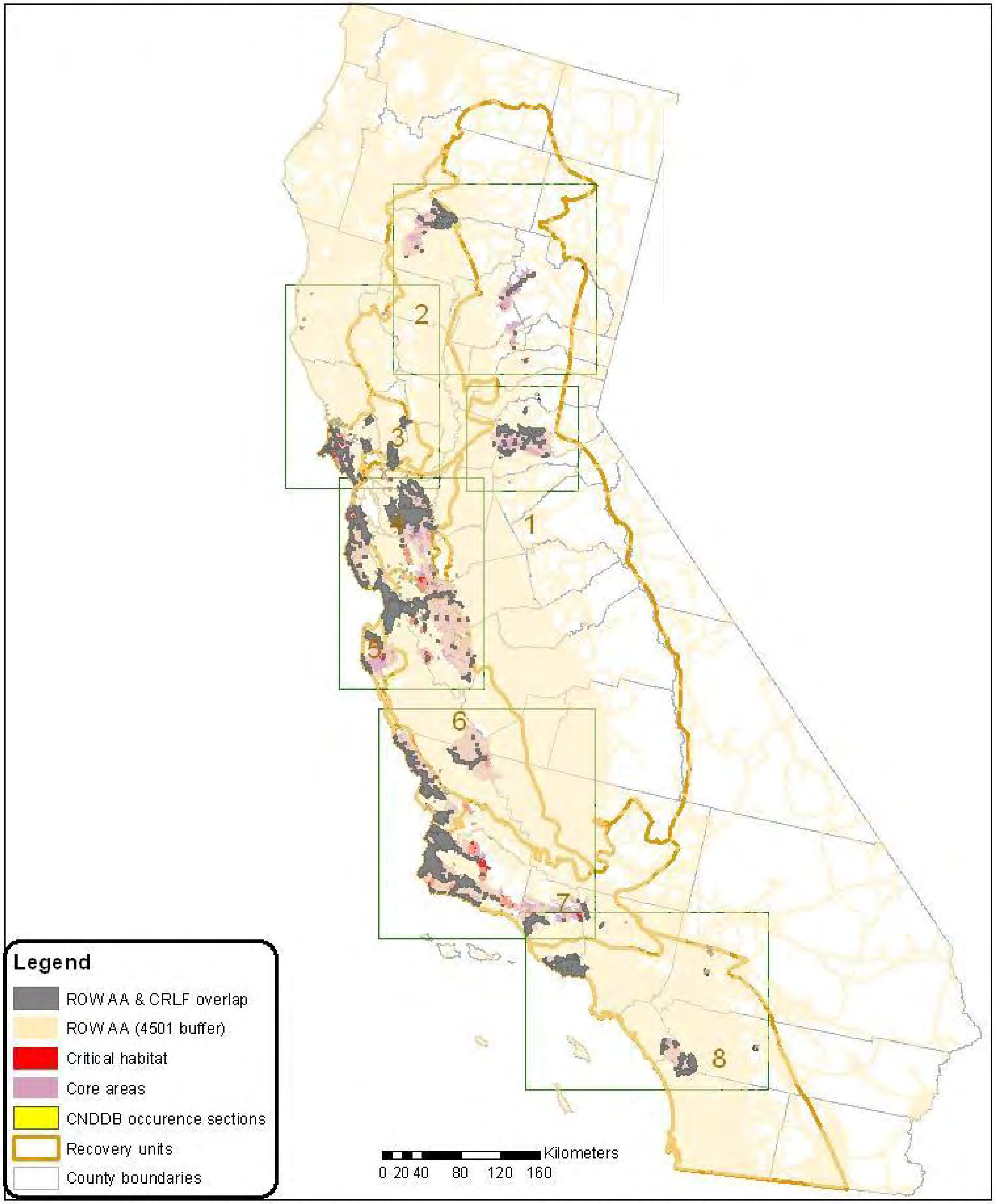


Compiled from California County boundaries (ESRI, 2002), USDA National Agriculture Statistical Service (NASS, 2002) Gap Analysis Program Orchard/ Vineyard Landcover (GAP) National Land Cover Database (NLCD) (MRLC, 2001)

Map created by US Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983).

Produced: 1/23/2008

Hexazinone Rights-of-Way (ROW) Action Area & CRLF Overlap



Compiled from California County boundaries (ESRI, 2002), USDA National Agriculture Statistical Service (NASS, 2002) Gap Analysis Program Orchard/ Vineyard Landcover (GAP) National Land Cover Database (NLCD) (MRLC, 2001)

Map created by US Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983).

Produced: 1/23/2008

Subsequent to defining the action area, an evaluation of usage information was conducted to determine the area where use of hexazinone may impact the CRLF. This analysis is used to characterize where predicted exposures are most likely to occur but does not preclude use in other portions of the action area. A more detailed review of the county-level use information was also completed. These data suggest that hexazinone has historically been used on a wide variety of agricultural and non-agricultural use sites. Further information regarding buffer analysis and down stream dilution analysis used to define the action area may be found in sections 3.2.5 and 3.2.6, respectively.

2.8 Assessment Endpoints and Measures of Ecological Effect

Assessment endpoints are defined as "explicit expressions of the actual environmental value that is to be protected."[4] Selection of the assessment endpoints is based on valued entities (e.g., CRLF, organisms important in the life cycle of the CRLF, and the PCEs of its designated critical habitat), the ecosystems potentially at risk (e.g,. waterbodies, riparian vegetation, and upland and dispersal habitats), the migration pathways of hexazinone (e.g., runoff, spray drift, etc.), and the routes by which ecological receptors are exposed to hexazinone-related contamination (e.g., direct contact, etc).

2.8.1 Assessment Endpoints for the CRLF

Assessment endpoints for the CRLF include direct toxic effects on the survival, reproduction, and growth of the CRLF, as well as indirect effects, such as reduction of the prey base and/or modification of its habitat. In addition, potential destruction and/or modification of critical habitat is assessed by evaluating potential effects to PCEs, which are components of the habitat areas that provide essential life cycle needs of the CRLF. Each assessment endpoint requires one or more "measures of ecological effect," defined as changes in the attributes of an assessment endpoint or changes in a surrogate entity or attribute in response to exposure to a pesticide. Specific measures of ecological effect are generally evaluated based on acute and chronic toxicity information from registrant-submitted guideline tests that are performed on a limited number of organisms. Additional ecological effects data from the open literature are also considered.

A complete discussion of all the toxicity data available for this risk assessment, including resulting measures of ecological effect selected for each taxonomic group of concern, is included in Section 4 of this document. A summary of the assessment endpoints and measures of ecological effect selected to characterize potential assessed direct and indirect CRLF risks associated with exposure to hexazinone is provided in Table 2.5.

Table 2.5 Summary of Assessment Endpoints and Measures of Ecological Effects for Direct and Indirect Effects of hexazinone on the California Red-legged Frog

Assessment Endpoint	Measures of Ecological Effects (Data Sources Reviewed)	Specific Selected Toxicity Value (basis)
Aquatic Phase (eggs, larvae, tadpoles, juveniles, and adults)[a]		
1. Survival, growth, and reproduction of CRLF	1a. Most sensitive fish acute LC_{50}	1a. Fathead minnow 274 mg/L

[4] From U.S. EPA (1992). *Framework for Ecological Risk Assessment.* EPA/630/R-92/001.

Table 2.5 Summary of Assessment Endpoints and Measures of Ecological Effects for Direct and Indirect Effects of hexazinone on the California Red-legged Frog		
Assessment Endpoint	**Measures of Ecological Effects (Data Sources Reviewed)**	**Specific Selected Toxicity Value (basis)**
individuals via direct effects on aquatic phases	1b. Most sensitive fish chronic NOAEC	1b. Fathead minnow early-life stage NOAEL = 17mg/Lai
	1c. Most sensitive fish early-life stage NOAEC	1c. same as 1b.
2. Survival, growth, and reproduction of CRLF individuals via effects to food supply (*i.e.,* freshwater invertebrates, non-vascular plants)	2a. Most sensitive (1) fish, (2) aquatic invertebrate, and (3) aquatic plant EC_{50} or LC_{50}	2a1. Fathead minnow 274 mg/L ai
		2a2. *Daphnia magna* 48-hr EC_{50} = 110 mg/L ai
		2a3. Green algae 96-hr EC_{50} = 0.007 mg/L ai
	2b. Most sensitive (1) aquatic invertebrate and (2) fish chronic NOAEC	2b1. *Daphnia magna* reproduction NOAEC = 20 mg/Lai
		2b2. Fathead minnow NOAEL = 17 mg/L ai
3. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat, cover, and/or primary productivity (*i.e.,* aquatic plant community)	3a. Vascular plant acute EC_{50}	3a. Lemna gibba 14-d EC50 = 0.0374 mg/l ai
	3b. Non-vascular plant acute EC_{50} (freshwater algae	3b. Green Algae 96-hr EC_{50} = 0.007 mg/l ai
4. Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation, required to maintain acceptable water quality and habitat in ponds and streams comprising the species' current range.	4a. Distribution of (1) seedling emergence and (2) vegetative vigor monocot EC_{25} values	4a1. Sorghum seedling emergence 21-d EC_{25} = 0.019 lbs ai/A
		4a2. Wheat vegetative vigor 21-d EC_{25} = 0.02 lbs ai/A
	4b. Distribution of (1) seedling emergence and (2) vegetative vigor EC_{25} values for dicots	4b1. Tomato seedling emergence 21-d EC_{25}= 0.0064 lbs ai/A
		4b2. Rape vegetative vigor 21-d EC_{25}= 0.011 lbs ai/A
Terrestrial Phase (Juveniles and adults)		
5. Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	5a. Most sensitive bird[2] acute LC_{50} or LD_{50}	5a. Mallard duck LD_{50} = 2258 mg ai/kg-bw
	5b. Most sensitive bird[2] chronic NOAEC	5b. Bobwhite quail reproduction NOAEC = 100 ppm
6. Survival, growth, and reproduction of CRLF individuals via effects on prey (*i.e.,*terrestrial invertebrates, small terrestrial vertebrates, including mammals and terrestrial phase amphibians)	6a. Most sensitive terrestrial (1) invertebrate and (2) vertebrate acute EC_{50} or LC_{50}	6a1. Honey bee acute contact LD_{50} >100 µg ai/bee
		6a2. Rat LD_{50} = 530 mg ai/kg-bw
	6b. Most sensitive terrestrial (1) invertebrate and (2) vertebrate chronic NOAEC	6b1. No chronic NOAEC data for terrestrial invertebrates
		6b2. Rat 2-generation reproduction study NOAEL = 200 mg/kg-bw

Table 2.5 Summary of Assessment Endpoints and Measures of Ecological Effects for Direct and Indirect Effects of hexazinone on the California Red-legged Frog		
Assessment Endpoint	**Measures of Ecological Effects (Data Sources Reviewed)**	**Specific Selected Toxicity Value (basis)**
7. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.,* riparian vegetation)	7a. Distribution of (1) seedling emergence and (2) vegetative vigor EC_{25} values for monocots	7a1. Sorghum seedling emergence 21-d EC_{25} = 0.019 lbs ai/A
		7a2. Wheat vegetative vigor 21-d EC_{25} = 0.02 lbs ai/A
	7b. Distribution of (1) seedling emergence and (2) vegetative vigor EC_{25} values for dicots	7b1. Tomato seedling emergence 21-d EC_{25} = 0.0064 lbs ai/A
		7b2. Rape vegetative vigor 21-d EC_{25} = 0.011 lbs ai/
[1] Adult frogs are no longer in the "aquatic phase" of the amphibian life cycle; however, submerged adult frogs are considered "aquatic" for the purposes of this assessment because exposure pathways in the water are considerably different than exposure pathways on land. [2] Birds are used as surrogates for terrestrial phase amphibians.		

2.8.2 Assessment Endpoints for Designated Critical Habitat

As previously discussed, designated critical habitat is assessed to evaluate actions related to the use of hexazinone that may alter the PCEs of the CRLF's critical habitat. PCEs for the CRLF were previously described in Section 2.6. Actions that may destroy or modify critical habitat are those that alter the PCEs. Therefore, these actions are identified as assessment endpoints. It should be noted that evaluation of PCEs as assessment endpoints is limited to those of a biological nature (i.e., the biological resource requirements for the listed species associated with the critical habitat) and those for which hexazinone effects data are available.

Assessment endpoints and measures of ecological effect selected to characterize potential modification to designated critical habitat associated with exposure to hexazinone are provided in Table 2.6. Modification to the critical habitat of the CRLF includes the following, as specified by USFWS (2006) and previously discussed in Section 2.6:

1. Alteration of water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs.

2. Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs.

3. Significant increase in sediment deposition within the stream channel or pond or disturbance of upland foraging and dispersal habitat.

4. Significant alteration of channel/pond morphology or geometry.

5. Elimination of upland foraging and/or aestivating habitat, as well as dispersal habitat.

6. Introduction, spread, or augmentation of non-native aquatic species in stream segments or ponds used by the CRLF.

7. Alteration or elimination of the CRLF's food sources or prey base.

Measures of such possible effects by labeled use of hexazinone on critical habitat of the CRLF are described in Table 2.6. Some components of these PCEs are associated with physical abiotic features (e.g., presence and/or depth of a water body, or distance between two sites), which are not expected to be measurably altered by use of pesticides. Assessment endpoints used for the analysis of designated critical habitat are based on the modification standard established by USFWS (2006).

Table 2.6. Summary of Assessment Endpoints and Measures of Ecological Effect for Primary Constituent Elements of Designated Critical Habitat

Assessment Endpoint	Measures of Ecological Effect[5] (Data Sources Reviewed)	Specific Selected Toxicity Value (basis)
	Aquatic Phase PCEs (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)	
Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.	a. Most sensitive aquatic plant EC_{50}	a. Green Algae 96-hr EC_{50} = 0.007 ppm ai
	b. Distribution of terrestrial monocot (1) seedling emergence and (2) vegetative vigor EC_{25}	b1. Sorghum seedling emergence 21-d EC_{25} = 0.019 lbs ai/A
		b2. Wheat vegetative vigor 21-d EC_{25} = 0.02 lbs ai/A
	c. Distribution of terrestrial dicot (1) seedling emergence and (2) vegetative vigor EC_{25} values	c1. Tomato seedling emergence 21-d EC_{25} = 0.0064 lbs ai/A
		c2. Rape vegetative vigor 21-d EC_{25} = 0.011 lbs ai/A
Alteration in water chemistry/quality including temperature, turbidity, and oxygen	a. Most sensitive EC_{50} values for aquatic	a. Green Algae 96-hr EC_{50} = 0.007 mg/L ai

[5] All toxicity data reviewed for this assessment are included in Appendices D through F.

Table 2.6. Summary of Assessment Endpoints and Measures of Ecological Effect for Primary Constituent Elements of Designated Critical Habitat		
Assessment Endpoint	**Measures of Ecological Effect[5] (Data Sources Reviewed)**	**Specific Selected Toxicity Value (basis)**
content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.[6]	b. Distribution of terrestrial monocot (1) seedling emergence and (2) vegetative vigor EC_{25} values	b1. Sorghum seedling emergence 21-d EC_{25} = 0.019 lbs ai/A
		b2. Wheat vegetative vigor 21-d EC_{25} = 0.02 lbs ai/A
	c. Distribution of terrestrial dicot (1) seedling emergence and (2) vegetative vigor EC_{25} values	c1. Tomato seedling emergence 21-d EC_{25} = 0.0064 lbs ai/A
		c2. Rape vegetative vigor 21-d EC_{25} = 0.011 lbs ai/A
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	a. Most sensitive EC_{50} or LC_{50} values for (1) fish or aquatic-phase amphibians and (2) aquatic invertebrates	a1. Fathead minnow 274 mg/L ai
		a2. *Daphnia magna* 48-hr EC_{50} = 110 mg/L ai
	b. Most sensitive reproductive NOAEC values for (1) fish or aquatic-phase amphibians and (2) aquatic invertebrates	b1. Fathead minnow early-life stage NOAEL = 17 mg/L ai
		b2. *Daphnia magna* reproduction NOAEC = 20 mg/L ai
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.,* algae)	a. Most sensitive aquatic plant EC_{50}	a. Green Algae 96-hr EC_{50} = 0.007 mg/L ai
Terrestrial Phase PCEs (Upland Habitat and Dispersal Habitat)		
Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance	a. Distribution of terrestrial monocot (1) seedling emergence and (2) vegetative vigor EC_{25} values	a1. Sorghum seedling emergence 21-d EC_{25} = 0.019 lbs ai/A a2. Wheat vegetative vigor 21-d EC_{25} = 0.02 lbs ai/A
	b. Distribution of terrestrial dicots (1) seedling emergence and (2) vegetative vigor EC_{25} values	b1. Tomato seedling emergence 21-d EC_{25} = 0.0063 lbs ai/A b2. Rape vegetative vigor 21-d EC_{25} = 0.011 lbs ai/A

[6] Physico-chemical water quality parameters such as salinity, pH, and hardness are not evaluated because these processes are not biologically mediated and, therefore, are not relevant to the endpoints included in this assessment.

Table 2.6. Summary of Assessment Endpoints and Measures of Ecological Effect for Primary Constituent Elements of Designated Critical Habitat		
Assessment Endpoint	**Measures of Ecological Effect[5] (Data Sources Reviewed)**	**Specific Selected Toxicity Value (basis)**
Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal	c. Most sensitive food source acute EC_{50} or LC_{50} and NOAEC values for terrestrial vertebrates (mammals) and invertebrates, birds, and freshwater fish.	C1a. rat LD_{50} = 530 mg ai/kg-bw (most sensitive terrestrial mammalian) c1b. rat 2-generation reproduction study NOAEC = 200 mg/kg-bw (most sensitive terrestrial mammalian) c2a. Honey bee acute contact LD50 > 100 µg ai/bee (most sensitive terrestrial invertebrate) c2b. No chronic NOAEC data for terrestrial invertebrates
Reduction and/or modification of food sources for terrestrial phase juveniles and adults		c3a. Mallard duck LD_{50} = 2258 mg ai/kg-bw (most sensitive bird test as surrogate for terrestrial-phase amphibian)
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.		C3b. Bobwhite quail reproduction NOAEC =100 ppm (most sensitive bird test as surrogate for terrestrial-phase amphibian)

2.9 Conceptual Model

2.9.1 Risk Hypotheses

Risk hypotheses are specific assumptions about potential adverse effects (i.e., changes in assessment endpoints) and may be based on theory and logic, empirical data, mathematical models, or probability models (U.S. EPA, 1998). For this assessment, the risk is stressor-linked, where the stressor is the release of hexazinone to the environment. The following risk hypotheses are presumed for this endangered species assessment:

- Labeled uses of hexazinone within the action area may directly affect the CRLF by causing mortality or by adversely affecting growth or fecundity;

- Labeled uses of hexazinone within the action area may indirectly affect the CRLF by reducing or changing the composition of food supply;

- Labeled uses of hexazinone within the action area may indirectly affect the CRLF and/or modify designated critical habitat by reducing or changing the composition of the aquatic plant community in the ponds and streams comprising the species' current range and designated critical habitat, thus affecting primary productivity and/or cover;

- Labeled uses of hexazinone within the action area may indirectly affect the CRLF and/or modify designated critical habitat by reducing or changing the composition of the terrestrial plant community (i.e., riparian habitat) required to maintain acceptable water quality and habitat in the ponds and streams comprising the species' current range and designated critical habitat;

- Labeled uses of hexazinone within the action area may modify the designated critical habitat of the CRLF by reducing or changing breeding and non-breeding aquatic habitat (via modification of water quality parameters, habitat morphology, and/or sedimentation);

- Labeled uses of hexazinone within the action area may modify the designated critical habitat of the CRLF by reducing the food supply required for normal growth and viability of juvenile and adult CRLFs;

- Labeled uses of hexazinone within the action area may modify the designated critical habitat of the CRLF by reducing or changing upland habitat within 200 ft of the edge of the riparian vegetation necessary for shelter, foraging, and predator avoidance.

- Labeled uses of hexazinone within the action area may modify the designated critical habitat of the CRLF by reducing or changing dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal.

- Labeled uses of hexazinone within the action area may modify the designated critical habitat of the CRLF by altering chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs.

2.9.2 Diagram

The conceptual model is a graphic representation of the structure of the risk assessment. It specifies the stressor hexazinone release mechanisms, biological receptor types, and effects endpoints of potential concern. The conceptual models for aquatic and terrestrial phases of the CRLF are shown in Figures 2.6 and 2.7, respectively, and the conceptual models for the aquatic and terrestrial PCE components of critical habitat are shown in Figures 2.8 and 2.9, respectively.

Exposure routes shown in dashed lines (long-range atmospheric transport) are not quantitatively considered because the contribution of those potential exposure routes to potential risks to the CRLF and modification to designated critical habitat is expected to be negligible.

<u>Exposure Pathways and Routes in Aquatic Phase Conceptual Model</u>

Eggs, larvae, tadpoles, juveniles, and adult frogs may potentially absorb hexazinone across their membranes or gills and integuments.

Aquatic animals that serve as prey to the juvenile and adult CRLF may be exposed via gills and their integument to hexazinone and its major degradates in surface water.

Aquatic vascular and non-vascular plants may sorb to their membranes or transfer across their membranes dissolved hexazinone and its major degradates in surface water.

Terrestrial plants in semi-aquatic areas (i.e. wetlands, riparian zones) may uptake dissolved hexazinone and its major degradates from soil pore water, surface water, or groundwater.

The exposure pathways discussed here in the aquatic phase conceptual model are modified to address attributes assessed in aquatic component of critical habitat and PCEs in conceptual model 2.8

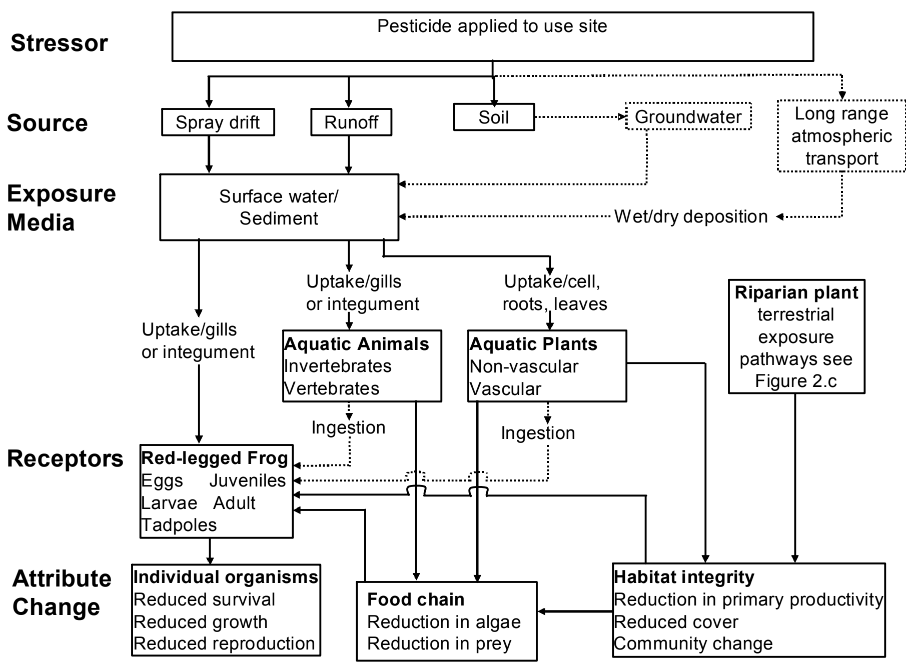


Figure 2.6 Conceptual Model for Pesticide Effects on Aquatic Phase of the Red-Legged Frog

<u>Exposure Pathways and Routes in Terrestrial Phase Conceptual Model</u>

Juvenile and adult frogs may experience dermal exposure to soil residues of hexazinone and its major degradates when seeking refuge in ground crevices from solar radiation or traversing across soils with hexazinone-related residues.

Juvenile and adult frogs may also incidentally ingest soil residues of hexazinone and its major degradates along with prey items.

CRLF prey items, small mammals (mice) and terrestrial insects, may uptake across their dermal/cuticle, residues of hexazinone and its major degradates.

The exposure pathways discussed here in the terrestrial phase conceptual model are modified to address attributes assessed in terrestrial component of critical habitat and PCEs in conceptual model 2.9.

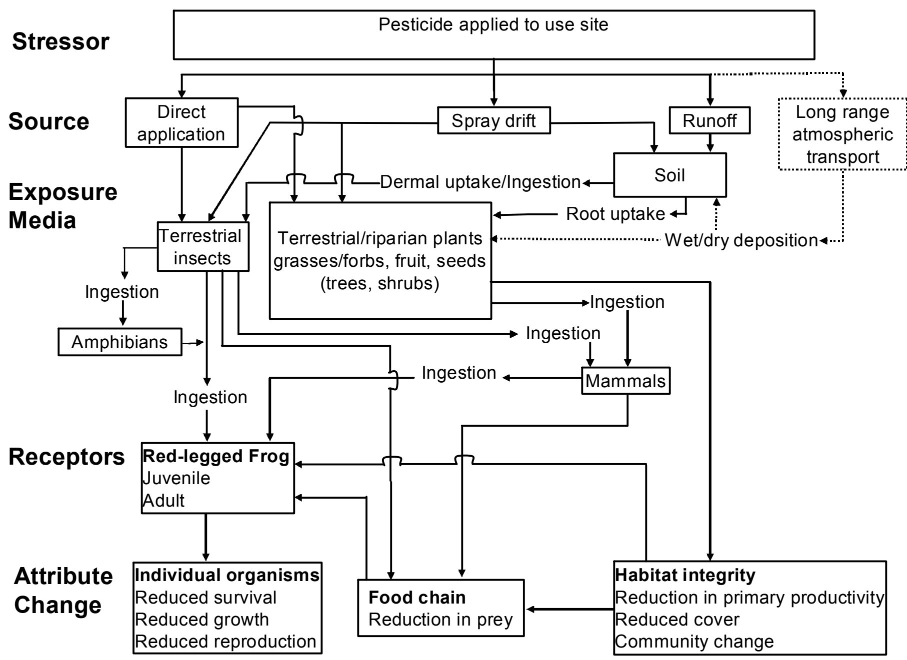


Figure 2.7 Conceptual Model for Pesticide Effects on Terrestrial Phase of Red-Legged Frog

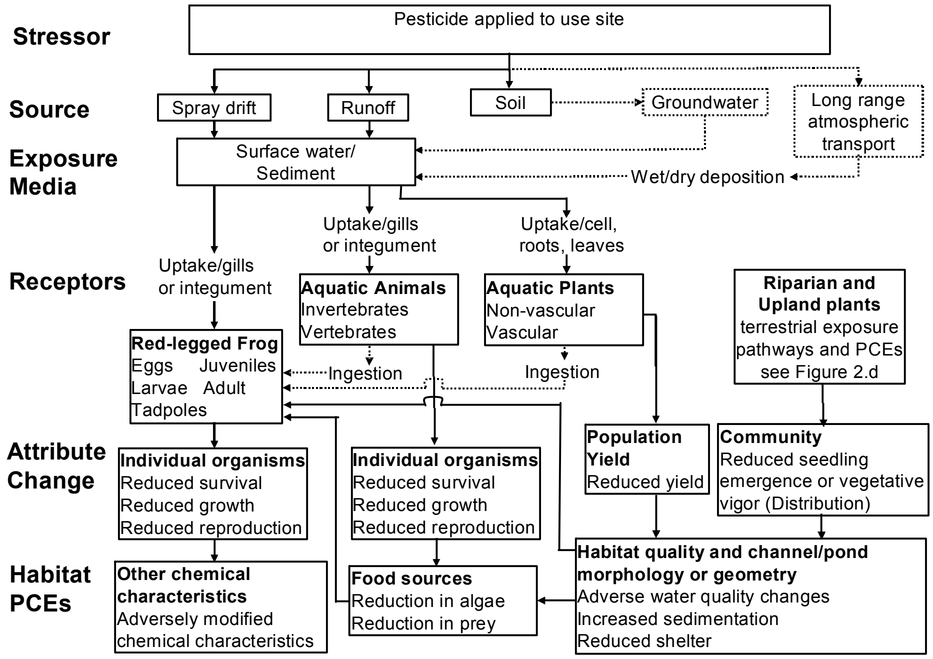


Figure 2.8 Conceptual Model for Pesticide Effects on Aquatic Components of Red-Legged Frog Critical Habitat

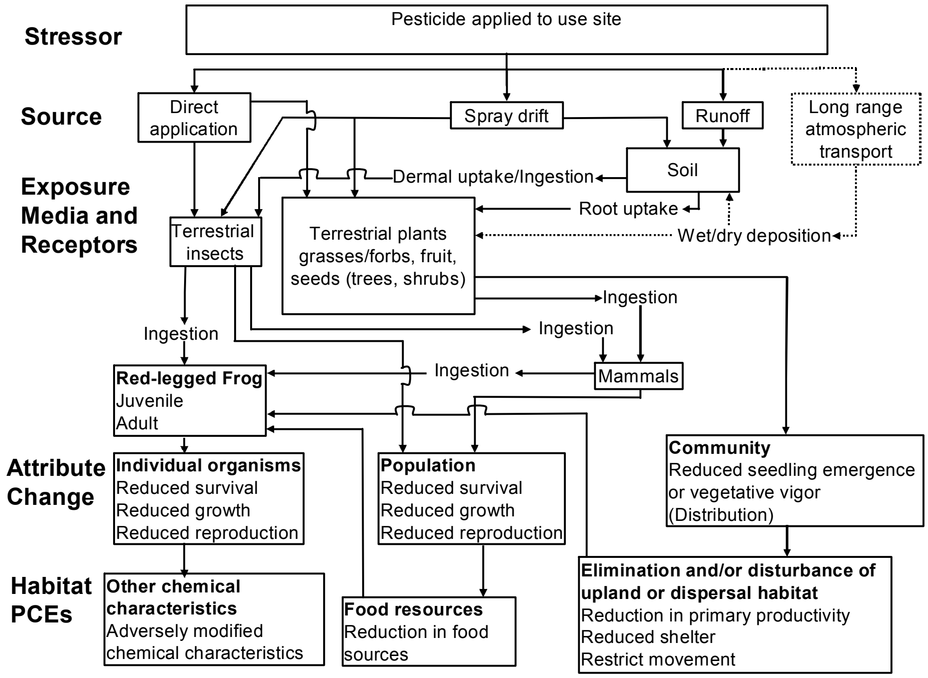


Figure 2.9 Conceptual Model for Pesticide Effects on Terrestrial Components of Red-Legged Frog Critical Habitat

2.10 Analysis Plan

The analysis plan is the final step in Problem Formulation. In order to address the risk hypothesis, the potential for adverse direct and indirect effects to the CRLF, its prey, and its habitat is estimated. The integration of exposure and ecological effects characterization determines the potential ecological risk from agricultural and non-agricultural uses of hexazinone, and the likelihood of direct and indirect effects to CRLF in aquatic and terrestrial habitats.

In the following sections, the use, environmental fate, and ecological effects of hexazinone are characterized and integrated to assess the risks. This is accomplished using a risk quotient (ratio of exposure concentration to effects concentration) approach. EECs are divided by acute and chronic toxicity values. The resulting RQs are then compared to the Agency's levels of concern (LOCs) (USEPA, 2004) (see Appendix B). Although risk is often defined as the likelihood and magnitude of adverse ecological effects, the risk quotient-based approach does not provide a quantitative estimate of likelihood and/or magnitude of an adverse effect. However, as outlined in the Overview Document (U.S. EPA, 2004), the likelihood of effects to individual organisms from particular uses of hexazinone is estimated using the probit dose-response slope and either the level of concern (discussed below) or actual calculated risk quotient value.

2.10.1 Measures of Exposure

The environmental fate properties of hexazinone along with available monitoring data indicate that runoff and spray drift are the principle potential transport mechanisms of hexazinone to the aquatic and terrestrial habitats of the CRLF. In this assessment, transport of hexazinone through runoff and spray drift is considered in deriving quantitative estimates of hexazinone exposure to CRLF, its prey and its habitats. Long-range transport of hexazinone is highly unlikely, given the chemical characteristics, formulation, and application methods. A vapor pressure of 2×10^{-7} mm Hg and Henry's Law Constant of 2×10^{-12} atm m^3/mole mean that hexazinone is not likely to volatilize and be transported offsite in the atmosphere. Some hexazinone residues may be transported as a component of field runoff, but dilution by receiving water bodies should limit the extent of long-range transport via surface water. Thus, the Agency believes that risk of long-range transport of hexazinone is minimal.

Measures of exposure are based on aquatic and terrestrial models that predict estimated environmental concentrations (EECs) of hexazinone using maximum labeled application rates and methods. The highest screening-level EEC based on granular use of hexazinone (non-crop/non-agricultural ROW at 12 lbs ai/A) or nongraular use (noncrop uses at 8 lb ai /A) was initially used to derive risk quotients. In cases where LOCs were not exceeded based on this use pattern, additional RQs were not derived because it was assumed that RQs for lower EECs would also not exceed LOCs. However, if LOCs were exceeded based on the highest EECs, use-specific RQs were also derived. The models used to predict aquatic EECs are the Pesticide Root Zone Model coupled with the Exposure Analysis Model System (PRZM/EXAMS). If there are any "May effect" determinations in the aquatic exposure assessment, an analysis of spray drift buffers needed to get below concentrations that exceed the endangered species level of concern will be conducted and a dilution model will be used and described in the risk characterization section of this risk assessment.

PRZM (v3.12beta, May 24, 2001) and EXAMS (v2.98.04, Aug. 18, 2002) are screening simulation models coupled with the input shell pe4v01.pl (Aug.8, 2003) to generate daily exposures and 1-in-10 year EECs of hexazinone that may occur in surface water bodies adjacent to application sites receiving hexazinone through runoff and spray drift. PRZM simulates pesticide application, movement and transformation on an agricultural field and the resultant pesticide loadings to a receiving water body via runoff, erosion and spray drift. EXAMS simulates the fate of the pesticide and resulting concentrations in the water body. The standard scenario used for ecological pesticide assessments assumes application to a 10-hectare agricultural field that drains into an adjacent 1-hectare water body that is 2 meters deep (20,000 m^3 volume) with no outlet. PRZM/EXAMS is used to estimate screening-level exposure of aquatic organisms to hexazinone. The measure of exposure for aquatic species is the 1-in-10 year return peak or rolling mean concentration. The 1-in-10 year peak is used for estimating acute exposures of direct effects to the CRLF, as well as indirect effects to the CRLF through effects to potential prey items, including: algae, aquatic invertebrates, fish, and frogs. The 1-in-10-year 60-day mean is used for assessing chronic exposure to the CRLF and fish and frogs serving as prey items.

The 1-in-10-year 21-day mean is used for assessing chronic exposure to aquatic invertebrates, which are also potential prey items.

Hexazinone residues are also estimated for waters downstream from the treated areas by assuming dilution with stream water (derived from land area) from unaffected sources, propagating downstream, until a point is reached beyond which there are no relevant LOC exceedences. Once the distribution of predicted stream water concentrations is obtained, it is further processed using a model that calculates expected dilution in the stream according to contributing land area. As the land area surrounding the field on which hexazinone is applied is enlarged, it encompasses a progressively greater drainage area; in effect, a progressively larger 'sub-watershed' is created, with a concomitant increase in dilution at the drainage point. This drainage point moves down-gradient along the stream channel as the sub-watershed is expanded. At a certain point the predicted stream concentrations will become sufficiently diluted; the region beyond this then falls outside the Action Area.

Terrestrial wildlife exposure estimates are typically calculated for birds and mammals, which are surrogates for terrestrial-phase amphibians and reptiles. These estimates focus on potential dietary exposures to the pesticide active ingredient and are estimated assuming the organisms are exposed to a singe pesticide residue on food items in a given exposure scenario. Exposure estimates for the terrestrial-phase CRLF and terrestrial invertebrates and mammals (serving as potential prey) assumed to be in the target area or in an area exposed to spray drift are derived using the T-REX model (version 1.3.1, 12/07/2006). This model incorporates the Kenega nomograph, as modified by Fletcher *et al.* (1994), which is based on a large set of actual field residue data. The upper limit values from the nomograph represented the 95th percentile of residue values from actual field measurements (Hoerger and Kenega, 1972). The Fletcher *et al.* (1994) modifications to the Kenega nomograph are based on measured field residues from 249 published research papers, including information on 118 species of plants, 121 pesticides, and 17 chemical classes. These modifications represent the 95th percentile of the expanded data set. For modeling purposes, direct exposures of the CRLF to hexazinone through contaminated food are estimated using the EECs for the small bird (20 g) which consumes small insects. Dietary-based and dose-based exposures of potential prey (small mammals) are assessed using the small mammal (15 g) which consumes short grass. The small bird (20g) consuming small insects and the small mammal (15g) consuming short grass are used because these categories represent the largest RQs of the size and dietary categories in T-REX that are appropriate surrogates for the CRLF and one of its prey items. Estimated exposures of terrestrial insects to hexazinone are bound by using the dietary based EECs for small insects and large insects. In addition, terrestrial exposures from granular applications (mg ai/square foot) for the CRLF are also estimated using T-REX (Appendix C).

If there are any "may effect" determinations in the terrestrial exposure assessment, terrestrial exposure and risk for the terrestrial-phase of the CRLF will be refined using T-HERPS (version 1.0, 2007), which is a modified version of T-REX (version 1.3.1*)* that allows for estimation of food intake for herptiles. Birds are typically used as surrogates for reptiles and terrestrial-phase amphibians. However, reptiles and terrestrial-phase amphibians (i.e., herptiles) tend to have much lower metabolic rates and lower caloric intake requirements than birds or mammals. As a consequence, birds are likely to consume more food than amphibians or reptiles on a daily dietary intake basis, assuming similar caloric content of the food item. T-REX (version 1.3.1.) has been altered to allow for an estimation of food intake for herptiles (T-HERPS) using the same basic procedure that T-REX uses to estimate avian food intake (see **Appendix D** for details).

EECs for terrestrial plants inhabiting dry and wetland areas are derived using TerrPlant (version 1.2.2, 12/26/2006)(Appendix E). This model uses estimates of pesticides in runoff and in spray drift to calculate EECs. EECs are based upon solubility, application rate and minimum incorporation depth.

Two spray drift models, AgDISP and AgDRIFT are used to assess exposures of terrestrial phase CRLF and its prey to hexazinone deposited on terrestrial habitats by spray drift. AgDISP (version 8.13; dated 12/14/2004) (Teske and Curbishley, 2003) is used to simulate aerial and ground applications using the Gaussian farfield extension

2.10.2 Measures of Effect

Data identified in Section 2.8 are used as measures of effect for direct and indirect effects to the CRLF. Data were obtained from registrant submitted studies or from literature studies identified by ECOTOX. The ECOTOXicology database (ECOTOX) was searched in order to provide more ecological effects data and in an attempt to bridge existing data gaps. ECOTOX is a source for locating single chemical toxicity data for aquatic life, terrestrial plants, and wildlife. ECOTOX was created and is maintained by the Agency's Office of Research and Development, and the National Health and Environmental Effects Research Laboratory's Mid-Continent Ecology Division.

As previously discussed in Section 2.8.1 and 2.8.2, assessment endpoints for the CRLF include direct toxic effects on survival, reproduction, and growth of the species itself, as well as indirect effects, such as reduction of the prey base and/or modification of its habitat. The assessment of risk for direct effects to the terrestrial-phase CRLF makes the assumption that toxicity of hexazinone to birds is similar to the terrestrial-phase CRLF. The same assumption is made for fish and aquatic-phase CRLF. Algae, aquatic invertebrates, fish, and amphibians represent potential prey of the CRLF in the aquatic habitat. Terrestrial invertebrates, small mammals, and terrestrial-phase amphibians represent potential prey of the CRLF in the terrestrial habitat. Aquatic, semi-aquatic, and terrestrial plants represent habitat of CRLF.

Acute (short-term) and chronic (long-term) toxicity information for hexazinone is characterized based on registrant-submitted studies and an updated review of the open literature. Based on an updated review of the open literature for October 2007, no endpoints for aquatic organisms were more sensitive than registrant submitted data. Although no open literature avian endpoints were more sensitive than registrant submitted data, there was an acute oral rat LD_{50} that was more sensitive than registrant submitted data. No endpoints for either aquatic or terrestrial plants were more sensitive than registrant submitted data. Therefore the more sensitive mammal acute oral LD_{50} = 530 mg/g (Kennedy 1984) was included and used in this assessment to modify the Action Area and evaluate direct and indirect effects to the CRLF and its critical habitat.

The acute measures of effect used for animals in this screening level assessment are the LD_{50}, LC_{50} and EC_{50}. LD stands for "Lethal Dose", and LD_{50} is the amount of a material, given all at once, that is estimated to cause the death of 50% of the test organisms. LC stands for "Lethal Concentration" and LC_{50} is the concentration of a chemical that is estimated to kill 50% of the test organisms. EC stands for "Effective Concentration" and the EC_{50} is the concentration of a chemical that is estimated to produce a specific effect in 50% of the test organisms. Endpoints for chronic measures of exposure for listed and non-listed animals are the NOAEL/NOAEC and NOAEC. NOAEL stands for "No Observed-Adverse-Effect-Level" and refers to the highest tested dose of a substance that has been reported to have no harmful (adverse) effects on test organisms. The NOAEC (*i.e.*, "No-Observed-Adverse-Effect-Concentration") is the highest test concentration at which none of the observed effects were statistically different from the control. The NOEC is the No-Observed-Effects-Concentration. For non-listed plants, only acute exposures are assessed (*i.e.*, EC_{25} for terrestrial plants and EC_{50} for aquatic plants).

2.10.3 Integration of Exposure and Effects

Risk characterization is the integration of exposure and ecological effects characterization to determine the potential ecological risk from agricultural and non-agricultural uses of hexazinone, and the likelihood of direct and indirect effects to CRLF in aquatic and terrestrial habitats. The exposure and toxicity effects data are integrated in order to evaluate the risks of adverse ecological effects on non-target species. For the assessment of hexazinone risks, the risk quotient (RQ) method is used to compare exposure and measured toxicity values. EECs are divided by acute and chronic toxicity values. The resulting RQs are then compared to the Agency's levels of concern (LOCs) (USEPA, 2004) (see Appendix B).

For this endangered species assessment, listed species LOCs are used for comparing RQ values for acute and chronic exposures of hexazinone directly to the CRLF. If estimated exposures directly to the CRLF of hexazinone resulting from a particular use are sufficient to exceed the listed species LOC, then the effects determination for that use is "may affect". When considering indirect effects to the CRLF due to effects to animal prey (aquatic and terrestrial invertebrates, fish, frogs, and mice), the listed species LOCs are also used. If estimated exposures to CRLF prey of hexazinone resulting from a particular use are sufficient to exceed the listed species LOC, then the effects determination for that use is a "may affect." If the acute RQ being considered also exceeds the non-listed species acute risk LOC, then the effects determination is a LAA. If the RQ is between the listed species LOC and the non-listed species LOC, then further lines of evidence (*i.e.* probability of individual effects, species sensitivity distributions) are considered in distinguishing between a determination of NLAA and a LAA. When considering indirect effects to the CRLF due to effects to algae as dietary items or plants as habitat, the non-listed species LOC for plants is used because the CRLF does not have an obligate relationship with any particular aquatic and/or terrestrial plant. If the RQ being considered for a particular use exceeds the non-listed species LOC for plants, the effects determination is LAA.

3 Exposure Assessment

Hexazinone products are formulated as granulars, pellets/tablets, emulsifiable concentrates, ready-to-use liquids, soluble concentrates/solids and a technical grade active ingredient. Products are applied using aerial or ground equipment or by hand, or using a hand-held, boom, knapsack or power sprayer. Use practice limitations prohibit application of hexazinone through any type of irrigation system. The majority of labels do not specify the allowed maximum application rate per year nor do they specify the maximum number of applications per year, therefore a conservative estimate will be used for this assessment.

Although the maximum number of applications from the label for pasture, alfalfa and Christmas tree uses is one, other uses do not specify a maximum rate or maximum number of applications per year. For this assessment, one application per year for those other uses is based on additional information from the label. Blueberry uses are modeled at a maximum of one application per year due to no application within 450 days of harvest. Pineapple is modeled as one application per year due to the growing season, which is nearly a year in length. Noncrop uses and all granular uses, non-crop/non-agricultural ROW, forest site preparation and rangeland, are modeled as one application per year due to the requirement for rain to activate hexazinone and because maximum effects are achieved after 12-24 months.

Risks from ground boom and aerial applications are considered in this assessment because they are expected to result in the highest off-target levels of hexazinone due to generally higher spray drift levels. Ground boom and aerial modes of application tend to use lower volumes of application applied in finer sprays than applications coincident with sprayers and spreaders and thus have a higher potential for off-target movement via spray drift.

3.1 Label Application Rates and Intervals

Nationally, hexazinone is an herbicide used to control a broad spectrum of weeds including undesirable woody plants in alfalfa, rangeland and pasture, woodland, pineapples, sugarcane and blueberries. It is also used on ornamental plants, forest trees and other non-crop areas. Hexazinone is registered for pre-emergent, post-emergence, layby, directed spray and basal soil applications. It is used as a non-selective herbicide in noncropland areas and as a selective herbicide in reforestation practices.

The purpose of the exposure assessment is to determine if the currently permitted label uses do not harm the CRLF, therefore conservative assumptions are developed for each use. Analysis of labeled use information is the critical first step in evaluating the action. The current label for hexazinone represents the FIFRA regulatory action; therefore, labeled use and application rates specified on the label form the basis of this assessment. The assessment of use information is critical to the development of the action area and selection of appropriate modeling scenarios and inputs.

A review of uses from the Label Use Information System (LUIS) produced by Office of Pesticide Programs (OPP) Biological and Economic Analysis Division (BEAD) revealed that many labels do not specify maximum number of applications per year/ crop cycle or maximum rates per year. This requires assumptions be made in the design for these conservative scenarios.

Although the maximum number of applications from the label for pasture, alfalfa and Christmas tree uses is one, other uses do not specify a maximum rate or maximum number of applications per year. For this assessment, one application per year for those other uses is based on additional information from the label: blueberry uses are modeled at a maximum of one application per year due to no application within 450 days of harvest; pineapple is modeled as one application per year due to the growing season, which is nearly a year in length; all noncrop and granular uses are modeled with one application per year. This assumption based on the availability for moisture to activate the pesticide and maximum effects not being achieved until 12-24 months after activation. Based on the assumption of a single application per year, the highest maximum rate identified for this assessment is identified for noncrop(12 lb/A).

Currently registered agricultural and non-agricultural uses of hexazinone within California are listed in Table 3.1 based on the conservative assumptions.

Table 3.1 Hexazinone Labeled Uses of and Application Information for the CRLF Risk Assessment.

Crop	Max Appl. Rate (lbs a.i./A)	Max # of Appl./Crop Cycle	Modeled Max # Appl	Seasonal Max Dose/Year (lbs a.i./A)
NON-CROP	12	NS	1	NS
NONAGRICULTURAL UNCULTIVATED AREAS/SOILS	8	NS	1	NS
AGRICULTURAL RIGHTS-OF-WAY/FENCEROWS/HEDGEROWS	8.	NS	1	NS
AGRICULTURAL UNCULTIVATED AREAS	8.	NS	1	NS
AIRPORTS/LANDING FIELDS	8.	NS	1	NS
SEWAGE DISPOSAL AREAS	8.	NS	1	NS
FOREST Site Preparation (g)	5	NS	1	NS
SUGARCANE	3.6	NS	1	NS
PINEAPPLE	3.6	NS	1	NS
CONIFER RELEASE	3	NS	1	NS
RANGELAND (g)	3	NS	1	NS
BLUEBERRY	3	NS	1	NS

CHRISTMAS TREE PLANTATIONS	2.	1	1	NS
ALFALFA	1.5	NS	1	1.5
PASTURES	1.1	1	1	NS
NS = Not stated on the label.				
1 Uses assessed based on memorandum from SRRD dated 07/31/2007				

3.2 Aquatic Exposure Assessment

3.2.1 Modeling Approach

Aquatic exposures are quantitatively estimated for all of assessed uses using scenarios that represent high exposure sites for hexazinone use. Each of these sites represents a 10 hectare field that drains into a 1-hectare pond that is 2 meters deep and has no outlet. Exposure estimates generated using the standard pond are intended to represent a wide variety of vulnerable water bodies that occur at the top of watersheds including prairie pot holes, playa lakes, wetlands, vernal pools, man-made and natural ponds, and intermittent and first-order streams. As a group, there are factors that make these water bodies more or less vulnerable than the standard surrogate pond. Static water bodies that have larger ratios of drainage area to water body volume would be expected to have higher peak EECs than the standard pond. These water bodies will be either shallower or have large drainage areas (or both). Shallow water bodies tend to have limited additional storage capacity, and thus, tend to overflow and carry pesticide in the discharge whereas the standard pond has no discharge. As watershed size increases beyond 10 hectares, at some point, it becomes unlikely that the entire watershed is planted to a single crop, which is all treated with the pesticide. Headwater streams can also have peak concentrations higher than the standard pond, but they tend to persist for only short periods of time and are then carried downstream.

Crop-specific management practices for all of the assessed uses of hexazinone were used for modeling, including application rates, number of applications per year, application intervals, buffer widths and resulting spray drift values modeled using standard EFED default values. These are 5% of the application rate for aerial application and 1% of the application rate for ground application. The date of first application was developed based on several sources of information including data provided by BEAD, a summary of individual applications from the CDPR PUR data, and Crop Profiles maintained by the USDA. More detail on the crop profiles and the previous assessments may be found at:

http://pestdata.ncsu.edu/cropprofiles/cropprofiles.cfm

The highest screening-level EEC (based on granular use of hexazinone on noncrop at 12 lbs ai/A or nongranular use for noncrop uses at 8 lb ai /A) was initially used to derive risk quotients. In cases where LOCs were not exceeded based on this use pattern, additional RQs were not derived because it was assumed that RQs for lower EECs would also not exceed LOCs. However, if LOCs were exceeded based on the highest EECs, use-specific RQs were also derived

3.2.2 Model Inputs

Table 3.2 summarizes the PRZM/EXAMS environmental fate data used for aquatic exposure Inputs for the hexazinone Endangered Species Assessment for the CRLF

Table 3.2 Hexazinone Model Inputs for PRZM/EXAMS for the CRLF Risk assessment

Parameter		Value	Comments	Source
Molecular Weight (grams/mole)		252.3		
Solubility (mg/L) Vapor Pressure (torr) Henry's Constant (atm m^3/mol) K_d (L/kg)		33,000 $2.0E^{-7}$ $2.0E^{-12}$ 0.45	10X reported value	product chemistry MRID41528101
K_{oc} (L/kg)		37.0		MRID41528101
Aerobic Soil Metabolism Half-life (days)		648 (216x3)	Based on 3X single aerobic soil metabolism linear first order half-life	MRID41807401 MRID42635001
Aerobic Aquatic Metabolism Half-life (days)		180 (60x3)	Based on 3X single aerobic aquatic metabolism linear first order half-life	MRID41811801
Anaerobic Aquatic Metabolism Half-life (days) Photodegradation in Water Half-life (days)		690 (230x3) Stable	Based on 3X single anaerobic aquatic metabolism linear first order half-life	MRID41807402 MRID41300801
Photodegradation in Soil Half-life (days)		Stable		MRID41300802
Hydrolysis Half-life (days)	pH 5	Stable		MRID41587301
	pH 7	Stable		MRID41587301
	pH 9	Stable		MRID41587301
Spray Drift Fraction		5% 1%	Aerial Ground	Default value

1 – Spray drift not included in final EEC due to edge-of-field estimation approach
2 – Inputs determined in accordance with EFED "*Guidance for Chemistry and Management Practice Input Parameters for Use in Modeling the Environmental Fate and Transport of Pesticides*" dated February 28, 2002

3.2.3 PRZM/EXAMS Aquatic Exposure Modeling Results

The aquatic EECs for the various scenarios and application practices are listed in Table 3.3. Peak EEC's for all uses permitted by the label range from 5.7 ug/l for blueberry to 156.6 ug/l for non-crop/non-agricultural ROW. Twenty-one and sixty-day EEC's range from 5.6 ug/l for blueberry to 153.7 ug/l for the noncropuse.

Table 3.3 Aquatic EECs in the Standard EXAMS Water Body (1 in 10 year EEC (ug/L))

Crop/Usage	Applic[a,b,c] Rate (#/ac)	Application Method[a]	Peak	21 day Average	60 day Average	Notes
NonAgRofWay/Fence	12	Ground Spray	156.6	153.7	148.8	
Noncrop: AgRofWay/Fence (Granular)	8.	Granule Applicator/ Broadcast	98.558	95.946	92.276	
Noncrop: AgUnCultivated (Granular)	8.	Aircraft/Granule Applicator/Soil Broadcast	84.49	83.338	81.291	
Forest Site preparation (Granular)	5.	Hand gun/ Soil Treatment	16.20	15.92	15.47	
Conifer Release	3	Ground Spray	52.139	51.144	49.744	
Rangeland (Granular)	3	Aircraft/Granule Applicator/Soil Broadcast	35.507	35.02	34.156	
BlueBerry	3	Sprayer/Spot Soil Treatment	5.6815	5.6033	5.4706	No Recorded Usage
Christmas Trees	2.	Air/Ground Spray	46.594	46.014	45.385	
Alfalfa	1.5	Aircraft/ Sprinkler Irrig/Chemigation/ Boomsprayer	24.788	24.437	23.583	
Pasture	1.1	Boom sprayer/ Ground sprayer/ Broadcast	10.629	10.48	10.2124	

[a] All crops/usages are one single application.
[b] All crops/usages are simulated at the highest rate allowed on any of the labels.
[c] Application rates for PRZM-EXAMS input are the labels multiplied by a conversion to kilograms/hectare (1.121 * lb/A)

3.2.4 Existing Monitoring Data

A critical step in the process of characterizing EECs is comparing the modeled estimates with available surface water monitoring data. Hexazinone has a limited set of monitoring data relevant to the CRLF assessment. There are only three measured Hexazinone concentrations above the limit of quantification in the California Department of Pesticide Regulation (CDPR) *data.* (http://www.cdpr.ca.gov/docs/sw/surfdata.htm) All three CDPR measured concentrations were recorded at the River Road sampling station on Orestimba Creek, tributary to the San Joaquin River in Stanislaus County. The concentrations of 0.06 µg/l, 0.5 µg/l and 0.154 µg/l were collected on January 31, 2001; February 14, 2001; and July 2, 2002, respectively. All three are non-targeted samples (i.e., study was not specifically designed to capture hexazinone concentrations in high use areas).

Hexazinone is not on the list of pesticides for which the NAQWA program samples in either of the two California sampling units. There is also no data for hexazinone or its degradates in the California Pesticide Air Monitoring Results: 1986-2000 (Kollman, 2002).

3.2.5 Spray Drift Buffer Analysis

When considering the terrestrial habitats of the CRLF, spray drift onto non-target areas from use sites could potentially result in exposures of the CRLF, its prey and its habitat to hexazinone. Therefore, it is necessary to estimate the distance from the application site where spray drift exposures do not result in LOC exceedences for organisms within the terrestrial habitat.

Since spray drift is the most likely means through which non-target terrestrial organisms will be potentially exposed to hexazinone, the AGDISP model (version 8.15) is used to estimate the terrestrial distance from the site of application to where RQs are predicted to fall below the endangered species LOC. The highest single maximum application rate allowed on the label was modeled to determine the maximum potential off-site estimated environmental concentrations (EECs) based on upper bound Kenaga values. The noncrop usage has the highest application rate (12 pounds per acre), but permits ground application only. The hexazinone usage with the highest aerial application rate is a conifer release forestry application (40 pounds per acre). These two applications produce the longest buffer widths. Typically aerial application is modeled for the assessment since aerial application produce much higher spray drift due the higher release heights**. Tables 3.4** and **3.5** have selected input parameters used in AGDISP modeling.

Table 3.4. AGDISP Input Parameters For Hexazinone Pineapple Application by Aerodyne Wasp Helicopter

Application. Method	Aerial (Aerodyne Wasp Helicopter)
Canopy Height (Forest)	30 ft
Release Height	15 ft
Swath Displacement	½ swath
Application Rate	4.0 pounds a.i. per acre
Spray Volume	5 gal·acre^{-1}
Non-volatile Fraction	0.3
Active Fraction	0.075
Specific Gravity (Carrier)	1.0
Specific Gravity (Hexazinone)	0.542
Fraction of Applied[1]	0.00087
Initial Average Deposition[2]	0.00348

[1] = LOC/RQ
[2] = (Fraction of applied) x (Application rate for forestry conifer release in lbs a.i/acre)

Table 3.5. AGDISP Input Parameters For Hexazinone Non-Crop Application by Ground Boom Sprayer	
Application. Method	**Aerial (Aerodyne Wasp Helicopter)**
Canopy Height (Forest)	none
Release Height	4 ft
Swath Displacement	½ swath
Application Rate	12.0 pounds a.i. per acre
Spray Volume	10 $gal \cdot acre^{-1}$
Non-volatile Fraction	0.15
Active Fraction	0.0375
Specific Gravity (Carrier)	1.0
Specific Gravity (Hexazinone)	0.542
Fraction of Applied[1]	0.00064
Initial Average Deposition[2]	0.0064
[1] = LOC/RQ [2] = (Fraction of applied) x (Application rate for non-agricultural rights-of-way in lbs a.i/acre)	

A single application was modeled for both applications because it is unlikely that the same terrestrial invertebrate would be exposed to the maximum amount of spray drift from multiple applications. There is also no data showing more than one application per year. For a terrestrial organism to receive the maximum concentration of hexazinone from multiple applications, it would require that each application is made under identical atmospheric conditions (*e.g.*, same wind speed and same wind direction) and the terrestrial organism being exposed is located in the same location (which receives the maximum amount of spray drift) after each application. Certain factors, including variations in topography, cover, and meteorological conditions over the transport distance are not accounted for by the AGDISP model (*i.e.*, it models spray drift from aerial and ground applications in a flat area with little to no ground cover and a steady, constant wind speed and direction). Therefore, in most cases, the drift estimates from AGDISP will overestimate exposure, especially as the distance increases from the site of application, since the model does not account for potential obstructions (*e.g.*, large hills, berms, buildings, trees, *etc.*).

Conservative assumptions are made regarding the droplet size distributions being modeled (‘ASAE Very Fine to Fine’ for rights-of way uses, the application method (*i.e.*, aerial), release heights and wind speeds. Alterations in any of these inputs would decrease the area of potential effect. As noted in Section 3.2.4, no hexazinone was detected in the air monitoring studies conducted in California. Therefore, it is unlikely that any terrestrial invertebrate outside the buffer from the site of hexazinone application would actually receive a level of exposure high enough to cause an adverse effect. The analysis of spray drift distances was completed using the Gaussian extension to AGDISP

For the terrestrial phase, an analysis was conducted using the most sensitive terrestrial endpoint, the terrestrial plant NOAEC of 0.00348 lbs ai/acre. This distance identifies those locations where terrestrial landscapes can be impacted by spray drift deposition alone (no runoff considered) at concentrations above the listed species LOC for terrestrial plants. The LOC was compared to the highest RQ for aerial applications to noncrop at 12.0 lbs ai/acre. In this analysis, the most sensitive endpoint was the NOAEC of 0.00348 lbs ai/A (0.00390 kg ai/hectare), which yielded a terrestrial spray drift distance of 4,501 feet. Each lower application rate yields a lower buffer distance. These distances represent the maximum extent where effects are possible using the most sensitive data and the endangered species LOC for plants (1.0).

In order to characterize the portion of the action area that is relevant to the CRLF and specific to the area where the effects determination (i.e. NLAA versus LAA) will be made, a similar analysis was conducted using the most sensitive non-endangered plant EC_{25} of 0.0064 lbs ai/acre. Typically the NOAEC is used when there is an obligate relationship between the species being assessed and endangered plants (or other taxa). However, there is no obligate relationship between the CRLF and any endangered plant; therefore the LAA/NLAA determination is based on the area defined by the non-listed species LOC (i.e., EEC/EC_{50}). Using the same approach described above, the maximum distance for the aerial use of hexazinone on noncrop at 12 lbs ai/acre is 3,3669 feet with reductions in distance for lower application rates. A summary of the modeled distances for these application rates is presented in Table 3.6.

The LOC was compared to the highest RQ for aerial applications to pineapple at 4.0 lbs ai/acre. In this analysis, the most sensitive endpoint was the NOAEC of 0.00348 lbs ai/A (0.00390 kg ai/hectare), which yielded a terrestrial spray drift distance of 4,200 feet. Each lower application rate yields a lower buffer distance. These distances represent the maximum extent where effects are possible using the most sensitive data and the endangered species LOC for plants (1.0).

In order to characterize the portion of the action area that is relevant to the CRLF and specific to the area where the effects determination (i.e. NLAA versus LAA) will be made, a similar analysis was conducted using the most sensitive non-endangered plant EC_{25} of 0.0064 lbs ai/acre. Using the same approach described above, the maximum distance for the aerial use of hexazinone on pineapple at 4 lbs ai/acre is 3,589 feet with reductions in distance for lower application rates. A summary of the modeled distances for these application rates is presented in Table 3.6.

Table 3.6 Summary of AgDISP Predicted Terrestrial Spray Drift Distances

Application Rate: lbs/ac (method)	Uses Represented	NOAEC Distance (ft)[1]	EC_{25} Distance (ft)[2]
12.0 (ground boom)	Non-crop	4501	3366
4.0 (helicopter)	Pineapple	4200	3589

[1] The NOAEC value is used to define the buffer associated with the full extent of the action area.
[2] The EC_{25} value is used to define the buffer associated with the relevant portion of the action area.

Given that the greatest buffer distance is 4501 feet for terrestrial plants, this value was used to define the action area (i.e., this buffer distance is added to the initial area of concern depicted in Figure 2.3. The action area (based on the buffer distance of 4501 feet) and the portion of the action area that is relevant to the CRLF (based on impacts to terrestrial plants at the non-listed LOC and a corresponding buffer distance of 3589 feet) is shown in Figure 2.4 and Figure 2.5.

3.2.6 Downstream Dilution Analysis

The final step in defining the action area is to determine the downstream extent of exposure in streams and rivers where the EEC could potentially be above levels that would exceed the most sensitive LOC. To complete this assessment, the greatest ratio of aquatic RQ to LOC was estimated. Using an assumption of uniform runoff across the landscape, it is assumed that streams flowing through treated areas (i.e. the initial area of concern) are represented by the modeled EECs; as those waters move downstream, it is assumed that the influx of non-impacted water will dilute the concentrations of hexazinone present.

The use of the "RQ to LOC ratio" provides information on the concentration that must be reached in downstream water to be below the LOC. Therefore, the analysis defines the point where the percentage of treated area with the watershed would yield sufficient non-impacted water to dilute the EECs to concentrations below the LOC. Further details on this approach are provided in Appendix B.

Using a NOAEC for non-vascular aquatic plants (the most sensitive species) of 7 ug/L and a maximum peak EEC for applications to non-crop (non-agricultural ROW) equal to 156.6 ug/L yields an RQ/LOC ratio of 39.15 (39.15).] Using the downstream dilution approach (described in more detail in **Appendix F**) yields a target percent crop area (PCA) of 4.2%. This value has been input into the downstream dilution approach for cultivated crop and pasture, forest and non-agricultural uses. Cultivated crop and pasture use added 4,853 kilometers, and resulted in a total of 68,257 kilometers of stream downstream from the initial area of concern. Forest use resulted in the largest addition, 22,416 kilometers, for a total of 160,771 kilometers of stream downstream from the initial area of concern. Non-agricultural uses resulted in the smallest addition, 1,868 kilometers, for a total of 105,929 kilometers of stream downstream from the initial area of concern. By way of comparison, there are 332,962 kilometers of streams within the initial area of concern all of which are assumed to be at the modeled EEC.

Similar to the spray drift buffer described above, the LAA/NLAA determination is based on the area defined by the point where concentrations exceed the non-vascular aquatic plant EC_{50} value, in this case 22.37 ug/L. Applying the same approach to downstream extent yields a RQ/LOC ratio of 22.37 (22.37) which equates to a downstream dilution factor of 27.8%. Cultivated crop and pasture use added 3,534 kilometers, and resulted in a total of 66,938 kilometers of stream downstream from the initial area of concern. Forest use resulted in the largest addition, 18,772 kilometers, for a total of 157,127 kilometers of stream downstream from the initial area of concern. Non-agricultural uses resulted in the smallest addition, 1,081 kilometers, for a total of 105,142 kilometers of stream downstream from the initial area of concern.

3.3 Terrestrial Exposure

3.3.1 Bird and Mammal Exposure (T-REX Model)

EFED estimates exposure of birds (Table 3.8) and mammals (Table 3.9) to pesticides using the Terrestrial Exposure Model (T-REX). T-REX uses the Kenaga nomagram, as modified by Fletcher *et al.* (1994) to determine pesticide residues on several categories of food items, and then calculates the potential dose an organism might receive from ingesting contaminated items using allometric equations. Dose estimates are based on an upper bound dose and the assumptions that the organism exclusively eats one type of food item and forages only in the treated and/or overspray areas. Hexazinone translocates in plant tissue and the residence time of the parent compound or any degradates is unknown. residues in the plant are not expected to exceed residues on the plant. Assessments for applications were completed for the non-crop/nonagricultural ROW granular uses (12 lb/A) noncrop uses (airports/sewage) (8 lb/A), forest site preparation (g) (5 lb/A), pineapple (3.6 lb/A), conifer release (3 lb/A), blueberry (3 lb/A), rangeland (g) (3 lb/A), Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) which comprise almost all recorded uses in California.

Table 3.7 Input Parameters for TREX and T-HERPS

Parameter	Value	Source
Percentage active ingredient for maximum rate	100%	Labels, application rate already adjusted
Number of applications	1	Labels
Application interval	None, single application	Labels
Dissipation half-life	35 days	Default

Table 3.8 TREX Avian Dose Estimates

Feeding Category	**Dietary-based EECs (mg/kg-food item)**	**Acute Dose-based EECs (mg/kg-bw)**	
		Small (20 g)	**Medium (100 g)**
Noncrop (airports)(8 lb/A)			
Short grass	1920.00	2186.69	1246.94
Tall grass	880.00	1002.23	571.52
Broadleaf plants/small insects	1080.00	1230.01	701.40
Fruits/pods/seeds/large insects	120.00	136.67	77.93
Pineapple (3.6 lb/A)			
Short grass	864.00	984.01	561.12
Tall grass	396.00	451.00	257.18
Broadleaf plants/small insects	486.00	553.51	315.63
Fruits/pods/seeds/large insects	54.00	61.50	35.07
Blueberry and conifer release (3 lb/A)			

Short grass	720.001	820.01	467.60
Tall grass	330.00	375.84	214.32
Broadleaf plants/small insects	405.00	461.25	263.03
Fruits/pods/seeds/large insects	45.00	51.25	29.23
Christmas Tree (2 lb/A)			
Short grass	480.00	546.67	311.74
Tall grass	220.00	250.56	142.88
Broadleaf plants/small insects	270.00	307.50	175.35
Fruits/pods/seeds/large insects	30.00	34.17	19.48
Alfalfa 1.5 lb ai/A			
Short grass	360.00	410.00	233.80
Tall grass	165.00	187.92	107.16
Broadleaf plants/small insects	202.50	230.63	131.51
Fruits/pods/seeds/large insects	22.50	25.63	14.61
Pasture (1.1 lb/A)			
Short grass	264.00	300.67	171.45
Tall grass	121.00	137.81	78.58
Broadleaf plants/small insects	148.50	169.13	96.44
Fruits/pods/seeds/large insects	16.50	18.79	10.72

Table 3.9 TREX Mammal Dose Estimates

Feeding Category	**Chronic Dietary-based EECs (mg/kg-food item)**	**Dose-based EECs (mg/kg-bw)**	
		Small (15 g)	**Medium (35 g)**
Noncrop (airports)(8 lb/A)			
Short grass	1920.00	1830.57	1265.17
Tall grass	880.00	839.01	579.87
Broadleaf plants/small insects	1080.00	1029.70	711.66
Fruits/pods/seeds/large insects	120.00	114.41	79.07
Granivore		25.42	17.57
Pineapple (3.6 lb/A)			
Short grass	864.00	823.76	569.33
Tall grass	396.00	377.56	260.94
Broadleaf plants/small insects	486.00	463.36	320.25
Fruits/pods/seeds/large insects	54.00	51.48	35.58
Granivore		11.44	7.91
Blueberry and conifer release (3 lb/A)			
Short grass	720.00	686.46	474.44
Tall grass	330.00	314.63	217.45
Broadleaf plants/small insects	405.00	386.14	266.87
Fruits/pods/seeds/large insects	45.00	42.90	29.65
Granivore		9.53	6.59
Christmas Tree (2 lb/A)			
Short grass	480.00	457.64	316.29
Tall grass	220.00	209.75	144.97
Broadleaf plants/small insects	270.00	257.42	177.91
Fruits/pods/seeds/large insects	30.00	28.60	19.77
Granivore		6.36	*4.39*
Alfalfa 1.5 lb ai/A Short grass	360.00	343.23	237.22
Tall grass	165.00	157.31	108.73
Broadleaf plants/small insects	202.50	193.07	133.44

Fruits/pods/seeds/large insects	22.50	21.45	14.83
Granivore		4.77	3.29
Pasture (1.1 lb/A)			
Short grass	264.00	251.70	173.96
Tall grass	121.00	115.36	79.73
Broadleaf plants/small insects	148.50	141.58	97.85
Fruits/pods/seeds/large insects	16.50	15.73	10.87
Granivore		3.50	2.42

Granular Applications

Terrestrial exposures from granular applications (mg ai/square foot) for the CRLF are also estimated using the T-REX Version 1.3.1. Broadcast treatment of hexazinone-treated granules assumes that 100% of the granules are unincorporated on the ground. Risk to terrestrial animals from ingesting granules is based on LD_{50}/ft^2 values. Although the habitat of the CRLF and its prey items are not limited to a square foot, there is presumably a direct correlation between the concentration of a pesticide in the environment (mg/ft^2) and the chance that an animal will be exposed to a concentration that could adversely affect its survival. Further description of the mg/ft^2 index is provided in U.S. EPA (1992 and 2004).

In order to derive an estimate of the granular exposure per square foot, the granular application rates for hexazinone were converted from lb ai/A to mg/ft^2 in Table 3.10 using the following equation: mg/ft^2 EEC = (application rate in lb ai/A x 453,590 mg/lb) / 4,560 ft^2/A). The LD_{50}/ft^2 values are calculated using the avian toxicity value (adjusted LD_{50} of the assessed animal and its weight classes) as a surrogate for the terrestrial-phase CRLF and the EEC (mg ai/ft^2).

Table 3.10 Terrestrial EECs for Granular Uses of Hexazione

Use	Application Rate (lb ai/A)	Number of Applications	EEC (mg/ft^2)
Non-crop (Nonagricultural ROW)	12	1	124.96
Forest Site preparation	5	1	52.06
Rangeland	3	1	31.24

Uncertainties associated with use of the T-REX model to estimate risk to the terrestrial-phase of the CRLF based on ingestion of hexazinone granules are discussed in Section 6.2.4.

EFED uses the T-Herps model to estimate the direct terrestrial effects to CRLF from ingestion of chemical residues with food/prey items. Because the dose experienced by a CRLF varies with its size, dose is estimated for a range of CRLF sizes: small (1.4 g), medium (37 g), and large (238 g) CRLF (Table 3.11). Because small frogs are modeled to have a higher metabolic rate than large CRLF, smaller frogs are estimated to receive a higher dose than larger frogs when ingesting the same type of food item. However, it is also assumed that small frogs are incapable of ingesting small herbivore mammals, small insectivore mammals, and small terrestrial phase amphibians (note the "N.A." at the small frog location within the last three columns of Table 3.11).

Because it is the small herbivore mammal food item that is estimated to have the highest concentration and only the medium and large CRLF ingest this food item, it is the medium CRLF (with their higher metabolic rate than the large CRLF) that receive the highest doses. Dose based estimates for individual exposure scenarios vary from 0.64 to 41.96 mg/kg-bw for small frogs, 0.20to 1196.78 mg/kg-bw for medium frogs, and 0.130 to 186.05 mg/kg-bw for large frogs.

Table 3.11. Assessment of Direct Effects on California Red-legged Frog (CRLF) Using Dose-Based Estimated Environmental Concentrations (EECs) of Hexazinone Based on the T-Herps Model for Small (1.4 g), Medium (37 g), and Large (238 g) CRLF.

Use	Maximum Application Rates[1] (lb a.i./A)	Dose-based EECs (mg/kg-bw) for Small, Medium, and Large CRLF (Small / Medium / Large)				
		Broadleaf Plants/ Small Insects	Fruit/ Pods/ Seeds/ Large Insects	Small Herbivore Mammals	Small Insectivore Mammals	Small Terrestrial Phase Amphibian
Agricultural Uses						
Noncrop: airport, Ag ROW	8	41.96 / 41.24 /27.03	4.66 / 4.58 / 3.00	N.A./ 1196.78/186.05	N.A. / 74.80 / 11.63	N.A. / 1.43 / 0.94
Pineapple	3.6	18.88 / 18.56 / 12.16	2.10 / 2.06 / 1.35	N.A. /538.55 / 83.72	N.A. / 33.66 / 5.23	N.A. /0.64 / 0.42
Blueberry and conifer release	3	15.73 / 15.46/ 10.14	1.75 /1.72 / 1.13	N.A. / 448.79 / 69.77	N.A. / 28.05 / 4.36	N.A. / 0.54 / 0.35
Christmas Tree	2	10.49 / 10.31 / 6.76	1.17 / 1.15 / 0.75	N.A. / 299.20 / 46.51	N.A. / 18.70 / 2.91	N.A. / 0.36 /0.23
Alfalfa	1.5	7.87 / 7.73 / 5.07	0.87 / 0.86 / 0.56	N.A. / 224.40 / 34.89	N.A. / 14.02 / 2.18	N.A. / 0.27 / 0.18
Pasture	1.1	5.77 / 5.67 / 3.72	0.64 /0.63 /0.41	N.A. / 164.56 / 25.58	N.A. / 10.28 / 1.60	N.A. /0.20 /0.13

Table 3.12 Assessment of Direct Effects on California Red-legged Frog (CRLF) Using Chronic Dose-Based Estimated Environmental Concentrations (EECs) of Hexazinone Based on the T-Herps Model						
Use	Maximum Application Rates[1] (lb a.i./A)	Chronic Dietary-based EECs (mg/kg)				
		Broadleaf Plants/ Small Insects	Fruit/ Pods/ Seeds/ Large Insects	Small Herbivore Mammals	Small Insectivore Mammals	Small Terrestrial Phase Amphibian
Agricultural Uses						
Noncrop: airport, Ag ROW	8	1080.00	120.00	1265.17	79.07	37.49
Pineapple	3.6	486.00	54.00	569.33	35.58	16.87
Blueberry and conifer release	3	405.00	45.00	474.44	29.65	14.06
Christmas Tree	2	270.00	30.00	316.29	19.77	9.37
Alfalfa	1.5	202.50	22.50	237.22	14.83	7.03
Pasture	1.1	148.50	16.50	173.96	10.87	5.15

Chronic dietary EECS are estimated using the T-Herps model to assess direct effects for the CRLF. Table 3.12 depicts the highest EECs for small herbivore mammals ranging from 173.96 to 1265.17 mg/kg. EECs for small insects ranged from 148.50 to 1080.0 mg/kg with EECs for large insect ranging from 16.50 to 120.00 mg/kg and EECs for small insectivore mammals ranging from 10.87 to 79.07. The lowest exposure for small amphibian resulted in EECs ranging from 5.15 to 37.49 mg/kg.

3.3.2 Terrestrial Invertebrate Exposure

Exposure of terrestrial invertebrates was estimated using the dietary-based EECs produced by T-REX for the two insect categories, small and large (Table 3.13). The value produced by T-REX, mg a.i./kg insect, is equivalent to µg a.i./g insect. The hexazinone residue for a bee (µg a.i./bee) using an adult honey bee weight of 0.128 g and multiplying it by the assumed weight of a honey bee (0.128 g) to establish a dose per bee. This method assumes that contact is the relevant route of exposure, rather than ingestion. This method of estimation is believed to be adequate for hexazinone.

Table 3.13 Terrestrial Invertebrate Estimated Dietary-Based Environmental Concentrations (EECs)		
Application Rate (lb ai/A)	**Insect Size Category**	**EECs (mg ai/kg insect)**
Noncrop 8 lb/A	Small insects	1080.00
	Large insects	120.00
	Large insects	
Conifer Release 3 lb/A	Small insects	405.00
	Large insects	45.00
Blueberry 3 lb/A	Small insects	405.00

	Large insects	45.00
Christmas Tree 2 lb/A	Small insects	270.00
	Large insects	30.00
Alfalfa 1.5 lb ai/A	Small insects	202.50
	Large insects	22.50
Pasture 1.1 lb/A	Small insects	148.50
	Large insects	16.50

3.4 Terrestrial Plant Exposure Assessment

Currently, EFED uses the TerrPlant Model (Version 1.2.2) to evaluate exposure of terrestrial plants to pesticides applied on agricultural fields. TerrPlant estimates a runoff component based on application rate and solubility of the compound, and a spray drift component based on application method. Because non-target plants are of concern for herbicide uses, EFED also used two spray drift models, AgDrift and AgDisp, to more fully evaluate spray drift effects. Screening level estimates from TerrPlant are presented here in the exposure section and in the risk estimation section. AgDrift is used in the risk characterization section to more fully evaluate potential off-site effects. AgDisp has an additional module which mathematically estimates drift beyond the range of AgDrift, which is based on empirical data, and has only been parameterized to approximately 950 ft from the application site. In general, spray drift is more dependent on the atmospheric physics of droplet transport than on the physico-chemical properties of the pesticide and carrier liquid.

3.4.1 TerrPlant

TerrPlant has two basic exposure scenarios. The first is an adjacent upland area, which is exposed to the pesticide via drift and dissolved concentrations in sheet runoff. The second is an adjacent semi-aquatic (wetland) area, which is exposed to the pesticide via drift and to dissolved concentrations in channelized runoff. Drift is calculated as a percentage of the application rate (1% for ground, and 5% for aerial, airblast, or spray chemigation) and is not adjusted for distance from the application site. The amount of dissolved pesticide in the runoff component is estimated based on solubility of the active ingredient. TerrPlant estimates are shown in Table 3.14. Total loading in upland areas (runoff plus drift) ranged from 0.007 lb ai/A (pasture, ground boom) to 0.84 lb ai/A (noncrop, aerial). Total loading in wetland areas (runoff plus drift) ranged from 0.5 lb ai/A (pasture, ground boom) to 6 lb ai/A (non-crop/non-agricultural ROW, granular). Pesticide loading to the different areas is affected by application rate and depth of incorporation. Concentrations of hexazinone in the runoff are more important in the wetland than for the upland. Thus, the specific crops used for the bounding estimates may not be the same. Based on the TerrPlant model, spray drift to either a wetland or an upland area ranged from 0 lb ai/A (non-crop/nonagricultural ROW, site preparartion and rangeland, granular) to 0.4 lb/A (noncrop, aerial). In this model, spray drift is strictly a function of application rate and method (ground vs. aerial). Loading estimates are presented in Table 3.14.

Table 3.14 Terrestrial Plant Exposure (TerrPlant)			
Crop and Application Rate (lb ai/A)	Total Loading (Runoff +Drift) EEC (lb ai/A)		Drift EEC (lb ai/A)
	Upland areas	Wetland areas	All areas
Noncrop: Non-agricultural rights-of-way, ground-boom (12.0 lb ai/A)	0.6	6	0
Noncrop (8 lb/A)	0.8	4.4	0.4
Forest site Preparation (5.0 lb/A)	0.25	2.5	0
Rangeland (g) (3 lb/A)	0.15	1.5	0
Conifer Release (3 lb/A)	0.24	2.04	0.04
Pineapple (3.6 lb/A)	0.36	1.98	0.18
Blueberry (3 lb/A)	0.3	1.65	0.15
Christmas trees (2 lb/A)	0.12	1.02	0.02
Alfalfa (1.5 lb/A)	0.15	0.82	0.07
Pasture (1.125 lb/A)	0.07	0.56	0.01
[a] Total loading to adjacent areas is a function of both runoff and spray drift, which are influenced by application rate, and depth of incorporation for soil incorporated compounds. For hexazinone, the highest and lowest EECs for the various receiving compartments are not always the same crop and/or application method.			

4. **Effects Assessment**

This assessment evaluates the potential for hexazinone to directly or indirectly affect the CRLF or modify its designated critical habitat. As previously discussed in Section 2.7, assessment endpoints for the CRLF include direct toxic effects on the survival, reproduction, and growth of CRLF, as well as indirect effects, such as reduction of the prey base or modification of its habitat. In addition, potential modification of critical habitat is assessed by evaluating effects to the PCEs, which are components of the critical habitat areas that provide essential life cycle needs of the CRLF. Direct effects to the aquatic-phase of the CRLF are based on toxicity information for freshwater fish, while terrestrial-phase effects are based on avian toxicity data. Because the frog's prey items and habitat requirements are dependent on the availability of freshwater fish and invertebrates, small mammals, terrestrial invertebrates, and aquatic and terrestrial plants, toxicity information for these taxa are also discussed. Acute (short-term) and chronic (long-term) toxicity information is characterized based on registrant-submitted studies and a comprehensive review of the open literature on hexazinone.

As described in the Agency's Overview Document (U.S. EPA, 2004), the most sensitive endpoint for each taxon is used for risk estimation. For this assessment, evaluated taxa include aquatic-phase amphibians, freshwater fish, freshwater invertebrates, aquatic plants, birds (surrogate for terrestrial-phase amphibians), mammals, terrestrial invertebrates, and terrestrial plants.

Toxicity endpoints are established based on data generated from guideline studies submitted by the registrant, and from open literature studies that meet the criteria for inclusion into the ECOTOX database maintained by EPA/Office of Research and Development (ORD) (U.S. EPA, 2004). Open literature data presented in this assessment were obtained from (October 2007) as well as ECOTOX information obtained on October 2007. In order to be included in the ECOTOX database, papers must meet the following minimum criteria:

(1) the toxic effects are related to single chemical exposure;
(2) the toxic effects are on an aquatic or terrestrial plant or animal species;
(3) there is a biological effect on live, whole organisms;
(4) a concurrent environmental chemical concentration/dose or application rate is reported; and
(5) there is an explicit duration of exposure.

Data that pass the ECOTOX screen are evaluated along with the registrant-submitted data, and may be incorporated qualitatively or quantitatively into this endangered species assessment. In general, effects data in the open literature that are more conservative than the registrant-submitted data are considered. The degree to which open literature data are quantitatively or qualitatively characterized is dependent on whether the information is relevant to the assessment endpoints (*i.e.*, maintenance of CRLF survival, reproduction, and growth) identified in Section 2.8. For example, endpoints such as behavior modifications are likely to be qualitatively evaluated, because quantitative relationships between modifications and reduction in species survival, reproduction, and/or growth are not generally available.

Citations of all open literature not considered as part of this assessment because they were either rejected by the ECOTOX screen or accepted by ECOTOX but not used (e.g., the endpoint is less sensitive and/or not appropriate for use in this assessment) are included in Appendix**G. Appendix G** also includes a rationale for rejection of those studies that did not pass the ECOTOX screen and those that were not evaluated as part of this endangered species risk assessment.

In addition to registrant-submitted and open literature toxicity information, other sources of information, including use of the acute probit dose response relationship to establish the probability of an individual effect and reviews of the Ecological Incident Information System (EIIS), are conducted to further refine the characterization of potential ecological effects associated with exposure to hexazinone. A summary of the available aquatic and terrestrial ecotoxicity information, and the incident information for hexazinone are provided in Sections 4.1 through 4.4, respectively.

4.1 Toxicity of Hexazinone to Aquatic Organisms

Toxicity to fish and aquatic invertebrates is categorized using the system shown in Table 4.1 (U.S.EPA, 2004). Toxicity categories for plants have not been defined.

Table 4.1 Categories of Acute Toxicity for Aquatic Organisms	
LC_{50} (ppm)	**Toxicity Category**
<0.1	Very highly toxic
>0.1-1	Highly toxic
>1-10	Moderately toxic
>10-100	Slightly toxic
>100	Practically nontoxic

Table 4.2 summarizes the most sensitive aquatic toxicity endpoints for the CRLF, based on an evaluation of both the submitted studies and the open literature, as previously discussed. A brief summary of submitted data considered relevant to this ecological risk assessment for the CRLF is presented below.

Table 4.2 Freshwater Aquatic Toxicity Profile for Hexazinone				
Assessment Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	MRID	**Comment**
Acute Direct Toxicity to Aquatic-Phase CRLF	**Fathead Minnow** Rainbow Trout Bluegill sunfish Bluegill sunfish	**96-hr LC_{50} = 274 mg/L** 96-hr LC_{50} >320 mg/L 96-hr LC_{50} >370 mg/L 96-hr LC_{50} = 505 mg/L	00104980 00104980 00104980 00076959	TGAI is Practically nontoxic
	Rainbow Trout Bluegill Sunfish	96-hr LC_{50} >1000 mg/L 96-hr LC_{50} > 585.6 mg/L	41235002 41235001	25% TEP is Practically nontoxic
Chronic Direct Toxicity to Aquatic-Phase CRLF	**Fathead minnow**	**Estimated chronic NOAEL = 17 ppm**	41406001	Endpoints were juvenile survival (day 39), length and weight.
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Freshwater Invertebrates (*i.e.* prey items)	***Daphnia magna***	**48-hr EC_{50} = 151.6 mg/L**	00116269	TGAI is practically nontoxic to freshwater invertebrates.
	Daphnia magna	48-hr EC_{50} = 339.9 mg/L	41235003	25% TEP is practically nontoxic to freshwater invertebrates.

Table 4.2 Freshwater Aquatic Toxicity Profile for Hexazinone				
Assessment Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	MRID	**Comment**
Indirect Toxicity to Aquatic-Phase CRLF via Chronic Toxicity to Freshwater Invertebrates (*i.e.* prey items)	***Daphnia magna***	**NOAEC = 20 mg/L**	00078041	Reproduction is the endpoint. Classified as supplemental due to dilution water and dilution preparation not reported.
	Daphnia magna	NOAEC = 29 mg/L LOAEC = 81 mg/L	41406002	Classified supplemental due to no information on dry weight of first generation reported. Endpoint was daphnid survival.
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Vascular Aquatic Plants	***Lemna gibba***	**EC_{50} = 0.0374 mg/L ai**	43225101	Endpoint is frond count. Study is classified as acceptable.
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Non-vascular Aquatic Plants	***Selenastrum capricornutum***	**120-hr EC_{50} =7 µg/L** **NOAEC = 4.0 µg/L**	41287001	Study is classified as acceptable. Results based on nominal concentrations.
	Anabaena flos-aquae	EC_{50}=210 µg/L	43302701	Study is classified as acceptable.
	Navicula pelliculosa	EC_{50}=12 µg/L	43302701	Study is classified as acceptable for a freshwater diatom.

Table 4.2 Freshwater Aquatic Toxicity Profile for Hexazinone				
Assessment Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	MRID	**Comment**
Values in bold were used in the assessment.				

4.1.1 Toxicity to Freshwater Fish

A comprehensive search of the open literature provided no toxicity information on lethal or sub lethal effects of hexazinone to amphibians.Given that no hexazinone toxicity data are available for aquatic-phase amphibians; freshwater fish data were used as a surrogate to estimate direct acute and chronic risks to the CRLF. Freshwater fish toxicity data were also used to assess potential indirect effects of hexazinone to the CRLF. Direct effects to freshwater fish resulting from exposure to hexazinone couldindirectly affect the CRLF via reduction in available food. As discussed in Section 2.5.3, over 50% of the prey mass of the CRLF may consist of vertebrates such as mice, frogs and fish (Hayes and Tennant, 1985).

A summary of acute and chronic freshwater fish data is provided below in Sections 4.1.1.1 through 4.1.1.2.

4.1.1.1 Freshwater Fish: Acute Exposure (Mortality) Studies

With respect to technical grade hexazinone, the reported acute 96-hour LC_{50} values were >320 mg/l for rainbow trout (MRID 00104980), and 274 mg/l for Fathead minnow (MRID 00104980). Two studies were available for bluegill sunfish, 96-hour LC_{50} >370 mg/L and 96-hour LC_{50} = 505 mg/L (MRID 00104980 and 00076959). The results of these tests indicate that hexazinone is practically nontoxic to fish. The fathead minnow LC_{50} = 274 mg/l will be used to assess direct and indirect (dietary) effects for the CRLF.

4.1.1.2 Freshwater Fish: Chronic Exposure (Growth/Reproduction) Studies

Chronic freshwater fish toxicity studies were used to assess potential direct effects via growth and reproduction to the aquatic-phase of the CRLF. A fish life cycle test with fathead minnow was submitted for hexazinone (MRID 41406001). A chronic NOAEC=17 mg/L was reported for juvenile survival (day 39), length and weight. This value will be used in this assessment.

4.1.2 Toxicity to Freshwater Invertebrates

Freshwater aquatic invertebrate toxicity data were used to assess potential indirect effects of hexazinone to the CRLF. Direct effects to freshwater invertebrates resulting from exposure to hexazinone could indirectly affect the CRLF via reduction in available food items. As discussed in Section 2.5.3, the main food source for juvenile aquatic- and terrestrial-phase CRLFs is thought to be aquatic invertebrates found along the shoreline and on the water surface, including aquatic sow bugs, larval alderflies and water striders.

A summary of acute and chronic freshwater invertebrate data is provided below in Section 4.1.2.1 through 4.1.2.2.

4.1.2.1 Freshwater Invertebrates: Acute Exposure Studies

Acute toxicity data for hexazinone are available for the preferred test species, *Daphnia magna*. A 48-hour EC_{50}=151.6 mg/l (MRID 00116269) indicates that hexazinone is practically nontoxic to aquatic invertebrates. This value will be used in this assessment.

4.1.2.2 Freshwater Invertebrates: Chronic Exposure Studies

Chronic toxicity data are available for daphnia magna. Two supplemental studies for daphnia resulted in NOEACs of 20 mg/L for reproduction (MRID 00078041) and 29 mg a.i./L for daphnid survival (MRID 41406002). The reproductive NOEAC will be used in this assessment as the most sensitive endpoint.

4.1.3 Toxicity to Aquatic Plants

Aquatic plant toxicity studies were used as one of the measures of effect to evaluate whether hexazinone may affect primary production and the availability of aquatic plants as food for CRLF tadpoles. Primary productivity is essential for indirectly supporting the growth and abundance of the CRLF.

An acceptable study is available for vascular aquatic plants using *Lemna gibba* with frond count as an endpoint. The EC_{50}=0.0374 mg/L (MRID 43225101) was used in this assessment.

Three studies for nonvascular aquatic plants were available. The acceptable nonvascular aquatic plant study with *Anabaena flos-aquae* resulted in an EC_{50}=210 µg/L (MRID 43302701). The acceptable study with the freshwater diatom *Navicula pelliculosa* resulted in an EC_{50}=12 µg/L (MRID 43302701). An acceptable study is available for nonvascular aquatic plants using *Selenastrum capricornutum.* The EC_{50}=7.0 µg/L with a NOAEC=4.0 µg/L (MRID 41287001) were based on nominal concentrations. The most sensitive value for *Selenastrum capricornutum* will be used to determine an LAA/NLAA effect, while the NOAEC will be used in the action area map.

4.2 Toxicity of Hexazinone to Terrestrial Organisms

Acute toxicity to terrestrial animals is categorized using the classification system shown in Table 4.3 (U.S. EPA, 2004). Toxicity categories for terrestrial plants have not been defined.

Table 4.3 Categories of Acute Toxicity for Avian and Mammalian Studies

Toxicity Category	Oral LD_{50}	Dietary LC_{50}
Very highly toxic	< 10 mg/kg	< 50 ppm
Highly toxic	10 - 50 mg/kg	50 - 500 ppm

Moderately toxic	51 - 500 mg/kg	501 - 1000 ppm
Slightly toxic	501 - 2000 mg/kg	1001 - 5000 ppm
Practically non-toxic	> 2000 mg/kg	> 5000 ppm

Table 4.4 summarizes the most sensitive terrestrial toxicity endpoints for the CRLF, based on evaluation of both submitted studies and the open literature. A brief summary of submitted and open literature data considered relevant to this ecological risk assessment for the CRLF is presented below.

Table 4.4 Terrestrial Toxicity Profile for Hexazinone

Assessment Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	MRID, Author and Date	**Comment**
Acute Direct Toxicity to Terrestrial-Phase CRLF (LD_{50})	Mallard duck acute oral	**LD_{50} = 2,258 mg ai/kg-bw**	00073988	Hexazinone is practically nontoxic to birds.
Acute Direct Toxicity to Terrestrial-Phase CRLF (LC_{50})	Mallard duck Bobwhite quail Bobwhite quail	LC_{50} >5,000 mg/kg	00104981 00072663 00107878	All studies were classified as acceptable. Hexazinone is practically nontoxic to birds
Chronic Direct Toxicity to Terrestrial-Phase CRLF	Bobwhite quail	NOAEC = 300 mg/kg	41764901	NOAEC was based on effects to the 14 day survivors.
	Mallard Duck	NOAEC > 1000 mg/kg	41764902	No observable treatment effects.
Indirect Toxicity to Terrestrial-Phase CRLF (via acute toxicity to mammalian prey items)	Rat acute oral/ rat	LD_{50}=530mg/Kg bw LD_{50} = 1200 mg ai/kg bw	(Kennedy 1984) 41235004	
Indirect Toxicity to Terrestrial-Phase CRLF (via chronic toxicity to mammalian prey items)	Rat reproduction	Chronic NOAEL = 200 mg/kg bw (2-generation reproduction study) LOAEC=2000 mg/kg	42066501	98% a.i in the diet. Endpoints are systemic and reproductive toxicity.
Indirect Toxicity to Terrestrial-Phase CRLF (via acute toxicity to terrestrial invertebrate prey items)	Honey bee	Acute contact LD_{50} > 100 ug/bee	41216502	
Indirect Toxicity to Terrestrial- and Aquatic-Phase CRLF (via toxicity to terrestrial plants)	Tomato *(dicot)*	Seedling emergence 21-d EC25 = 0.0064 lbs ai/A. NOAEC = 0.00348 lb/A	43162501	Based on weight.
	Sorghum *(monocot)*	Seedling emergence 21-d EC25 = 0.019 lbs ai/A. NOAEC = 0.0139	43162501	Based on weight.

Table 4.4 Terrestrial Toxicity Profile for Hexazinone				
Assessment Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	MRID, Author and Date	**Comment**
	Rape *(dicot)*	Vegetative Vigor 21-d EC25 = 0.011 lbs ai/A. NOAEC = 0.0071 lb/A	43162501	Based on weight.
	Wheat *(monocot)*	Vegetative Vigor 21-d EC25 = 0.020 lbs ai/A. NOAEC= 0.010 lb/A	43162501	Based on total weight

4.2.1 Toxicity to Birds

As specified in the Overview Document, the Agency uses birds as a surrogate for terrestrial-phase amphibians when amphibian toxicity is not available (U.S.EPA, 2004). No terrestrial-phase amphibian data are available for hexazinone, therefore acute and chronic avian toxicity data are used to assess the potential direct effects of hexazinone to terrestrial-phase CRLFs.

4.2.1.1 Birds: Acute Exposure (Mortality) Studies

Acute oral toxicity data are available for quail (LD_{50} = 2,258 mg/kg MRID 00073988) using 98% a.i. The results indicate that hexazinone is practically nontoxic to birds. This result will be used in this assessment.

Dietary acute toxicity studies resulted in an LC50 > 5,000 mg/kg for mallard duck (MRID 0010498), and bobwhite quail (MRID 00107878 and MRID 00072663) indicating that hexazinone is practically nontoxic to birds. These studies are not used quantitatively in this assessment because no mortality was observed at the highest test concentration.

4.2.1.2 Birds: Chronic Exposure (Growth, Reproduction) Studies

Chronic reproductive toxicity data is available for the bobwhite quail (NOAEC = 300 mg/kg MRID 417649-01). The endpoint was a statistically significant increase in food consumption for survivors from day 14 through test termination. This endpoint will be used in this assessment.

Chronic reproductive toxicity data is available for the mallard duck (NOAEC >1,000 mg/kg, MRID 41764902). However, this is not the most sensitive endpoint reported, therefore this value will not be used in the assessment.

4.2.2 Toxicity to Mammals

4.2.2.1 Mammals: Acute Exposure (Mortality) Studies

A registrant submitted rat acute oral (LD_{50}=1,200 mg/kg, MRID 41235004) was reported. The results indicate that hexazinone is practically nontoxic to mammals.

There is an open literature endpoint that is more sensitive than the registrant submitted data. An acute oral toxicity study is available for rat LD_{50} = 530 mg/kg (Kennedy 1984). The results indicate that hexazinone is practically nontoxic to mammals. This result will be used in this assessment.

4.2.2.2 Mammals: Chronic (Growth, Reproduction) Studies

An acceptable two-generation rat study (MRID 42066501) resulted in a systemic and reproductive NOAC=200 mg/kg. This value will be used in this assessment to calculate RQs for dietary effects for the CRLF.

4.2.3 Toxicity to Terrestrial Invertebrates

Terrestrial invertebrate toxicity data are used to assess potential indirect effects of hexazinone to the terrestrial-phase CRLF. Effects to terrestrial invertebrates resulting from exposure to hexazinone could also indirectly affect the CRLF via reduction in available food.

An acceptable honey bee study (LD50 > 100 μg/bee, MRID 41216502) indicates that hexazinone is practically nontoxic to terrestrial invertebrates. This value will be used qualitatively in the risk description.

4.2.4 Toxicity to Terrestrial Plants

Terrestrial plant toxicity data are used to evaluate the potential for hexazinone to affect riparian zone and upland vegetation within the action area for the CRLF. Impacts to riparian and upland (i.e., grassland, woodland) vegetation could result in indirect effects to both aquatic- and terrestrial-phase CRLFs, as well as modification to designated critical habitat PCEs via increased sedimentation, alteration in water quality, and reduction in of upland and riparian habitat that provides shelter, foraging, predator avoidance and dispersal for juvenile and adult CRLFs.

Plant toxicity data from both registrant-submitted studies and studies in the scientific literature were reviewed for this assessment. Registrant-submitted studies are conducted under conditions and with species defined in EPA toxicity test guidelines. Sub-lethal endpoints such as plant growth, dry weight, and biomass are evaluated for both monocots and dicots, and effects are evaluated at both seedling emergence and vegetative life stages. Guideline studies generally evaluate toxicity to ten crop species. A drawback to these tests is that they are conducted on herbaceous crop species only, and extrapolation of effects to other species, such as the woody shrubs and trees and wild herbaceous species, contributes uncertainty to risk conclusions.

Commercial crop species have been selectively bred, and may be more or less resistant to particular stressors than wild herbs and forbs. The direction of this uncertainty for specific plants and stressors, including hexazinone, is largely unknown. Homogenous test plant seed lots also lack the genetic variation that occurs in natural populations, so the range of effects seen from tests is likely to be smaller than would be expected from wild populations.

The results of the Tier II seedling emergence and vegetative vigor toxicity tests on non-target plants are summarized in **Table 4.4.**

Toxicity data are available for seedling emergence and vegetative vigor for terrestrial plant exposure to hexazinone. The results from the seedling emergence test study resulted in an EC_{25} of 0.0064 lbs/A for tomato. The monocot EC_{25} result for sorghum was 0.019, lbs/A.

The vegetative vigor EC25 for dicots was 0.011 for rape. The EC_{25} for emergence for monocots was 0.020 for wheat. These values will be used in this assessment.

4.3 Incident Database Review

A review of the EIIS database for ecological incidents involving hexazinone was completed on November 8, 2007. The results of this review for terrestrial, plant, and aquatic incidents are discussed below in Sections 4.5.1 through 4.5.3, respectively.

4.3.1 Terrestrial Animal Incidents

No terrestrial animal incidents were reported for hexazinone.

4.3.2 Plant Incidents

Nine hexazinone incidents have been reported for terrestrial plants. One incident, I000548-001 reporting non-target plant exposure, was classified as highly probable. Velpar ULW, a product with hexazinone, was applied using an aerial application method on a forest site in Florida. No application rate or total magnitude was reported.

Five reports were classified as possible.

I016312-001: Westar and Velpar, products containing hexazinone, were applied on a forest site in Oregon (2005). Aerial and ground methods were used to apply 2 lbs/A with a total magnitude of 300 acres. The caller reported the appearance of reddening on Ponderosa Pine needle growth. Damage could be due to the application of multiple products.

I016680-001: Velpar, a product containing hexazinone, was applied on a right-of-way site in Oregon (2005). Thirteen acres of vineyards were damaged through spray in a ground application. No application rate was reported. Damage could be due to the application of multiple products.

I000611-001: Velpar L, a product containing hexazinone, was applied in Georgia (1993). No report of use site, total magnitude, or application rate was provided. The report cited foliar damage (mottling and yellow-spotting) to the trees. Grasses were not affected.

I014407-036: A product containing hexazinone was applied on an alfalfa site in Washington (1994). No report of total magnitude, or application rate was provided. The damage to yield was to be assessed after the crop was harvested.

I015265-001: Outstar, a product containing hexazinone, was applied at a forest product site in Texas (2004) The loss of sixty-five percent of 129 ares of loblolly pine seedlings was reported, but. no application rate or method was provided. Damage could be due to the application of multiple products.

Two incidents reported were classified as misuse, I005822-001 reporting damage to flora and I007984-008 reporting damage to a tree. A report of trees dying, I0012233-001, was classified as unlikely. None of these incidents were used in this risk assessment.

4.3.3 Aquatic Animal Incidents

No aquatic animal incidents were reported for hexazinone**.**

5. Risk Characterization

Risk characterization is the integration of the exposure and effects characterizations. Risk characterization is used to determine the potential for direct and/or indirect effects to the CRLF or for modification to its designated critical habitat from the use of hexazinone in CA. The risk characterization provides an estimation (Section 5.1) and a description (Section 5.2) of the likelihood of adverse effects; articulates risk assessment assumptions, limitations, and uncertainties; and synthesizes an overall conclusion regarding the likelihood of adverse effects to the CRLF or its designated critical habitat (i.e., "no effect," "likely to adversely affect," or "may affect, but not likely to adversely affect").

5.1 Risk Estimation

Risk is estimated by calculating the ratio of exposure to toxicity. This ratio is the risk quotient (RQ), which is then compared to pre-established acute and chronic levels of concern (LOCs) for each category evaluated (**Appendix B**). For acute exposures to the CRLF and its animal prey in aquatic habitats, as well as terrestrial invertebrates, the LOC is 0.05. For acute exposures to the CRLF and mammals, the LOC is 0.1. The LOC for chronic exposures to CRLF and its prey, as well as acute exposures to plants is 1.0.

Risk to the aquatic-phase CRLF is estimated by calculating the ratio of exposure to toxicity using 1-in-10 year EECs based on the label-recommended hexazinone usage scenarios summarized in **Table 3.3** and the appropriate aquatic toxicity endpoint from **Table 4.1**. Risks to the terrestrial-phase CRLF and its prey (*e.g*. terrestrial insects, small mammals and terrestrial-phase frogs) are estimated based on exposures resulting from applications of hexazinone (**Tables 3.5 through 3.6**) and the appropriate toxicity endpoint from **Table 4.3**. Exposures are also derived for terrestrial plants, as discussed in Section 3.3 and summarized in **Table 3.7**, based on the highest application rates of hexazinone use within the action area.

5.1.1 5.1.1 Use of Probit Slope Response Relationship to Provide Information on the Endangered Species Levels of Concern

The Agency uses the probit dose response relationship as a tool for providing additional information on the potential for acute direct effects to individual listed species and indirect effects to aquatic or terrestrial animals that may affect the listed species of concern (U.S. EPA, 2004). As part of the risk characterization, an interpretation of acute RQ for listed species is discussed. This interpretation is presented in terms of the chance of an individual event (i.e., mortality or immobilization) should exposure at the EEC actually occur for a species with sensitivity to hexazinone on par with the acute toxicity endpoint selected for RQ calculation. To accomplish this interpretation, the Agency uses the slope of the dose response relationship available from the toxicity study used to establish the acute toxicity measures of effect for each taxonomic group that is relevant to this assessment. The individual effects probability associated with the acute RQ is based on the mean estimate of the slope and an assumption of a probit dose response relationship. In addition to a single effects probability estimate based on the mean, upper and lower estimates of the effects probability are also provided to account for variance in the slope, if available.

Individual effect probabilities are calculated based on an Excel spreadsheet tool IECV1.1 (Individual Effect Chance Model Version 1.1) developed by the U.S. EPA, OPP, Environmental Fate and Effects Division (June 22, 2004). The model allows for such calculations by entering the mean slope estimate (and the 95% confidence bounds of that estimate) as the slope parameter for the spreadsheet. In addition, the acute RQ is entered as the desired threshold. Probit analyses will be provided for direct and indirect effects (section 5.1). Calculations from probit analyses are reviewed under direct and indirect effects sections.

5.1.2 Exposures in the Aquatic Habitat

The highest screening-level aquatic EEC (based on non-granular use of hexazinone on non-agricultural Right-Of-Way at 12 lbs ai/A was initially used to derive risk quotients. In cases where LOCs were not exceeded based on this use pattern, additional RQs were not derived because it was assumed that RQs for lower EECs would also not exceed LOCs. However, if LOCs were exceeded based on the highest EECs, use-specific RQs were also derived.

5.1.2.1 Direct Effects to Aquatic-Phase CRLF

Direct effects to the aquatic-phase CRLF are based on peak EECs in the standard pond and the lowest acute toxicity value for freshwater fish. In order to assess direct chronic risks to the CRLF, 60-day EECs and the lowest chronic toxicity value for freshwater fish are used. As shown in Table 5.1, acute and chronic RQs for the highest EEC for applications of hexazinone are well below their respective LOCs; therefore, direct effects associated with acute and chronic exposure to hexazinone are not expected to occur for the aquatic-phase CRLF. These RQs are further characterized in Section 5.2.1.1. The highest screening-level EEC (non-crop/non-agricultural ROW at 12 lbs ai/A) was initially used to derive risk quotients. LOCs were not exceeded based on this use pattern, therefore additional RQs were not derived because it was assumed that RQs for lower EECs would also not exceed LOCs. A preliminary no effect determination for acute and chronic exposure for the aquatic-phase CRLF is based no LOC exceedence for any scenario.

Table 5.1 Summary of Direct Effect RQs for the Aquatic-phase CRLF

Direct Effects to CRLF[a]	Surrogate Species	Toxicity Value (µg/L)	EEC (µg/L)	RQ	LOC Exceedence and Risk Interpretation
Acute Direct Toxicity	Fathead minnow	LC_{50} = 274,000	Peak: 156.6	0.0006	No[b]
Chronic Direct Toxicity		NOAEC = 17,000	60 day 136.6	0.009	No[c]

[a] RQs associated with acute and chronic direct toxicity to the CRLF are also used to assess potential indirect effects to the CRLF based on a reduction in freshwater fish and frogs as food items.
[b] RQ < acute endangered species LOC of 0.05.
[c] RQ < chronic LOC of 1.0.

5.1.2.2 Indirect Effects to Aquatic-Phase CRLF via Reduction in Prey (non-vascular aquatic plants, aquatic invertebrates, fish, and frogs)

Non-vascular Aquatic Plants

Indirect effects of hexazinone to the aquatic-phase CRLF (tadpoles) via reduction in non-vascular aquatic plants in its diet are based on peak EECs from the standard pond and the lowest acute toxicity value for aquatic non-vascular plants. As shown in Table 5.2, RQs exceed the acute risk LOC (RQ ≥1.0) for aquatic plants for applications of hexazinone to non-crop (12 lb ai/A), noncrop (agricultural ROW, and forest site preparation (5.0 lbs/A), conifer release (3 lb ai/A), Christmas trees (2 lbs/A), alfalfa (1.5 lbs/A) and pasture (1.1 lbs/A) with RQ values ranging from 1.52 to 22.37. There is no exceedence for blueberries (3 lbs/A). The preliminary effects determination is "may affect" based on indirect effects to aquatic-phase CRLFs due to a reduction in non-vascular aquatic plants as food items.

Table 5.2 Summary of Acute RQs Used to Estimate Indirect Effects to the CRLF via Effects to Non-Vascular Aquatic Plants (diet of CRLF in tadpole life stage and habitat of aquatic-phase CRLF)			
Uses	**Application rate (lb ai/A) and type**	**Peak EEC (µg/L)**	**Indirect effects RQ* (food and habitat)**
Non-crop (Non-agricultural ROW)	12 spray	156.6	**22.37**
Agricultural uncultivated	8 spray	84.49	**12.07**
Agricultural ROW	8 spray	98.56	**14.08**
Airports, Sewage	8 spray	70.85	**10.12**
Forest Site Preparation	5	16.20	**2.31**
Blueberry	3 (liquid)	5.68	0.81
Christmas Trees	2 (liquid)	46.59	**6.66**
Alfalfa	1.5 (liquid)	24.79	**3.54**
Pasture	1.125 (liquid)	10.63	**1.52**
* = LOC exceedences (RQ ≥ 1) are bolded and shaded. RQ = use-specific peak EEC / [lemna EC50=37.4 µg/L].			

Aquatic Invertebrates

Indirect acute effects to the aquatic-phase CRLF via effects to prey (invertebrates) in aquatic habitats are based on peak EECs in the standard pond and the lowest acute toxicity value for freshwater invertebrates. For chronic risks, 21-day EECs and the lowest chronic toxicity value for invertebrates are used to derive RQs. As shown in Table 5.3 all acute and chronic RQs are well below their respective LOCs; therefore, indirect effects associated with acute and chronic exposure to hexazinone are not expected to occur for the aquatic-phase CRLF. RQs were calculated only for the use that resulted in the highest EEC (foliar use on non-crop /non-agricultural ROW at 12 lb ai/A) because none of the acute or chronic LOCs were exceeded. A "no effect" determination for acute and chronic aquatic invertebrates is based on the highest EECs application rates.

Table 5.3 Summary of Acute and Chronic RQs Used to Estimate Indirect Effects to the CRLF via Direct Effects on Aquatic Invertebrates as Dietary Food Items (prey of CRLF juveniles and adults in aquatic habitats)					
Uses	**Application rate (lb ai/A) and type**	**Peak EEC (µg/L)**	**21-day EEC (µg/L)**	**Indirect Effects Acute RQ***	**Indirect Effects Chronic RQ***
Non-crop (Non-agriculture ROW)	12 (granular)	156.6	153.7	0.001	0.005

* = LOC exceedences (acute RQ $\geq$ 0.05; chronic RQ $\geq$ 1.0) are bolded and shaded. Acute RQ = use-specific peak EEC / [daphnia 1.52 ppm]. Chronic RQ = use-specific 21-day EEC / [chronic daphnia 20 ppm].

Fish and Frogs

Fish and frogs also represent potential prey items of adult aquatic-phase CRLFs. RQs associated with acute and chronic direct toxicity to the CRLF (**Table 5.1**) are used to assess potential indirect effects to the CRLF based on a reduction in freshwater fish and frogs as food items. Given that acute and chronic RQs for direct toxicity to the CRLF are less than LOCs, indirect effects based on a reduction of fish and frogs as prey items are not expected. Therefore, a preliminary "no effect" determination is based on no RQ exceedence for any LOCs.

5.1.1.3 Indirect Effects to CRLF via Reduction in Habitat and/or Primary Productivity (Freshwater Aquatic Plants)

Indirect effects to the CRLF via direct toxicity to aquatic plants are estimated using the most sensitive non-vascular and vascular plant toxicity endpoints. Because there are no obligate relationships between the CRLF and any aquatic plant species, the most sensitive EC_{50} values, rather than NOAEC values, were used to derive RQs. Table 5.4 includes RQs for vascular plants (RQs for non-vascular plants are presented in Section 5.1.2.1 and Tables 5.2). RQs for Christmas tree, conifer release, airports, agricultural ROW, agricultural uncultivated, and non-crop/non-agricultural ROW exceed the LOC for indirect effects to the CRLF via effects to vascular plants. RQs range from 1.25-4.19 for effects to vascular plants for granular and liquid applications. RQs for forest site preparation, blueberry, rangeland, alfalfa and pasture do not exceed the LOC (RQs 0.15-0.95). In addition to the LOC exceedence for vascular plants LOCs are exceeded for non-vascular aquatic plants for all applications of hexazinone as previously discussed in Section 5.1.2.2 and summarized in Table 5.2. A "may affect" determination for indirect effects to CRLF via reduction in habitat and/or primary production is based on the LOC exceedence for both non-vascular and vascular aquatic plants.

Table 5.4 Summary of Acute RQs Used to Estimate Indirect Effects to the CRLF via Effects to Vascular Aquatic Plants (diet of CRLF in tadpole life stage and habitat of aquatic-phase CRLF)

Uses	Application rate (lb ai/A) and type	Peak EEC (µg/L)	Indirect effects RQ* (food and habitat)
Non-crop (NonAgricultural ROW)	12	156.6	**4.19**
Agricultural uncultivated	8	84.49	**2.26**
Agricultural ROW	8	98.56	**2.63**
Airports, Sewage	8	70.85	**1.89**
Forest Site Preparation (5 lb/A)	5	16.20	0.43
Conifer Release	3	52.14	**1.39**

Blueberry	3	5.68	0.15
Rangeland	3	35.51	0.95
Christmas Trees	2	46.59	**1.25**
Alfalfa	1.5	24.79	0.66
Pasture	1.1	10.63	0.28
[a] RQs used to estimate indirect effects to the CRLF via toxicity to non-vascular aquatic plants are summarized in Table 5.2. * = LOC exceedences (RQ $\geq$ 1) are bolded and shaded. RQ = use-specific peak EEC / [lemna EC50=37.4 µg/L].			

5.1.3 Exposures in the Terrestrial Habitat

5.1.3.1 Direct Effects to Terrestrial-phase CRLF

As previously discussed in Section 3.3, potential direct effects to terrestrial-phase CRLFs are based on granular and liquid applications of hexazinone.

Acute Effects

Potential direct acute effects to the terrestrial-phase CRLF are derived by considering dose- and dietary-based EECs modeled in T-REX for a small bird (20 g) consuming small invertebrates (Table 5.5) and acute oral and sub acute dietary toxicity endpoints for avian species. Dietary RQs could not be estimated because no mortality was observed at the highest tested level of hexazinone (LC_{50} >5,000 mg/kg-diet, MRIDs 00107878, 001040498 and 00072663).

Potential direct acute effects of non-granular hexazinone applications to terrestrial-phase CRLF are evaluated by calculating RQs from the most sensitive avian acute oral LD_{50}=2,258 mg/kg. All non-granular uses exceed endangered species LOCs, with RQs ranging from alfalfa (0.14 to) 0.76 (Table 5.5) except pasture (0.10). The preliminary effects determination for direct acute effects to the terrestrial-phase CRLF is “may effect”.

Table 5.5 Terrestrial Direct Effects to the CRLF from Hexazinone Exposure

Surrogate Species	Toxicity Value (mg/kg)	Uses	EEC (ppm)	RQ
Bobwhite Quail	LD_{50} = 2258	Noncrop (8 lb/A)	1537.51	**0.76**
		Pineapple (3.6 lb/A)	553.51	**034**
		Blueberry/Conifer release (3 lb/A)	461.25	**0.28**
		Christmas Trees	307.50	**0.19**

		(2.0 lb/A)		
		Alfalfa (1.5 lb/A)	230.63	**0.14**
		Pasture (1.1 lb/A)	169.13	0.10

Chronic Non-granular liquid spray applications

As shown in Table 5.6, chronic RQs, which range from 1.35 to 3.6, exceed LOCs for all non-granular uses of hexazinone. No exceedences resulted for pasture, alfalfa, or Christmas trees uses, with RQs ranging from 0.50 to 0.90. Therefore, the preliminary effects determination is "may effect" for direct chronic effects to the terrestrial-phase CRLF.

Table 5.6 Summary of Chronic RQs* Used to Estimate Direct Effects to the Terrestrial-phase CRLF (non-granular application)

Uses	Application rate (lb ai/A) and type	Dietary-based Chronic RQ[1]
Noncrop: Ag ROW, airport	8	**3.6**
Pineapple	3.6	**1.62**
Blueberry/conifer release (3 lbs/A)	3	**1.35**
Christmas Tree	2	0.90
Alfalfa	1.5	0.68
Pasture	1.1	0.50

* = LOC exceedences (chronic RQ $\geq$ 1) are bolded and shaded.
[1] Based on bobwhite quail chronic reproduction NOAEC = **3**00 mg/kg-diet (MRID 417649-01).

Granular applications

The RQ for direct effects of granular applications of hexazinone was calculated using the bird as a surrogate for the terrestrial-phase CRLF. The EEC for the granular application of 12 lb/A is shown in Table 5.7. Using the avian LD_{50} (2258 mg/kg) and the EEC (93.72) from the LDft2 model, the RQ is 0.04, which does not exceed the endangered species LOC.

EEC/toxicity value = RQ

93.72/2258 = 0.04

The highest screening-level EEC (non-crop/non-agricultural ROW at 12 lbs ai/A) was initially used to derive risk quotients. LOCs were not exceeded based on this use pattern, therefore additional RQs were not derived because it was assumed that RQs for lower EECs would also not exceed LOCs.

Table 5.7 Comparison of Granular EECs to Adjusted LD_{50} Value Used to Estimate Direct Effects to the Terrestrial-phase CRLF (granular application)

Use	Application Rate (lb ai/A)	EEC (mg/ft^2)	Adjusted LD_{50} Value (mg/kg-bw)[1]	
			20 g (juvenile)	100g (adult)
Non-crop/NonAg ROW	12	93.72	1626.73	2070.91

[1] Adjusted Avian $LD_{50} = LD_{50}\,(AW/TW)^{(1.15-1)}$

Based on no LOC exceedence, there is a No Effect determination for acute effects from granular uses of hexazinone.

Chronic Effects from Granular Applications

Potential direct chronic effects of granular hexazinone applications to the terrestrial-phase CRLF are derived by considering dietary-based exposures modeled in T-REX for a small bird (20g) consuming small invertebrates. Chronic effects are estimated using the lowest available toxicity data for birds. EECs are divided by toxicity values to estimate chronic dietary-based RQs.

As shown in Table 5.8, chronic RQs, which range from 1.35 to 5.48, exceed LOCs for modeled granular uses of hexazinone. Therefore, the preliminary effects determination is "may effect" for direct chronic effects to the terrestrial-phase CRLF.

Table 5.8 Summary of Chronic RQs* Used to Estimate Direct Effects to the Terrestrial-phase CRLF (granular application)

Uses	Application rate (lb ai/A)	Dietary-based Chronic RQ[1]
Non-crop:Non-agricultural ROW areas	12	**5.4**
Forest Site Preparation	5	**2.25**
Rangeland	3	**1.35**

* = LOC exceedences (chronic RQ ≥ 1) are bolded and shaded. [1] Based on bobwhite quail chronic reproduction NOAEC = 300 mg/kg-diet (MRID 417649-01).

5.1.3.2 Indirect Effects to Terrestrial-Phase CRLF via Reduction in Prey (mammals, and frogs)

Mammals

Risks associated with ingestion of small mammals by large terrestrial-phase CRLFs are derived for dietary-based and dose-based exposures modeled in T-REX for a small mammal (15g) consuming short grass. Acute and chronic effects are estimated using the most sensitive mammalian toxicity data. EECs are divided by the toxicity value to estimate acute and chronic dose-based RQs as well as chronic dietary-based RQs. The potential for acute and chronic indirect effects for the terrestrial-phase CRLF via reduction in small mammals is shown in Table 5.9. All non-granular uses result in RQs that exceed the acute and chronic LOCs. Chronic dose-based RQs range from 11.45 to 83.32, while dietary based chronic RQs range from 1.32 to 9.60. Acute RQs range from 0.22 to 1.57. Therefore, the preliminary effects determination for indirect effects to terrestrial-phase CRLFs via reduction in small mammals (exposed to non-granular applications of hexazinone) as dietary food items is "may affect" for both acute and chronic exposure for noncrop uses, conifer release, Christmas trees, pineapple and blueberry. There were chronic LOC exceedences for alfalfa and pasture, but not acute exceedences.

Table 5.9 Summary of Acute and Chronic RQs* Used to Estimate Indirect Effects to the Terrestrial-phase CRLF via Direct Effects on Small Mammals as Dietary Food Items (non-granular application)

Use (Application Rate)	Chronic RQ		Acute RQ
	Dose-based Chronic RQ[1]	Dietary-based Chronic RQ[2]	Dose-based Acute RQ[3]
Noncrop:Agricultural ROW, Agricultural uncultivated, airport (8 lbs/A)	**83.32**	**9.60**	**1.57**
Blueberry, Rangeland, conifer release (3 lbs/A)	**31.23**	**3.60**	**0.59**
Christmas Trees (2 lbs/A)	**20.82**	**2.40**	0.39
Alfalfa (1.5 lb/A)	**15.62**	**1.80**	0.29
Pasture (1.125 lbs/A)	**11.45**	**1.32**	0.22

* = LOC exceedences (acute RQ ≥ 0.1 and chronic RQ ≥ 1) are bolded and shaded.
[1] Based on dose-based EEC and Hexazinone rat NOAEL = 10 mg/kg-bw.
[2] Based on dietary-based EEC and Hexazinone rat NOAEC = 200 mg/kg-diet.
[3] Based on dose-based EEC and Hexazinone rat acute oral LD_{50} = 530 mg/kg-bw.

Frogs

An additional prey item of the adult terrestrial-phase CRLF is other species of frogs. In order to assess risks to these organisms, dietary-based and dose-based exposures modeled in T-REX for a small bird (20g) consuming small invertebrates are used. The RQs from the T-REX model are discussed presented in section 5.1.2.1, Therefore the preliminary effects determination for indirect effects to terrestrial-phase CRLFs via reduction in small amphibians (exposed to non-granular applications of hexazinone as dietary food items) is "may affect".

5.1.3.3 Indirect Effects to CRLF via Reduction in Terrestrial Plant Community (Riparian and Upland Habitat)

Potential indirect effects to the CRLF resulting from direct effects on riparian and upland vegetation are assessed using RQs from terrestrial plant seedling emergence and vegetative vigor EC_{25} data as a screen. Based on the results of the submitted terrestrial plant toxicity tests, it appears that dicot plants are more sensitive in the e seedling emergence test than the vegetative vigor test. However, all tested plants, with the exception of cucumber (invalid), exhibited adverse effects in both the seedling emergence and vegetative vigor test, following exposure to hexazinone. The results of these tests indicate that a variety of terrestrial plants that may inhabit riparian and upland zones may be sensitive to hexazinone exposure.

For monocot and dicot plants inhabiting dry and semi-aquatic areas, the LOC (RQ $\geq$ 1.0) is exceeded for exposures resulting from applications of all non-granular and granular uses of hexazinone (Tables 5.10 and 5.11). Dry area RQs range from 3.47 to 31.58 and semi-aquatic area RQs from 29.53-315.79 for monocots. Dry area RQs range from 10.31 to 93.75 and semi-aquatic areas RQs from 87.66 to 937.5 for dicots. In addition, spray drift RQs exceed LOCs for all non-granular uses of hexazinone for dicot plants. Spray drift RQs exceed LOCs for all monocot plants except for pasture. For both monocots and dicots, spray draft does not exceed for non-crop/non-agricultural ROW, forest site preparation and rangeland granular applications. The preliminary effects determination for indirect effects to terrestrial- and aquatic-phase CRLFs via reduction in the terrestrial plant community is "may affect".

Table 5.10 RQs* for Monocots Inhabiting Dry and Semi-Aquatic Areas Exposed to Hexazinone via Runoff and Drift

Use	Application rate (lbs a.i./A)	Application method	Drift Value (%)	Spray drift RQ	Dry area RQ	Semi-aquatic area RQ
Non-crop/Non-agricultural ROW	12	Granular Spray		<0.1	**31.58**	**315.79**
Agricultural	8	Spray	0.05	**21.05**	**42.11**	**231.58**

uncultivated, Airports, Sewage						
Forest Site Preparation	5.0	Granular	0	<0.1	**13.16**	**131.58**
Ag ROW, Sugarcane	3.625	Liquid	0.05	**9.54**	**19.08**	**104.93**
Blueberries/Conifer release	3	Spray	0.05	**7.89**	**15.79**	**86.84**
Rangeland	3	Granular	0	<0.1	**7.89**	**78.95**
Christmas Trees	2	Spray	0.01	**1.05**	**6.32**	**53.68**
Alfalfa	1.5	Spray	0.05	**3.95**	**7.89**	**43.42**
Pasture	1.1	Spray	0.01	.0.58	**3.47**	**29.53**
* = LOC exceedences (RQ ≥ 1) are bolded and shaded.						

Table 5.11 RQs* for Dicots Inhabiting Dry and Semi-Aquatic Areas Exposed to Hexazinone via Runoff and Drift

Use	Application rate (lbs a.i./A)	Application method	Drift Value (%)	Spray drift RQ	Dry area RQ	Semi-aquatic area RQ
Non-crop/Non-agricultural ROW	12	Granular	0	<0.1	**93.75**	**937.50**
Agricultural uncultivated, Airports, Sewage	8	Spray	0.05	**62.50**	**125.00**	**687.50**
Forest Site Preparation	5.0	Granular	0	<0.1	**39.06**	**390.63**
Ag ROW, Sugarcane	3.625	Spray	0.05	**28.32**	**56.64**	**311.52**
Blueberries/conifer release	3	Spray	0.05	**23.44**	**46.88**	**257.81**
Rangeland	3	Granular	0	<0.1	**23.44**	**234.38**
Christmas Trees	2	Spray	0.01	**3.13**	**18.75**	**159.38**
Alfalfa	1.5	Spray	0.05	**11.72**	**23.44**	**128.91**
Pasture	1.1	Spray	0.01	**1.72**	**10.31**	**87.66**
* = LOC exceedences (RQ ≥ 1) are bolded and shaded.						

5.1.4 Primary Constituent Elements of Designated Critical Habitat

5.1.4.1 Aquatic-Phase (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)

Three of the four assessment endpoints for the aquatic-phase primary constituent elements (PCEs) of designated critical habitat for the CRLF are related to potential effects to aquatic and/or terrestrial plants:

- Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.

- Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.

- Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.,* algae).

The preliminary effects determination for aquatic-phase PCEs of designated habitat related to potential effects on aquatic and/or terrestrial plants is "may affect", based on the risk estimation provided in Sections 5.1.1.2, 5.1.1.3, and 5.1.2.3.

The remaining aquatic-phase PCE is "alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source." To assess the impact of hexazinone on this PCE, acute and chronic freshwater fish and invertebrate toxicity endpoints, as well endpoints for aquatic non-vascular plants are used as measures of effects. RQs for these endpoints were calculated in Sections 5.1.1.1 and 5.1.1.2. Based on these results, the preliminary effects determination for alteration of characteristics necessary for normal growth and viability of the CRLF is "no effect" (see Section 5.1.1.1). However, aquatic vascular and non-vascular aquatic plant food items of the CRLF may be affected; therefore the preliminary effects determination for potential impacts to these food items is "may affect" (see Section 5.1.1.2).

5.1.4.2 Terrestrial-Phase (Upland Habitat and Dispersal Habitat)

Two of the four assessment endpoints for the terrestrial-phase PCEs of designated critical habitat for the CRLF are related to potential effects to terrestrial plants:

- Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance

- Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal

The preliminary effects determination for terrestrial-phase PCEs of designated habitat related to potential effects on terrestrial plants is "habitat modification", based on the risk estimation provided in Section 5.1.2.3.

The third terrestrial-phase PCE is "reduction and/or modification of food sources for terrestrial phase juveniles and adults." To assess the impact of hexazinone on this PCE, acute and chronic toxicity endpoints for birds, mammals, and terrestrial invertebrates are used as measures of effects. RQs for these endpoints, which were calculated in Section 5.1.2.2, exceed the LOCs for all hexazinone non-granular uses. Uses of hexazinone are not expected to cause direct acute effects to frog prey items of the terrestrial-phase CRLF; however, chronic effects to small insectivorous mammals that ingest granules may occur. The preliminary effects determination for habitat modification via impacts of non-granular uses of hexazinone to terrestrial-phase CRLF food items is "habitat modification".

The fourth terrestrial-phase PCE is based on alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source. Direct acute effects, via mortality, are expected for the terrestrial-phase CRLF (see Section 5.2.1.2). Chronic reproductive effects are possible for all non-granular uses of hexazinone. Therefore the preliminary effects determinations for habitat modification are "habitat modification" via direct acute effects to terrestrial-phase CRLFs and "habitat modification" based on chronic exposures to non-granular applications of hexazinone.

5.2 Risk Description

The risk description synthesizes an overall conclusion regarding the likelihood of adverse impacts leading to an effects determination (*i.e.*, "no effect," "may affect, but not likely to adversely affect," or "likely to adversely affect") for the CRLF and its designated critical habitat.

If the RQs presented in the Risk Estimation (Section 5.1) show no direct or indirect effects for the CRLF, and no modification to PCEs of the CRLF's designated critical habitat, a "no effect" determination is made, based on hexazinone's use within the action area. However, if direct or indirect effects LOCs are exceeded or there are modifications to PCEs of the CRLF's critical habitat, the Agency concludes a preliminary "may affect" determination for the FIFRA regulatory action regarding hexazinone.

Following a "may affect" determination, additional information is considered to refine the potential for exposure at the predicted levels based on the life history characteristics (*i.e.*, habitat range, feeding preferences, etc.) of the CRLF. Based on the best available information, the Agency uses the refined evaluation to distinguish those actions that "may affect, but are not likely to adversely affect" from those actions that are "likely to adversely affect" the CRLF and its designated critical habitat.

The criteria used to make determinations that the effects of an action are "not likely to adversely affect" the CRLF and its designated critical habitat include the following:

- Significance of Effect: Insignificant effects are those that cannot be meaningfully measured, detected, or evaluated in the context of a level of effect where "take" occurs for even a single individual. "Take" in this context means to harass or harm, defined as the following:

    - Harm includes significant habitat modification or degradation that results in death or injury to listed species by significantly impairing behavioral patterns such as breeding, feeding, or sheltering.
    -
    - Harass is defined as actions that create the likelihood of injury to listed species to such an extent as to significantly disrupt normal behavior patterns which include, but are not limited to, breeding, feeding, or sheltering.

- Likelihood of the Effect Occurring: Discountable effects are those that are extremely unlikely to occur.

- Adverse Nature of Effect: Effects that are wholly beneficial without any adverse effects are not considered adverse.

A description of the risk and effects determination for each of the established assessment endpoints for the CRLF and its designated critical habitat is provided in Sections 5.2.1 through 5.2.3.

Table 5.12 Effects Determination Summary for Direct and Indirect Effects of Hexazinone on the CRLF		
Assessment Endpoint	**Effects Determination[1]**	**Basis for Determination**
Aquatic-Phase CRLF (Eggs, Larvae, and Adults)		
Direct Effect s of Hexazinone on the Aquatic-Phase CRLF		
Survival, growth, and reproduction of CRLF individuals via direct effects on aquatic phases	Using fish as a surrogate: No Effect	Using freshwater fish as a surrogate, no acute and chronic LOCs are exceeded for applications of Non-agricultural Rights–of-way (12 lb/A-granular), there is no expectation for adverse effects for lower rates: noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas Tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
Indirect Effect s of Hexazinone on the Aquatic –Phase CRLF		

Survival, growth, and reproduction of CRLF individuals via effects to food supply (*i.e.,* freshwater invertebrates, non-vascular plants, fish, and frogs)	Freshwater invertebrates: No effect	Using freshwater invertebrates, no acute and chronic LOCs are exceeded for applications of Non-agricultural Rights–of-way (12 lb/A-granular). Due to no exceedence for the 12 lb/A rate, it is assumed that there would also be no exceedence for the lower rates: Noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
	Indirect Effects of Prey Reduction for Non-vascular aquatic plants for all uses: May Affect Non-vascular aquatic plants: No Effect	The May Affect for hexazinone uses related to applications for non-agricultural ROW (12 lb/A)non-crop uses (8 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), rangeland (3 lbs/A), Christmas trees (2.0 lb ai/A), alfalfa (1.5 lb ai/A) and pasture (1.1lb ai/A), exceed LOCs; therefore, indirect effects to tadpoles that feed on algae are possible. RQs range from 22.37 to 1.52. Blueberry uses (RQ = 0.81) resulted in no LOC exceedence.
	Indirect Effects of Prey Reduction for Fish as surrogate for Frogs for all uses: No effect	Using freshwater fish as a surrogate, no acute and chronic LOCs are exceeded for applications of non-crop/non-agricultural Rights–of-way (12 lb/A-granular), Noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A). .

Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat, cover, and/or primary productivity (*i.e.,* aquatic plant community)	Non-vascular aquatic plants: May Affect No Effect	Hexazinone uses related to applications on Non-crop/non-agricultural Rights–of-Way (12 lb/A-granular), Noncrop uses (8 lb/A), forest site preparation (5 lb/A), pineapple (3.6 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas Tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) exceed LOCs. There is no LOC exceedence for blueberry (3 lb/A) (RQ=0.81).
	Indirect Effects on habita for Vascular aquatic plants for all uses: May Affect No Effect	The "May Affect" is based on the LOC exceedence for vascular aquatic plants for liquid applications of hexazinone to non-crop/non-agricultural ROW (12 lb/A), non-crop uses (8 lbs/A), Forest Site Preparation (5 lb/A), Conifer Release (3 lb/A).RQs range from 4.19 to 1.25. No LOC exceedence resulted for blueberry (3 lb/A), rangeland (3 lb/A) Christmas tree (2 lb/A), alfalfa (1.5 lb/A) or pasture (1.1 lb/A).
Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation, required to maintain acceptable water quality and habitat in ponds and streams comprising the species' current range.	Forested and grassy/herbaceous riparian vegetation: May Affect	Riparian vegetation may be affected because terrestrial plant RQs exceed LOCs. RQs for semi-aquatic areas range from 87.66 to 937.50 Hexazinone effects on shading, bank stabilization, and structural diversity of riparian areas in the action area are expected. Aquatic-phase CRLFs may be indirectly affected by adverse effects to sensitive herbaceous vegetation (based on all hexazinone uses), which provides habitat and cover for the CRLF and attachment sites for its egg masses.
Terrestrial-Phase CRLF (Juveniles and adults)		
Direct Effect s of Hexazinone on the Terrestrial-Phase CRLF		

Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	Nongranular acute for medium size frog using the bird as a surrogate: May affect No Effect for medium sized frog	An adverse effect is expected based on weight of evidence for acute avian toxicity. The T-Rex analysis resulted in an endangered species exceedence. A "No effect"determination for pasture (1.1 lb/A) is due to no LOC exceedence using the bird as a surrogate for the CRLF for the 37 g animal resulting from the T-REX analysis.
	Nongranular Acute Small Frog May Affect:	The May Affect resulted from LOC exceedence using the bird as a surrogate in the T-Herps analysis.
	Nongranular Acute Large Frog: May Affect:	The May Affect resulted from LOC exceedence using the bird as a surrogate in the T-HERPS analysis.
	Acute Granular for direct effects on the CRLF: No Effect	The no effect determination is due to no LOC exceedence using the bird as a surrogate from the LDft^2 results for noncrop non-agricultural ROW, forest site preparation (5 lb/A) and rangeland (3 lb/A).

	Nongranular Chronic Direct Effects Using the Bird as a Surrogate: May Affect:	The May affect is based on non-listed LOC exceedence using the bird as a surrogate from the T-REX analysis.
	Granular: May Affect	Granular formulations for non-crop/non-agricultural ROW, forest site preparation and rangeland resulted in LOC exceedences resulting from the T-REX analysis. RQs range from 1.35 to 5.4.
Indirect Effect s of Hexazinone on the Terrestrial –Phase CRLF		
	Nongranular Acute Terrestrial Invertebrates (Large Insect Prey): May Affect Nongranular Acute No Effect	An May Affect determination for large insect prey is based on the uncertainty regarding the effects of hexazinone on terrestrial invertebrates due to related toxicity values reporting no mortality at the highest concentration tested for noncrop (8 lb/A), pineapple (3.6 lb/A) and blueberry/conifer release (3 lb/A). A "No effect" determination for large insect prey for Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) uses was based on no LOC exceedence from the T-REX analysis.

	Indirect prey reduction: Mammals: Non-granular Acute May Affect Granular Acute Mammal Prey Reduction: No Effect (granular uses)	The May Affect is based on T-Rex LOC exceedence results for 15 g mammal with a diet of shortgrass for non-crop (8 lb/A) through pasture (1.1 lb/A) uses. A No effect determination is based on the LDft[2] results from the T-REX analysis for non-crop/non-agricultural ROW (12 lb/A), forest site preparation (5 lb/A) and rangeland (3 lb/A).
	Indirect prey reduction-Mammal Nongranular Chronic: May Affect:	Chronic reproductive effects are possible, based on non-granular uses of hexazinone. The May affect is due to LOC exceedences from the T-Rex analysis for all nongranular uses.
	Amphibian using the bird as a surrogate Nongranular Acute May Affect	The May affect is due to LOC exceedence using the bird as a surrogate from the T-Rex analysis.

	<u>Amphibian using the bird as a surrogate</u> <u>Nongranular Chronic</u> May Affect:	Chronic reproductive effects are possible, based on non-granular uses of hexazinone resulting from the T-REX analysis using the bird as a surrogate.
	<u>Amphibian using the bird as a surrogate</u> Nongranular Chronic No Effect	Christmas tree, alfalfa and pasture did not result in LOC exceedences broadleaf. RQs for broadleaf range from 0.5 (3 lb/A) to 0.90 (1.5 lb/A) resulting from the T-Rex analysis. .
Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.,* riparian vegetation)	<u>Woody and grassy/herbaceous riparian vegetation:</u> May Affect	Riparian woody and herbaceous vegetation may be affected because terrestrial plant RQs are above LOCs.

Table 5.13 Effects Determination Summary for the Critical Habitat Impact Analysis		
Assessment Endpoint	**Effects Determination**	**Basis for Determination**
Aquatic-Phase PCEs (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)		
Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile	Habitat modification	Due to the MOA which interferes with photosynthesis and RNA ,sensitive herbaceous riparian vegetation may be affected based on all modeled uses of hexazinone; therefore, critical habitat may be modified by an increase in sediment

and adult CRLFs.		deposition and reduction in herbaceous riparian vegetation that provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult aquatic-phase CRLFs.
Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.[7]	Habitat modification	Sensitive non-vascular aquatic plants may be affected; therefore, critical habitat may be modified via turbidity and reduction in oxygen content necessary for normal growth and viability of juvenile and adult aquatic-phase CRLFs.
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	No effect to growth and viability Habitat modification based on alteration of food source	Direct effects to the aquatic-phase CRLF, via mortality, growth, and/or fecundity, are not expected. However, critical habitat of the CRLF may be modified via hexazinone-related impacts to non-vascular aquatic plants as food items for tadpoles. LOCs are exceeded for non-vascular uses for non-crop/non-agricultural ROW (12 lb/A), noncrop (8 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas Trees (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
	No Habitat Modification	There was no LOC exceedence for blueberry uses for nonvascular plants.
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.,* algae)	Habitat modification No Habitat Modification	Based on the results of the effects determinations for aquatic plants, critical habitat of the CRLF may be modified via hexazinone-related impacts to non-vascular aquatic plants as food items for tadpoles. LOCs are exceeded for uses for non-crop/non-agricultural ROW (12 lb/A), noncrop uses (8 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas trees (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A). Blueberry use resulted in no LOC exceedence.
Terrestrial-Phase PCEs (Upland Habitat and Dispersal Habitat)		

[7] Physico-chemical water quality parameters such as salinity, pH, and hardness are not evaluated because these processes are not biologically mediated and, therefore, are not relevant to the endpoints included in this assessment.

Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance	Habitat modification	Based on MOA, modification to critical habitat may occur via hexazinone-related impacts to sensitive woody and herbaceous vegetation, which provide habitat and cover for the terrestrial-phase CRLF and its prey, based on all assessed uses of hexazinone**.** Terrestial incident reports support a "habitat modification" determination.
Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal	Habitat modification	Based on the MOA for hexazione modification to dispersal habitat may occur via hexazinone-related impacts to sensitive woody and herbaceous vegetation, which provide habitat and cover for the terrestrial-phase CRLF and its prey, based on all assessed uses of hexazinone**.** Terrestrial incident reports support the "habitat modification' determination.
Reduction and/or modification of food sources for terrestrial phase juveniles and adults	Habitat modification	Based on the characterization of indirect effects to terrestrial-phase CRLFs via reduction in the prey base, critical habitat may be modified via a reduction in mammals and terrestrial-phase amphibians as food items.
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	Habitat modification	Direct acute effects, via mortality, are expected for the terrestrial-phase CRLF. Chronic reproductive effects are also possible. Therefore, hexazinone may adversely critical habitat by altering chemical characteristics necessary for normal growth and viability of terrestrial-phase CRLFs and their mammalian and amphibian food sources.

5.2.1 Direct Effects

5.2.1.1 Aquatic-Phase CRLF

The aquatic-phase considers life stages of the frog that are obligatory aquatic organisms, including eggs and larvae. It also considers submerged terrestrial-phase juveniles and adults, which spend a portion of their time in water bodies that may receive runoff and spray drift containing hexazinone.

As shown in Table 5.1, acute and chronic RQs based on the highest modeled EECs for hexazinone use on non-crop/non-agricultural ROW (12 lb ai/A) and the most sensitive freshwater fish data (used as a surrogate for aquatic-phase amphibians) are well below the Agency's acute and chronic risk LOCs.

All uses resulted in no RQs exceeding either the acute or chronic LOCs for fish.

No open literature was available that provided more sensitive endpoints than registrant submitted studies. No relevant incident reports are available for hexazinone. In summary, the Agency concludes a "no effect" determination for direct effects to the aquatic-phase CRLF, via mortality, growth, or fecundity, based on all available lines of evidence.

5.2.1.2 Terrestrial-Phase CRLF

Direct Acute Effects Determination

Direct acute terrestrial effects were further refined following the "May Affect" determination using the T-Herps model to adjust for size and dietary effects.Based on acute avian toxicity data as a surrogate for the terrestrial-phase amphibians, direct acute mortality is expected for the terrestrial-phase CRLF via exposure to hexazinone applications due to LOC exceedences resulting from the T-Herps refinement analysis (Table 5.16).

For the medium-sized frog (37 g) there is an "LAA" determination for acute indirect effects using the 20 g bird as a surrogate based on T-Herps small herbivore mammal results for noncrop uses (8 lb/A).

Pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas tree (2 lb/A) uses for small herbivore mammals resulted in RQs falling between the listed and non-listed LOCs. Therefore, the probit analysis will be used to further analyze those uses.

There was a "LAA" determination based on the probit results for pineapple, blueberry, conifer release and Christmas tree uses. The probability of an individual effect to the bird as as a surrogate for the CRLF for pineapple, blueberry and conifer release with a slope of 4.2132 were ~1 in 222.

There was a "NLAA" determination due to discountable effects based on the probability of an individual effect to the bird as as a surrogate for the CRLF for Christmas tree from the probit analysis. Usinga slope of 4.2132, there was ~1 in 10,600.

There is an NLAA determination for alfalfa and pasture uses for the 37g frog consuming small herbivore mammals due to no LOC exceedence resulting from the T-Herps analysis.

There is an NLAA determination for the small frog (1.4 g) for all scenarios due to no LOC exceedence resulting from the T-Herps analysis.

There is an NLAA determination for the large frog (238 g) for all scenarios due to no LOC exceedence resulting from the T-Herps analysis.

There is a no effect determination for acute direct effects for the terrestrial-phase CRLF based on results for non-crop/non-agricultural ROW, forest site preparation and rangeland granular applications for which LOCs were not exceeded resulting from the T-Rex LDft^2 analysis, as well as no reported terrestrial animal incidents.

Table 5.16. Assessment of Direct Effects on California Red-legged Frog (CRLF) Using Dose-Based Estimated Environmental Concentrations (EECs) of Hexazinone Based on the T-Herps Model for Small (1.4 g), Medium (37 g), and Large (238 g) CRLF.						
		RQs (mg/kg-bw) for Small, Medium, and Large CRLF (Small / Medium / Large)				
Use	**Maximum Application Rates (Lbs. ai/A)**	**Broadleaf Plants/Small Insects**	**Fruit/ Pods/ Seeds/ Large Insects**	**Small Herbivore Mammals**	**Small Insectivore Mammals**	**Small Terrestrial Phase Amphibian**
Noncrop: airport, Ag ROW	8	0.02 / 0.02 / 0.01	0.0 / 0.0 / 0.0	N.A./**0.53*** / 0.08	N.A. / 0.03 / 0.01	N.A. / 0.0 / 0.0
Pineapple	3.6	0.01/0.01/0.01	0.0/0.0/0.0/	N.A./0.**24***/0.04	N.A.//0.01/0.0	N.A./0.0/0.0/
Blueberry/ conifer release	3	0.01 / 0.01 / 0.0	.0 /0.0 /0.0	N.A. / **0.20*** /0.03	N.A. / .01 / 0.0	N.A. / 0.0 /0.0
Christmas Tree	2	0.0 / 0.0 / 0.0	0.0 /0.0 /0.0	N.A. / **0.13*** / 0.02	N.A. / 0.01 /0.0	N.A. / .0.0/0.0
Alfalfa	1.5	0.0 /0.0 / 0.0	.0.0/ 0.0 /0.0	N.A. / 0.1 /0.02	N.A. / 0.1 /0.0	N.A. / .0.0/ 03.0
Pasture	1.1	0.0 / 0.0 / 0.0	.0.0 / 0.0 /0.0	N.A. / 0.07 /0.01	N.A. /0.0 /0.0	N.A. /0.0 /0.0
* RQ > acute endangered species LOC of 0.1.						

Direct Chronic Terrestrial Effects Determination

Direct chronic terrestrial effects were further refined following the “May Affect” determination using the T-Herps model to adjust for size and dietary effects. Table 5.17 summaraizes the effects of chronic hexazinone exposure using the bird as a surrogate for the CRLF.There is an LAA determination for the CRLF using the bird as a surrogate for broadleaf plants/small insects for noncrop, conifer release and blueberry uses due to LOC exceedence resulting from the T-Herps analysis.

.

There is an LAA determination for the CRLF using the bird as a surrogate consuming small herbivore mammals for noncrop, conifer release, blueberry and Christmas tree uses due to LOC exceedence resulting from the T-Herps analysis.

.

There is an NLAA determination for the CRLF using the bird as a surrogate consuming broadleaf/small insects for Christmas tree, alfalfa and pasture uses due to no LOC exceedence resulting from the T-Herps analysis.

.

There is an NLAA determination for the CRLF using the bird as a surrogate consuming small herbivore mammals for alfalfa and pasture uses due to no LOC exceedence resulting from the T-Herps analysis.
.
There is an NLAA determination for the CRLF using the bird as a surrogate consuming fruits/seeds/large insects, small insectivore mammals and small amphibians as prey due to no LOC exceedence resulting from the T-Herps analysis.

Table 5.17 Assessment of Direct effects on Terrestrial phase California Red-legged Frog (CRLF) Using Dietary based Chronic Risk Quotients (RQs) From the T-Herps Model for Hexazinone.

Scenario Group: Crop/Site	Chronic Dietary-based RQs for Terrestrial Phase CRLF Direct Effects				
	Small Insect Prey	Large Insect Prey	Small Herbivore Mammal Prey	Small Insectivore Mammal Prey	Small Terrestrial Phase Amphibian Prey
NonCrop (8 lb/A)	**3.60***	0.40	**4.22***	0.26	0.12
Blueberry /conifer release (3 lb/A)	**1.35***	.0.15	**1.58***	0.10	0.05
Christmas Trees (2 lb/A)	.0.90	0.10	**1.05***	0.07	0.03
Alfalfa (1.5 lb/A)	0.68	.0.08	0.79	0.05	0.02
Pasture (1.1 lb/A)	0.50	0.06	0.58	0.04	0.02

* RQ > chronic LOC of 1

5.2.2 Indirect Effects (via Reductions in Prey Base)

5.2.2.1 Algae (non-vascular plants)

As discussed in Section 2.5.3, the diet of CRLF tadpoles is composed primarily of unicellular aquatic plants (*i.e.*, algae and diatoms) and detritus. Based on RQs for algae (Table 5.2), liquid applications of hexazinone to pasture (1.1 lb/A), alfalfa (1.5 lb/A), Christmas trees (2 lb a/A), blueberry (3 lb ai/A), conifer release (3 lb ai/A), and granular applications of hexazinone to non-crop/non-agricultural ROW (12 lb/A), forest site preparation (5lb/A) and rangeland (3 lb ai/A) may affect this food source. RQs for non-vascular plants were based on the most sensitive EC_{50} value of 7 µg/L for freshwater algae (*Selenastrum*).

Typically, algal populations are relatively dynamic, although the presence of hexazinone in the water may result in an overall reduction in biomass, and/or a shift in community composition as more sensitive species are eliminated. Although recovery of algal populations has been shown to occur, if the timing of hexazinone applications co-occur with the presence of tadpole life stages of the CRLF (from December to March), a reduction in algae as a food source for the tadpole may occur.

There is an LAA determination for indirect dietary effects based on nonagricultural ROW, noncrop, conifer release, Christmas tree, alfalfa and pasture for nonagricultural ROW, noncrop, conifer release and Christmas tree uses and mode of action for non-vascular plants based on LOC exceedence and mode-of-action for hexazinone.

There is a no effect determination for indirect effects for the aquatic-phase CRLF based on blueberry using non-vascular plants resulting from no LOC exceedence, as well as no open literature or reported incidents.

5.2.2.2 Aquatic Invertebrates

The potential for hexazinone to elicit indirect effects to the CRLF via effects on freshwater invertebrate food items is dependent on several factors including: (1) the potential magnitude of effect on freshwater invertebrate individuals and populations; and (2) the number of prey species potentially affected relative to the expected number of species needed to maintain the dietary needs of the CRLF. Together, these data provide a basis to evaluate whether the number of individuals within a prey species is likely to be reduced such that it may indirectly affect the CRLF.

As previously discussed in Section 5.1.1.2, acute RQs for the highest application rate (industrial outdoor uses at 10 lbs/A)Table 5.3) calculated using modeled peak aquatic EECs and the 48-hour EC_{50} for the water flea, *Daphnia magna*, do not exceed the acute LOC for hexazinone exposure.

Chronic RQs for invertebrates were less than the Agency's LOC, based on the highest 21-day modeled EECs for all hexazinone uses Therefore, chronic risks to freshwater invertebrates and potential indirect effects to aquatic-phase CRLFs that consume them as prey are not expected.

All uses resulted in no RQs exceeding either the acute or chronic LOCs for aquatic invertebrates. No open literature was available that provided more sensitive endpoints than registrant submitted studies. No incident reports for aquatic invertebrates are available for hexazinone. In summary, the Agency concludes a “no effect” determination for indirect dietary effects to the aquatic-phase CRLF, via mortality, growth, or fecundity of aquatic invertebrates, based on all available lines of evidence.

5.2.2.3 Fish and Aquatic-phase Frogs

No endangered species acute or chronic LOCs were exceeded for any uses for freshwater fish used as a surrogate for aquatic-phase amphibians. Therefore, indirect effects to the CRLF via a reduction in freshwater fish and other aquatic-phase frog species as prey items is not expected, and the effects determination for this assessment endpoint is "no effect". No incident reports are available for fish or aquatic-phase frogs. No open literature provides a more sensitive endpoint than registrant data for fish or frogs.

5.2.2.4 Terrestrial Invertebrates

When the terrestrial-phase CRLF reaches juvenile and adult stages, its diet is mainly composed of terrestrial invertebrates.

In order to assess the risks of hexazinone to terrestrial invertebrates the honey bee is used as a surrogate for terrestrial invertebrates. The toxicity value for terrestrial invertebrates is calculated by multiplying the lowest available acute contact LD_{50} of >100 µg a.i. /bee by 1 bee/0.128g, which is based on the weight of an adult honey bee. EECs (µg a.i. /g of bee) calculated by T-REX for small and large insects are divided by the calculated toxicity value for terrestrial invertebrates, which is >781 µg a.i. /g of bee. Given that the toxicity endpoint is non-definitive (*i.e.*, the LD_{50} value is greater than the highest test concentration), the reported RQ values represent an upper bound. The resulting non-definitive RQ values for large insect and small insect exposures bound the potential range of exposures for terrestrial insects to hexazinone. RQs may exceed the LOC for small insects, with RQs ranging from <1.57 to <0.22. With the exception of the 1.1, 1.5 and 2 hexazinone use rates (for pasture, alfalfa and Christmas trees) on large insects, RQ values may exceed the LOC (RQ ≥ 0.05). Christmas tree (<0.04), alfalfa (<0.03) and pasture (<0.02) resulted in no RQ exceedence (Table 5.18).

In addition to calculating RQs, other sources were reviewed for weight of evidence.

No probit analysis was conducted for terrestrial invertebrate effects due to no observed dose-response. No open literature for terrestrial invertebrates was available to provide additional information. No relevant incident reports were available for terrestrial invertebrates.

Table 5.18 Summary of RQs Used to Estimate Indirect Effects to the Terrestrial-phase CRLF via Direct Effects on Terrestrial Invertebrates as Dietary Food Items

Use	Small Insect RQ*	Large Insect RQ*
Agricultural uncultivated, airport (8 lbs/A)	**<1.57**	**<0.17**
Pineapple (3.6 lbs/A)	**<0.71**	**<0.08**
Blueberry, Conifer release, Rangeland (3 lbs/A)	**<0.59**	**<0.06**
Christmas Trees (2 lbs/A)	**<0.39**	<0.04
Alfalfa (1.5 lbs/A)	**<0.26**	<0.03
Pasture (1.12 lbs/A)	**<0.22**	<0.02

* = LOC exceedences (RQ ≥ 0.05) are bolded and shaded. Because a definitive endpoint was not established for terrestrial invertebrates (i.e., the value is greater than the highest test concentration), the RQ represents an upper bound value.

There is uncertainty based on no mortality at the highest tested concentration for the toxicity study, which is reflected in the results for this analysis. The RQs need to exceed an LOC = 0.05 to establish an adverse effect. RQs for Christmas tree, alfalfa and pasture are below the LOC.

There is an "LAA" determination for indirect small insect dietary effects for terrestrial invertebrates based on the uncertainty due to no mortality at the highest tested concentration for the honeybee toxicity study for all scenarios. There was no acceptable open literature or reported incidents for terrestrial invertebrates.

5.2.2.5 Mammals

Life history data for terrestrial-phase CRLFs indicate that large adult frogs consume terrestrial vertebrates, including mice. A reported acute oral rat LD_{50}=530 mg/kg was used to assess indirect effects of hexazinone on the CRLF (Kennedy 1984). This value was lower than the registrant submitted value of 1,200 mg/kg. The acute RQs resulting from the 15 g mammal eating short grass were further refined using the probit analysis Table 5.19).

An analysis of potential adverse mammal population affects was assessed for hexazinone uses at the LOC threshold (.1) and at the RQ between the endangered species LOC and the acute risk LOC using the acute oral rat LD_{50} of 530 mg/kg and a default probit slope of 4.5. The likelihood of individual effect for hexazinone is 1 in 294,000 (with respective upper and lower bounds of 1 in 44 and 1 in 8.86E+18).

Noncrop (RQ=1.57), pineapple (RQ=0.71) and blueberry/conifer release (RQ=0.59) uses resulted in RQs exceeding the non-listed LOC (0.5) and therefore a "LAA" determination was concluded.

Although RQs for Christmas tree, alfalfa and pasture did not exceed the nonlisted LOC, RQs fell between the endangered species and nontarget LOCs. Therefore, these uses were also refined using the probit analysis.

For Christmas tree uses, the corresponding estimated chance of an individual acute mortality/immobilization to a terrestrial mammal at an RQ level of 0.39 is 1 in 30.4 (with respective upper and lower bounds of 1 in 859 and 1 in 4.84).

For alfalfa uses, the corresponding estimated chance of an individual acute mortality/immobilization to a terrestrial mammal at an RQ level of 0.29 is 1 in 129 (with respective upper and lower bounds of 1 in 7.09 and 1 in 1.04).

For pasture uses, the corresponding estimated chance of an individual acute mortality/immobilization to a terrestrial mammal at an RQ level of 0.22 is 1 in 648 (with respective upper and lower bounds of 1 in 1,530,000 and 1 in 1.53).

Based on this assessment, the potential reduction in abundance of terrestrial mammals as food for these uses would be < 1% at most (range 0.001%-0.03%); therefore a "not likely to adversely affect" determination can be made.

Table 5.19 Hexazinone Uses That Exceed the Endangered Species LOC (Based on Mammal Toxicty Data)

Use	LOC or RQ	Likelihood of Individual Effect (1 in …)	Probability of Affect
Acute Endangered Species LOC hexazinone uses	0.1	~1 in 2.94E +05	3.4E-06
Christmas Tree	0.39	~1 in 30.4	0.0329%
Alfalfa	0.29	~1 in 129	0.008%
Pasture	0.22	~1 in 648	0.001%

[1] Hexazinone acute oral rat LD_{50}=530 mg/kg (MRID# 4074902); Probit slope 4.5 (default)
[2] Chance of individual effect and probability of affect were only calculated if the LOC was between 0.1-0.5

5.2.2.6 Terrestrial-phase Amphibians

Terrestrial-phase adult CRLFs also consume frogs. RQ values representing direct exposures of hexazinone to terrestrial-phase CRLFs are used to represent exposures of hexazinone to frogs in terrestrial habitats.

An analysis of the likelihood of individual mortality as an indirect dietary effect indicates that at the listed species LOC, *i.e.*, RQ=0.1, the likelihood of individual mortality for hexazinone is 1 in 79,000 (Table 5.20)

Table 5.20 Hexazinone Uses That Exceed the Endangered Species LOC Using the Bird As a Surrogate for Amphibians

Use	LOC or RQ	Likelihood of Individual Effect (1 in …)	Probability of Affect
Acute Endangered Species LOC hexazinone uses	0.1	~1 in 79,000	1.26E-05%
Pineapple	0.34	~1 in 41.3	0.00242%
Blueberry	0.28	~1 in 101	0.00992%
Christmas Tree	0.19	~1 in 842	0.00119%
Alfalfa	0.14	~1 in 6230	0.000161%
Pasture	0.10	~1 in 79,400	0.0000126%

[1] Hexazinone acute oral quail LD_{50} =2258 mg/kg (MRID# 0073988); Probit slope 4.2132
[2] Chance of individual effect and probability of affect were only calculated if the LOC was between 0.1-0.5

There is an "LAA" determination for indirect effects of hexazinone for the CRLF consuming amphibians based on the RQ (0.76) for noncrop uses exceeding the non-listed LOC (0.5).

Based on this assessment, the potential reduction in abundance of amphibians as food for these uses would be < 1% at most (range 0.0000161%-0.00242%); therefore a "not likely to adversely affect" determination can be made for pineapple, blueberry, Christmas tree and alfalfa.

There is a "no effect" determination for pasture (RQ=0.1) based on no LOC exceedence.

5.2.3 Indirect Effects (via Habitat Effects)

5.2.3.1 Aquatic Plants (Vascular and Non-vascular)

Aquatic plants serve several important functions in aquatic ecosystems. Non-vascular aquatic plants are primary producers and provide the autochthonous energy base for aquatic ecosystems. Vascular plants provide structure, rather than energy, to the system, as attachment sites for many aquatic invertebrates, and refugia for juvenile organisms, such as fish and frogs. Emergent plants help reduce sediment loading and provide stability to nearshore areas and lower stream banks. In addition, vascular aquatic plants are important as attachment sites for egg masses of CRLFs.

Potential indirect effects to the CRLF based on impacts to habitat and/or primary production were assessed using RQs from freshwater aquatic vascular and non-vascular plant data.

Based on RQs for non-vascular plants (previously described in Section 5.2.2.1 and summarized in Table 5.2), LOCs are exceeded for all modeled uses of hexazinone. RQs for vascular plants exceed the LOC for all uses except rangeland, blueberry, alfalfa and pasture. Therefore, indirect effects to the CRLF via direct effects to vascular plants as habitat are expected.

In summary, the overall effects determination for indirect effects of hexazinone to CRLFs via impacts to habitat and/or primary production through direct effects to non-vascular plants and vascular plants is "likely to adversely affect" or "LAA" for hexazinone exposure.

5.2.3.2 Terrestrial Plants

Terrestrial plants serve several important habitat-related functions for the CRLF. In addition to providing habitat and cover for invertebrate and vertebrate prey items of the CRLF, terrestrial vegetation also provides shelter for the CRLF and cover from predators while foraging. Upland vegetation including grassland and woodlands provides cover during dispersal. Riparian vegetation helps to maintain the integrity of aquatic systems by providing bank and thermal stability, serving as a buffer to filter out sediment, nutrients, and contaminants before they reach the watershed, and serving as an energy source.

Loss, destruction, and alteration of habitat were identified as a threat to the CRLF in the USFWS Recovery Plan (USFWS, 2002). Herbicides can adversely impact habitat in a number of ways. In the most extreme case, herbicides in spray drift and runoff from the site of application have the potential to kill (or reduce growth and/or biomass in) all or a substantial amount of the vegetation, thus removing or impacting structures which define the habitat, and reducing the functions (*e.g.*, cover, food supply for prey base) provided by the vegetation.

Hexazinone is a systemic herbicide that is absorbed by the plant through both the leaves and the roots. It acts by inhibiting photosynthesis within the targeted plant. Based on the available toxicity data for terrestrial plants, it appears that emerged dicot seedlings are similar in sensitivity to hexazinone in the vegetative vigor test. This is demonstrated by the difference in dicot response to the two guideline studies. The dicot EC_{25} values for the seedling emergence and vegetative vigor tests are 0.0064 lb ai/A and 0.011 lb ai/A, respectively. Monocots show similar levels of sensitivity in the seedling emergence and vegetative vigor toxicity tests.

Riparian vegetation typically consists of three tiers of vegetation, which include a groundcover of grasses and forbs, an understory of shrubs and young trees, and an overstory of mature trees. Frogs spend a considerable amount of time resting and feeding in riparian vegetation; the moisture and cover of the riparian plant community provides good foraging habitat, and may facilitate dispersal in addition to providing pools and backwater aquatic areas for breeding (USFWS, 2002). According to Hayes and Jennings (1988), the CRLF tends to occupy waterbodies with dense riparian vegetation including willows (*Salix* sp.). Upland habitat includes grassland and woodlands, as well as scrub/shrub habitat. While no guideline data are available on the toxicity of woody plants, the available toxicity information indicates that hexazinone is likely to cause adverse effects to non-target woody plants. In addition, hexazinone is labeled for use around fields for woody species, as well as uses associated with forestry, reforestation, conifer release and Christmas trees. Therefore, hexazinone is generally toxic to woody plants. Woody trees and shrubs in both upland and riparian habitats are expected to intercept some of the hexazinone that might otherwise be deposited on the more sensitive herbaceous species. Additionally, in natural systems, older plants, fallen leaves, and other debris often provide a litter layer, which may serve to protect newly emerging herbaceous plants.

As shown in Tables 5.10 and 5.11, RQs exceed LOCs for monocots and dicots inhabiting dry and semi-aquatic areas exposed to hexazinone via runoff and drift. In general, it appears that dicots are more sensitive than monocots to hexazinone in semi-aquatic areas. Dicots in semi-aquatic and dry areas are approximately 3 times more sensitive than monocots in similar areas; as well as sensitivity to hexazinone in spray drift. In summary, based on exceedence of the terrestrial plant LOCs for all hexazinone use patterns following runoff and spray drift to semi-aquatic and dry areas, the following general conclusions can be made with respect to potential harm to riparian habitat:

- Hexazinone may enter riparian areas via runoff and/or spray drift where it may be taken up by the plant by the leaves and roots of sensitive plants.

- Comparison of seedling emergence EC_{25} values to EECs estimated using Terrplant suggests that existing vegetation may be affected or inhibition of new growth may occur. Inhibition of new growth could result in degradation of high quality riparian habitat over time because as older growth dies from natural or anthropogenic causes, plant biomass may be prevented from being replenished in the riparian area. Inhibition of new growth may also slow the recovery of degraded riparian areas that function poorly due to sparse vegetation because hexazinone deposition onto bare soil would be expected to inhibit the growth of new vegetation. As stated previously, hexazinone is persistent and mobile; therefore, it is likely to be transported from soil surfaces during runoff events.

Based on a review of the hexazinone incidents for terrestrial plants, only three have been reported. In the first incident, a section of lawn grass was damaged following application of hexazinone to a swimming pool. In the remaining two incidents, both of which occurred on May 9, 2000, 130 acres of corn was damaged following aerial application of hexazinone and atrazine to corn, although both incidents were reported as “unlikely”. Although the reported number of hexazinone incidents for terrestrial plants is low, and due to uses either not relevant for this assessment (*i.e.* application to swimming pools) or cancelled (aerial application to corn), an absence of reports does not necessarily provide evidence of an absence of incidents. The only plant incidents that are reported are those that are alleged to occur on more than 45 percent of the acreage exposed to the pesticide. Therefore, an incident could impact 40% of an exposed crop and not be included in the EIIS database (unless it is reported by a non-registrant, such as a state agency, where data are not systemically collected.

In summary, terrestrial plant RQs are above LOCs; therefore, upland and riparian vegetation may be affected. Woody plants are generally sensitive to environmentally-relevant hexazinone concentrations; therefore, effects on shading, bank stabilization, structural diversity (height classes) of vegetation, and woodlands are expected. Given that both upland and riparian areas are comprised of a mixture of both sensitive woody (trees and shrubs) and sensitive grassy herbaceous vegetation, CRLFs may be indirectly affected by adverse effects to both woody and herbaceous vegetation which provides habitat and cover for the CRLF and its prey. Therefore, the effects determination for this assessment endpoint is "likely to adversely affect" or "LAA" for all assessed hexazinone use patterns.

As previously described in Section 3.2.5, downwind spray drift buffers were developed to determine the distance required to dissipate spray drift to below the LOC, based on both NOAEC and EC_{25} levels for terrestrial plants. Dissipation to the no effect level was modeled in order to provide potential buffer distances that are protective of endangered terrestrial plant species; this distance beyond the site of application is considered as the action area for hexazinone. However, because no obligate relationship exists between the CRLF and terrestrial plants, the portion of the action area that is relevant to the CRLF is defined by the dissipation distance to the EC_{25} level (*i.e.*, the potential buffer distance required to protect non-endangered terrestrial plant species). The spray drift distances presented in Table 3.4 were derived based on the most sensitive EC_{25} value for dicots in the seedling emergence test (0.0063 lb ai/A). Based on the maximum hexazinone aerial application rate of 12 lb ai/A a spray drift buffer of 3,366 feet from the site of application is required to dissipate to levels below the LOC (for the portion of the action area that is relevant to the CRLF). The vegetative vigor toxicity test is intended to assess the potential effects on plants following deposition of hexazinone on the leaves and above-ground portions of plants, which are more likely to receive exposure via spray drift. Therefore, spray drift distances are derived for the vegetative vigor endpoint, as well as the seedling emergence endpoint, for both monocots and dicots, in Table 5.15. As discussed in Section 3.2.5, the drift buffers for the more sensitive seedling emergence endpoint for dicots were derived using the AgDISP model with the Gaussian extension because the 1,000 foot limit of the AgDrift model was exceeded. However, spray drift dissipation distances reported for the vegetative vigor endpoints and for the monocot seedling emergence endpoint were based on the Agdrift model because the limits of the model were not exceeded using the spray drift parameters provided in Section 3.2.5. As shown in Table 5.15, adverse effects to terrestrial plants might reasonably be expected to occur up to 3589 feet from the use site for aerial applications and 3366 feet from the use site for ground applications of hexazinone. In some cases, topography (such as an intervening ridge) or weather conditions (such as prevailing winds towards or away from the frog habitat) could affect the estimates presented in Table 5.15. However, analysis of these site-specific details is beyond the scope of this assessment.

5.2.4 Modification to Designated Critical Habitat

5.2.4.1 Aquatic-Phase PCEs

Three of the four assessment endpoints for the aquatic-phase primary constituent elements (PCEs) of designated critical habitat for the CRLF are related to potential effects to aquatic and/or terrestrial plants:

- Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.

- Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.

- Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.,* algae).

The effects determinations for indirect effects to the CRLF via direct effects to aquatic and terrestrial plants are used to determine whether modification to critical habitat may occur.

Based on the results of the effects determinations for aquatic plants (see Sections 5.2.2.1 and 5.2.3.1), critical habitat of the CRLF may be modified via hexazinone-related impacts to non-vascular aquatic plants as food items for tadpoles and habitat for aquatic-phase CRLFs. Critical habitat may be modified by an increase in sediment deposition and associated turbidity (via impacts to herbaceous riparian vegetation), potential reduction in oxygen (via impacts to the aquatic plant community and primary productivity), and reduction in herbaceous riparian vegetation that provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult aquatic-phase CRLFs. Hexazinone uses may result in modification to critical habitat via direct effects to non-vascular plants for all modeled applications. Based on the results of the effects determination for terrestrial plants (see Section 5.2.3.2), hexazinone-related effects on shading (*i.e.*, temperature), bank stabilization, and structural diversity (height classes) of vegetation are expected because woody plants are generally sensitive to environmentally-relevant concentrations of hexazinone. However, modification to critical habitat may occur via hexazinone-related impacts to sensitive herbaceous vegetation, which provide habitat and cover for the CRLF and its prey, based on all assessed uses of hexazinone.

The remaining aquatic-phase PCE is "alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source." Other than impacts to algae as food items for tadpoles (discussed above), this PCE was assessed by considering direct and indirect effects to the aquatic-phase CRLF via acute and chronic freshwater fish and invertebrate toxicity endpoints as measures of effects. As discussed in Section 5.2.1.1, direct effects to the aquatic-phase CRLF, via mortality, growth, and/or fecundity are not expected. In addition, hexazinone-related effects to freshwater invertebrates and freshwater fish as food items are also not likely to occur (see Sections 5.2.2.2 and 5.2.2.3). Therefore, hexazinone is not likely to modify critical habitat by altering chemical characteristics necessary for normal growth and viability of aquatic-phase CRLFs and their non-plant food sources.

5.2.4.2 Terrestrial-Phase PCEs

Two of the four assessment endpoints for the terrestrial-phase PCEs of designated critical habitat for the CRLF are related to potential effects to terrestrial plants:

- Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or drip line surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance.

- Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal.

As discussed above, modification to critical habitat may occur via hexazinone-related impacts to sensitive herbaceous vegetation, which provides habitat, cover, and a means of dispersal for the terrestrial-phase CRLF and its prey, based on all monocot and dicot assessments for hexazinone uses. Modification to critical habitat is expected to occur in woodland areas because woody plants are sensitive to environmentally relevant concentrations of hexazinone. Terrestrial plant incident reports support the habitat modification detetmination.

The third terrestrial-phase PCE is "reduction and/or modification of food sources for terrestrial phase juveniles and adults." To assess the impact of hexazinone on this PCE, acute and chronic toxicity endpoints for terrestrial invertebrates, mammals, and terrestrial-phase frogs are used as measures of effects. Based on the characterization of indirect effects to terrestrial-phase CRLFs via reduction in the prey base (see Section 5.2.2.4 for terrestrial invertebrates, Section 5.2.2.5 for mammals, and 5.2.2.6 for frogs), critical habitat may be modified via a reduction in mammals and terrestrial-phase amphibians as food items. No open literature studies are available for terrestrial invertebrates, mammals or frogs resulting in more sensitive endpoints than registrant submitted studies. No incident reports are available for terrestrial invertebrates, mammals or frogs.

The fourth terrestrial-phase PCE is based on alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source. As discussed in Section 5.2.1.2, direct acute effects, via mortality, are expected for the terrestrial-phase CRLF. Chronic reproductive effects are possible for all uses of hexazinone except alfalfa and pasture. Therefore, hexazinone may adversely affect critical habitat by altering chemical characteristics necessary for normal growth and viability of terrestrial-phase CRLFs and their mammalian and amphibian food sources.

6 Uncertainties

6.1 Exposure Assessment Uncertainties

6.1.1 Maximum Use Scenario

The screening-level risk assessment focuses on characterizing potential ecological risks resulting from a maximum use scenario, which is determined from labeled statements of maximum application rate and number of applications with the shortest time interval between applications. The frequency at which actual uses approach this maximum use scenario may be dependant on pest resistance, timing of applications, cultural practices, and market forces.

6.1.2 Aquatic Exposure Modeling of Hexazinone

The standard ecological water body scenario (EXAMS pond) used to calculate potential aquatic exposure to pesticides is intended to represent conservative estimates, and to avoid underestimations of the actual exposure. The standard scenario consists of application to a 10-hectare field bordering a 1-hectare, 2-meter deep (20,000 m^3) pond with no outlet. Exposure estimates generated using the EXAMS pond are intended to represent a wide variety of vulnerable water bodies that occur at the top of watersheds including prairie pot holes, playa lakes, wetlands, vernal pools, man-made and natural ponds, and intermittent and lower order streams. As a group, there are factors that make these water bodies more or less vulnerable than the EXAMS pond. Static water bodies that have larger ratios of pesticide-treated drainage area to water body volume would be expected to have higher peak EECs than the EXAMS pond. These water bodies will be either smaller in size or have larger drainage areas. Smaller water bodies have limited storage capacity and thus may overflow and carry pesticide in the discharge, whereas the EXAMS pond has no discharge. As watershed size increases beyond 10-hectares, it becomes increasingly unlikely that the entire watershed is planted with a single crop that is all treated simultaneously with the pesticide. Headwater streams can also have peak concentrations higher than the EXAMS pond, but they likely persist for only short periods of time and are then carried and dissipated downstream.

The Agency acknowledges that there are some unique aquatic habitats that are not accurately captured by this modeling scenario and modeling results may, therefore, under- or over-estimate exposure, depending on a number of variables. For example, aquatic-phase CRLFs may inhabit water bodies of different size and depth and/or are located adjacent to larger or smaller drainage areas than the EXAMS pond. The Agency does not currently have sufficient information regarding the hydrology of these aquatic habitats to develop a specific alternate scenario for the CRLF. CRLFs prefer habitat with perennial (present year-round) or near-perennial water and do not frequently inhabit vernal (temporary) pools because conditions in these habitats are generally not suitable (Hayes and Jennings 1988). Therefore, the EXAMS pond is assumed to be representative of exposure to aquatic-phase CRLFs. In addition, the Services agree that the existing EXAMS pond represents the best currently available approach for estimating aquatic exposure to pesticides (USFWS/NMFS 2004).

In general, the linked PRZM/EXAMS model produces estimated aquatic concentrations that are expected to be exceeded once within a ten-year period. The Pesticide Root Zone Model is a process or "simulation" model that calculates what happens to a pesticide in an agricultural field on a day-to-day basis. It considers factors such as rainfall and plant transpiration of water, as well as how and when the pesticide is applied. It has two major components: hydrology and chemical transport. Water movement is simulated by the use of generalized soil parameters, including field capacity, wilting point, and saturation water content. The chemical transport component can simulate pesticide application on the soil or on the plant foliage. Dissolved, adsorbed, and vapor-phase concentrations in the soil are estimated by simultaneously considering the processes of pesticide uptake by plants, surface runoff, erosion, decay, volatilization, foliar wash-off, advection, dispersion, and retardation.

Uncertainties associated with each of these individual components add to the overall uncertainty of the modeled concentrations. Additionally, model inputs from the environmental fate degradation studies are chosen to represent the upper confidence bound on the mean values that are not expected to be exceeded in the environment approximately 90 percent of the time. Mobility input values are chosen to be representative of conditions in the environment. The natural variation in soils adds to the uncertainty of modeled values. Factors such as application date, crop emergence date, and canopy cover can also affect estimated concentrations, adding to the uncertainty of modeled values. Factors within the ambient environment such as soil temperatures, sunlight intensity, antecedent soil moisture, and surface water temperatures can cause actual aquatic concentrations to differ for the modeled values.

Unlike spray drift, tools are currently not available to evaluate the effectiveness of a vegetative setback on runoff and loadings. The effectiveness of vegetative setbacks is highly dependent on the condition of the vegetative strip. For example, a well-established, healthy vegetative setback can be a very effective means of reducing runoff and erosion from agricultural fields. Alternatively, a setback of poor vegetative quality or a setback that is channelized can be ineffective at reducing loadings. Until such time as a quantitative method to estimate the effect of vegetative setbacks on various conditions on pesticide loadings becomes available, the aquatic exposure predictions are likely to overestimate exposure where healthy vegetative setbacks exist and underestimate exposure where poorly developed, channelized, or bare setbacks exist.

] In order to account for uncertainties associated with modeling, available monitoring data were compared to PRZM/EXAMS estimates of peak EECs for the different uses. As discussed above, several data values were available from NAWQA for Hexazinone concentrations measured in surface waters receiving runoff from agricultural areas. The specific use patterns (e.g. application rates and timing, crops) associated with the agricultural areas are unknown, however, they are assumed to be representative of potential Hexazinone use areas.

6.1.3 Action Area Uncertainties

An example of an important simplifying assumption that may require future refinement is the assumption of uniform runoff characteristics throughout a landscape. It is well documented that runoff characteristics are highly non-uniform and anisotropic, and become increasingly so as the area under consideration becomes larger. The assumption made for estimating the aquatic action area (based on predicted in-stream dilution) was that the entire landscape exhibited runoff properties identical to those commonly found in agricultural lands in this region. However, considering the vastly different runoff characteristics of: a) undeveloped (especially forested) areas, which exhibit the least amount of surface runoff but the greatest amount of groundwater recharge; b) suburban/residential areas, which are dominated by the relationship between impermeable surfaces (roads, lots) and grassed/other areas (lawns) plus local drainage management; c) urban areas, that are dominated by managed storm drainage and impermeable surfaces; and d) agricultural areas dominated by Hortonian and focused runoff (especially with row crops), a refined assessment should incorporate these differences for modeled stream flow generation. As the zone around the immediate (application) target area expands, there will be greater variability in the landscape; in the context of a risk assessment, the runoff potential that is assumed for the expanding area will be a crucial variable (since dilution at the outflow point is determined by the size of the expanding area). Thus, it important to know at least some approximate estimate of types of land use within that region. Runoff from forested areas ranges from 45 – 2,700% less than from agricultural areas; in most studies, runoff was 2.5 to 7 times higher in agricultural areas (e.g., Okisaka et al., 1997; Karvonen et al., 1999; McDonald et al., 2002; Phuong and van Dam 2002). Differences in runoff potential between urban/suburban areas and agricultural areas are generally less than between agricultural and forested areas. In terms of likely runoff potential (other variables – such as topography and rainfall – being equal), the relationship is generally as follows (going from lowest to highest runoff potential):

Three-tiered forest < agroforestry < suburban < row-crop agriculture < urban.

There are, however, other uncertainties that should serve to counteract the effects of the aforementioned issue. For example, the dilution model considers that 100% of the agricultural area has the chemical applied, which is almost certainly a gross over-estimation. Thus, there will be assumed chemical contributions from agricultural areas that will actually be contributing only runoff water (dilutant); so some contributions to total contaminant load will really serve to lessen rather than increase aquatic concentrations. In light of these (and other) confounding factors, Agency believes that this model gives us the best available estimates under current circumstances.

6.1.4 Usage Uncertainties

County-level usage data were obtained from California's Department of Pesticide Regulation Pesticide Use Reporting (CDPR PUR) database. Four years of data (2002 – 2005) were included in this analysis because statistical methodology for identifying outliers, in terms of area treated and pounds applied, was provided by CDPR for these years only. No methodology for removing outliers was provided by CDPR for 2001 and earlier pesticide data; therefore, this information was not included in the analysis because it may misrepresent actual usage patterns. CDPR PUR documentation indicates that errors in the data may include the following: a misplaced decimal; incorrect measures, area treated, or units; and reports of diluted pesticide concentrations. In addition, it is possible that the data may contain reports for pesticide uses that have been cancelled. The CPDR PUR data does not include home owner applied pesticides; therefore, residential uses are not likely to be reported. As with all pesticide usage data, there may be instances of misuse and misreporting. The Agency made use of the most current, verifiable information; in cases where there were discrepancies, the most conservative information was used.

6.1.5 Terrestrial Exposure Modeling of Hexazinone

The Agency relies on the work of Fletcher et al. (1994) for setting the assumed pesticide residues in wildlife dietary items. These residue assumptions are believed to reflect a realistic upper-bound residue estimate, although the degree to which this assumption reflects a specific percentile estimate is difficult to quantify. It is important to note that the field measurement efforts used to develop the Fletcher estimates of exposure involve highly varied sampling techniques. It is entirely possible that much of these data reflects residues averaged over entire above ground plants in the case of grass and forage sampling.

It was assumed that ingestion of food items in the field occurs at rates commensurate with those in the laboratory. Although the screening assessment process adjusts dry-weight estimates of food intake to reflect the increased mass in fresh-weight wildlife food intake estimates, it does not allow for gross energy differences. Direct comparison of a laboratory dietary concentration- based effects threshold to a fresh-weight pesticide residue estimate would result in an underestimation of field exposure by food consumption by a factor of 1.25 – 2.5 for most food items.

Differences in assimilative efficiency between laboratory and wild diets suggest that current screening assessment methods do not account for a potentially important aspect of food requirements. Depending upon species and dietary matrix, bird assimilation of wild diet energy ranges from 23 – 80%, and mammal's assimilation ranges from 41 – 85% (U.S. Environmental Protection Agency, 1993). If it is assumed that laboratory chow is formulated to maximize assimilative efficiency (e.g., a value of 85%), a potential for underestimation of exposure may exist by assuming that consumption of food in the wild is comparable with consumption during laboratory testing. In the screening process, exposure may be underestimated because metabolic rates are not related to food consumption.

For the terrestrial exposure analysis of this risk assessment, a generic bird or mammal was assumed to occupy either the treated field or adjacent areas receiving a treatment rate on the field. Actual habitat requirements of any particular terrestrial species were not considered, and it was assumed that species occupy, exclusively and permanently, the modeled treatment area. Spray drift model predictions suggest that this assumption leads to an overestimation of exposure to species that do not occupy the treated field exclusively and permanently.

T-HERPS Modeling

Uncertainties in the Mammal and Herptile Prey Item EEC

T-HERPS calculates EECs for terrestrial-phase herptiles that consume mammals and other terrestrial phase herptiles. The amount of chemical estimated to be in the prey animal, in most cases, is thought to be a conservative estimate of potential dietary exposure because T-HERPS assumes that a small prey animal is consuming its daily intake of contaminated food before being consumed by the assessed species. Depuration of the pesticide from the prey item due to excretion or metabolism was not included in the estimation. Therefore, the EECs for chemicals that are short-lived in an animal are expected to represent an over-estimate of exposure. However, for chemicals that are bioaccumulative and are not readily degraded or excreted in an animal, the resulting exposure estimates could be low-end estimates because body burdens within the prey species would be expected to increase over time for bioaccumulative chemicals, resulting in potential body burdens that exceed the estimated daily dose calculated by T-HERPS. In addition, potential residues on the surface of potential prey items (e.g. in the fur) were not estimated by T-HERPS. Additional residues would be expected to be on prey item surface as well as within the prey item. Residues could be on prey items by several pathways including direct deposition of spray drift or by contact of the prey animal with contaminated soil or foliage.

In addition, the mammal prey item assessment assumes consumption of a 35-gram mammal by the assessed species. A body weight of 35 grams was chosen because it represents a higher end body weight of deer mice (U.S. EPA, 1993). Use of larger sized prey mammals would result in higher dose-based RQs, but lower dietary-based RQs. It is uncertain if dose-based or dietary-based RQs are more appropriate for this exposure pathway. Therefore, in cases where neither dietary-based nor dose-based RQs exceed

LOCs, effects of using a smaller mammal prey item (i.e., 15 grams) on the dietary based RQs should be considered by the assessor.

Uncertainties Associated with the Food Intake Allometric Equation

The daily food intake is estimated in T-HERPS using an iguanid lizard allometric equation as presented in U.S. EPA (1993). This equation is used in T-HERPS to estimate potential exposures to all herptiles, including the CRLF. Allometric equations specific for terrestrial-phase amphibians were not identified. To test the assumption that use of the iguanid lizard allometric equation results in a reasonable approximation of terrestrial phase amphibian food intake, measured food intake values reported for juvenile bullfrogs (*Rana catesbeiana*) of various weights reported by Modzelewski and Culley (1974, as cited in U.S. EPA, 1993) were compared to estimates derived using the iguanid food intake allometric equation incorporated into T-HERPS for the same body weight range.

The analysis suggests that food intake values for juvenile bullfrogs in the Modzelewski and Culley (1974) study are reasonably approximated using the allometric equation for iguanid lizards. The data in juvenile bullfrogs reported daily food intake values that range from approximately 3% to 7% of their body weight. Estimates of daily food intake using T-HERPS for the same range of body weights (13 grams to 100 grams) ranged from approximately 3% to 5% body weight daily. This analysis suggests that use of the iguanid lizard allometric equation results in a reasonable approximation of food intake reported for terrestrial phase frogs.

An additional uncertainty of T-HERPS is associated with temperature influence on the food intake allometric equation. Given that terrestrial phase frogs are poikilothermic, temperature may impact feeding rate. Temperature has not specifically been incorporated into the food ingestion allometric equation, and is not directly considered in T-HERPS.

Uncertainties associated with the Feeding Behavior of the Assessed Species

The allometric equation used to estimate daily food intake assumes a typical or constant food intake rate daily. In reality, the amount of food consumed (and, therefore, potential exposures to pesticides) may vary significantly from day to day, depending on a number of factors including availability of particular food items and energy needs.

T-HERPS estimates potential exposures for a number of food items. EECs for a particular food item are calculated with the assumption that one food item is consumed daily. Terrestrial-phase herptiles may receive 100% of their daily diet from one food item for a particular day, especially if larger prey, such as a small mammal, is available. However, many terrestrial-phase herptiles (including the California red-legged frog) may consume a variety of food items in a given day. T-HERPS estimates potential exposures resulting from consumption of a range of food items for the purpose of giving a high-end and low-end bounding estimate. All exposure values may be used in characterizing potential exposures.

6.1.6 Spray Drift Modeling

It is unlikely that the same organism would be exposed to the maximum amount of spray drift from every application made. In order for an organism to receive the maximum concentration of hexazinone from multiple applications, each application of hexazinone would have to occur under identical atmospheric conditions (e.g., same wind speed and same wind direction) and (if it is an animal) the animal being exposed would have to be located in the same location (which receives the maximum amount of spray drift) after each application. Additionally, other factors, including variations in topography, cover, and meteorological conditions over the transport distance are not accounted for by the AgDRIFT/AGDISP model (*i.e*., it models spray drift from aerial and ground applications in a flat area with little to no ground cover and a steady, constant wind speed and direction). Therefore, in most cases, the drift estimates from AgDRIFT/AGDISP may overestimate exposure, especially as the distance increases from the site of application, since the model does not account for potential obstructions (*e.g*., large hills, berms, buildings, trees, *etc*.). Furthermore, conservative assumptions are made regarding the droplet size distributions being modeled ('ASAE Very Fine' for agricultural uses), the application method (*i.e*., aerial), release heights and wind speeds. Alterations in any of these inputs would decrease the area of potential effect.

6.2 Effects Assessment Uncertainties

6.2.1 Age Class and Sensitivity of Effects Thresholds

It is generally recognized that test organism age may have a significant impact on the observed sensitivity to a toxicant. The acute toxicity data for fish are collected on juvenile fish between 0.1 and 5 grams. Aquatic invertebrate acute testing is performed on recommended immature age classes (e.g., first instar for daphnids, second instar for amphipods, stoneflies, mayflies, and third instar for midges).

Testing of juveniles may overestimate toxicity at older age classes for pesticide active ingredients that act directly without metabolic transformation because younger age classes may not have the enzymatic systems associated with detoxifying xenobiotics. In so far as the available toxicity data may provide ranges of sensitivity information with respect to age class, this assessment uses the most sensitive life-stage information as measures of effect for surrogate aquatic animals, and is therefore, considered as protective of the CRLF.

6.2.2 Use of Surrogate Species Effects Data

Guideline toxicity tests and open literature data on hexazinone are not available for frogs or any other aquatic-phase amphibian; therefore, freshwater fish are used as surrogate species for aquatic-phase amphibians. Therefore, endpoints based on freshwater fish ecotoxicity data are assumed to be protective of potential direct effects to aquatic-phase amphibians including the CRLF, and extrapolation of the risk conclusions from the most sensitive tested species to the aquatic-phase CRLF is likely to overestimate the potential risks to those species. Efforts are made to select the organisms most likely to be affected by the type of compound and usage pattern; however, there is an inherent uncertainty in extrapolating across phyla. In addition, the Agency's LOCs are intentionally set very low, and conservative estimates are made in the screening level risk assessment to account for these uncertainties.

6.2.3 Sub lethal Effects

When assessing acute risk, the screening risk assessment relies on the acute mortality endpoint as well as a suite of sub lethal responses to the pesticide, as determined by the testing of species response to chronic exposure conditions and subsequent chronic risk assessment. Consideration of additional sub lethal data in the assessment is exercised on a case-by-case basis and only after careful consideration of the nature of the sub lethal effect measured and the extent and quality of available data to support establishing a plausible relationship between the measure of effect (sub lethal endpoint) and the assessment endpoints.

To the extent to which sub lethal effects are not considered in this assessment, the potential direct and indirect effects of hexazinone on CRLF may be underestimated.

6.2.4 6.2.4 Location of Wildlife Species

For the terrestrial exposure analysis of this risk assessment, a generic bird or mammal was assumed to occupy either the treated field or adjacent areas receiving a treatment rate on the field. Actual habitat requirements of any particular terrestrial species were not considered, and it was assumed that species occupy, exclusively and permanently, the modeled treatment area. Spray drift model predictions suggest that this assumption leads to an overestimation of exposure to species that do not occupy the treated field exclusively and permanently.

7 Risk Conclusions

In fulfilling its obligations under Section 7(a)(2) of the Endangered Species Act, the information presented in this endangered species risk assessment represents the best data currently available to assess the potential risks of hexazinone to the CRLF and its designated critical habitat.

Based on the best available information, the Agency makes a Likely to Adversely Affect determination for the CRLF from the use of hexazinone. Additionally, the Agency has determined that there is the potential for modification of CRLF designated critical habitat from the use of the chemical.

In order to confirm that uses of hexazinone have the potential to affect the CRLF and its critical habitat through direct applications to spray drift to non-target areas, it is necessary to determine whether the final action area for hexazinone uses overlap with CRLF habitats. Using ArcGIS 9.2, the National Land-Cover Dataset (NLCD, 2001), and the

CRLF habitat information provided by the USFWS, the Agency has identified the areas where indirect effects to the CRLF and modification to designated critical habitat are anticipated to occur. These areas are depicted with a 4,200 foot buffer for cultivated crops, forest and pasture (Fig. 2.4) and non-crop/non-agricultural ROW (2.5).

Modification to CRLF designated critical habitat could potentially occur in 89% (approximately 20,846 out of 23,440 acres) of the currently designated habitat area. Based on the results of this effects determination, the CRLF may be indirectly affected within 930 core areas within the eight recovery units.

A summary of the risk conclusions and effects determinations for the CRLF and its critical habitat, given the uncertainties discussed in Section 6, is presented in Tables 5.12 and 5.13.

In fulfilling its obligations under Section 7(a)(2) of the Endangered Species Act, the information presented in this endangered species risk assessment represents the best data currently available to assess the potential risks of hexazinone to the CRLF and its designated critical habitat.

Based on the best available information, the Agency makes a Likely to Adversely Affect determination for the CRLF from the use of hexazinone. Additionally, the Agency has determined that there is the potential for modification of CRLF designated critical habitat from the use of the chemical. No direct effect for the aquatic-phase CRLF is expected using fish as a surrogate due to no LOC exceedence.

No indirect dietary effect is expected for the aquatic-phase CRLF consuming aquatic invertebrates for hexazinone based on no LOC exceedence. A "May Affect" determination for indirect dietar effects for the CRLF consuming non-vascular aquatic plants effect is due to the LOC exceedence for all uses except blueberry. An "LAA" determination for non-vascular aquatic plants resulted for all uses of hexazinone except blueberry due to mode-of-action for hexazinone. A "May Affect" determination is due to LOC exceedence for non-agricultural uncultivated areas (10 lb/A), conifer release (4 lb/A) and Christmas tree (2 lb/A) uses for vascular plants and the "LAA" determination is due to the mode-of-action for hexazinone.

A determination of "May Affect" for alteration of critical habitat for monocots and dicots was based on LOC exceedence for all hexazinone uses. The "LAA" determination is due to the mode-of-action for hexazinone, which interferes with photosynthesis and RNA production.

A determination of habitat modification for critical habitat was based on LOC exceedence resulting from the TerrPlant analysis for all hexazinone uses. The "LAA" determination is due to the mode-of-action for hexazinone, which interferes with photosynthesis and RNA production.

A summary of the risk conclusions and effects determinations for the CRLF and its critical habitat, given the uncertainties discussed in Section 6, is presented in Tables 7.1 and 7.2.

Table 7.1 Effects Determination Summary for Direct and Indirect Effects of Hexazinone on the CRLF		
Assessment Endpoint	**Effects Determination**[1]	**Basis for Determination**
Aquatic-Phase CRLF (Eggs, Larvae, and Adults)		
Direct Effect s of Hexazinone on the Aquatic-Phase CRLF		
Survival, growth, and reproduction of CRLF individuals via direct effects on aquatic phases	Using fish as a surrogate: No Effect	Using freshwater fish as a surrogate, no acute and chronic LOCs are exceeded for applications of Non-agricultural Rights–of-way (12 lb/A-granular), there is no expectation for adverse effects for lower rates: noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas Tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
Indirect Effect s of Hexazinone on the Aquatic –Phase CRLF		
Survival, growth, and reproduction of CRLF individuals via effects to food supply (*i.e.,* freshwater invertebrates, non-vascular plants, fish, and frogs)	Freshwater invertebrates: No effect	Using freshwater invertebrates, no acute and chronic LOCs are exceeded for applications of Non-agricultural Rights–of-way (12 lb/A-granular). Due to no exceedence for the 12 lb/A rate, it is assumed that there would also be no exceedence for the lower rates: Noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
	Indirect Effects of Prey Reduction for Non-vascular aquatic plants for all uses: May Affect	The May Affect for hexazinone uses related to applications for non-agricultural ROW (12 lb/A)non-crop uses (8 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), rangeland (3 lbs/A), Christmas trees (2.0 lb ai/A), alfalfa (1.5 lb ai/A) and pasture (1.1lb ai/A), exceed LOCs; therefore, indirect effects to tadpoles that feed on algae are possible. RQs range from 22.37 to 1.52.

	LAA Non-vascular aquatic plants: No Effect	The LAA determination is due to mode of action Blueberry uses (RQ = 0.81) resulted in no LOC exceedence.
	Indirect Effects of Prey Reduction for Fish as surrogate for Frogs for all uses: No effect	Using freshwater fish as a surrogate, no acute and chronic LOCs are exceeded for applications of non-crop (12 lb/A-granular), Noncrop uses (8 lb/A), forest site preparation (5 lb/A), conifer release (3 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A), Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A). .
Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat, cover, and/or primary productivity (*i.e.,* aquatic plant community)	Non-vascular aquatic plants: May Affect LAA No Effect	Hexazinone uses related to applications on Non-crop (12 lb/A-granular), Noncrop uses (8 lb/A), forest site preparation (5 lb/A), pineapple (3.6 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas Tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) exceed LOCs. Indirect effects to tadpoles that feed on algae are possible due to the MOA, which interferes with photosynthesis and RNA. There is no LOC exceedence for blueberry (3 lb/A) (RQ=0.81).

	Indirect Effects on habita for Vascular aquatic plants for all uses: May Affect LAA	The "May Affect" is based on the LOC exceedence for vascular aquatic plants for liquid applications of hexazinone to non-crop (12 lb/A), non-crop uses (8 lbs/A), Forest Site Preparation (5 lb/A), Conifer Release (3 lb/A).RQs range from 4.19 to 1.25. The "LAA" determination is based on the Mode of Action.
	No Effect	No LOC exceedence resulted for blueberry (3 lb/A), rangeland (3 lb/A) Christmas tree (2 lb/A), alfalfa (1.5 lb/A) or pasture (1.1 lb/A).
Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation, required to maintain acceptable water quality and habitat in ponds and streams comprising the species' current range.	Fforested and grassy/herbaceous riparian vegetation: May Affect LAA < 3366 ft (ground) NLAA ≥ 3366 ft (ground) LAA <3589 ft (aerial); NLAA ≥ 3589 ft (aerial)	Riparian vegetation may be affected because terrestrial plant RQs exceed LOCs. RQs for semi-aquatic areas range from 87.66 to 937.50 Hexazinone effects on shading, bank stabilization, and structural diversity of riparian areas in the action area are expected. Aquatic-phase CRLFs may be indirectly affected by adverse effects to sensitive herbaceous vegetation (based on all hexazinone uses), which provides habitat and cover for the CRLF and attachment sites for its egg masses. The LAA determination is based on MOA for ground applications of hexazinone within a drift buffer of 3366 ft based on the AGDISP results. There is an "NLAA' determination fro animals outside the drift buffer of 3589 ft resulting from the AGDISP model. The LAA determination is based on MOA for aerial applications of hexazinone within a drift buffer of 3589 ft based on the AGDISP results. There is an "NLAA" determination for animals outside the drift buffer of 3589 ft resulting from the AGDISP model.
Terrestrial-Phase CRLF (Juveniles and adults)		
Direct Effect s of Hexazinone on the Terrestrial-Phase CRLF		

Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	Nongranular acute for medium size frog using the bird as a surrogate: May affect LAA for the medium size frog NLAA for the medium size frog No Effect for medium sized frog	An adverse effect is expected based on weight of evidence for acute avian toxicity. The T-Rex analysis resulted in an endangered species exceedence for all crops. Further refinement using the T-HERPS analysis resulted in an endangered species exceedence for the CRLF consuming small herbivore mammals with RQs ranging from 0.53 for (8 lb/A) resulting from T-Rex analysis. Further refinement using a T-HERPS analysis resulted in RQS falling between the endangered species (0.1) and acute risk (0.5) LOCs exceedence for the CRLF consuming small herbivore mammals for pineapple, blueberry, conifer release and Christmas tree uses for a 37 g animal. The RQs from the T-HERPS analysis were analyzed using probit analysis and resulted in discountable effects for prey reduction. A "No effect"determination for pasture (1.1 lb/A) is due to no LOC exceedence using the bird as a surrogate for the CRLF for the 37 g animal resulting from the T-REX analysis.
	Nongranular Acute Small Frog May Affect: NLAA	The May Affect resulted from LOC exceedence using the bird as a surrogate from the T-Rex analysis for all crops. An "NLAA" determination is due to no LOC exceedence for any use for the small CRLF (1.4 g) from the T-Herps analysis for consumption of small insect, large insect small, small herbivore mammal, insectivore mammals or small amphibian prey..
	Nongranular Acute Large Frog: May Affect: NLAA	The May Affect resulted from LOC exceedence using the bird as a surrogate in the T-Rex analysis for all crops. An "NLAA" determination is due to no LOC exceedence for any use for the juvenile CRLF (238 g) from the T-Herps analysis for consumption of small insect, large insect small, small herbivore mammal, insectivore mammals or small amphibian prey.

	Acute granular direct effects on the CRLF: No Effect	The no effect determination is due to no LOC exceedence using the bird as a surrogate from the LDft[2] results for noncrop non-agricultural ROW, forest site preparation (5 lb/A) and rangeland (3 lb/A).
	Nongranular chronic direct effects using the bird as a surrogate: May Affect:	The May affect is based on LOC exceedence using the bird as a surrogate from te T-REX analysis. T-Herps was used to refine the May Affect from T-REX to either an "LAA" or "NLAA" determination. Chronic reproductive effects are possible, based on non-granular uses of hexazinone.
	Direct chronic effects Using the bird as a surrogate: LAA for small herbivore mammal prey	T-Herps was used to refine the May Affect from T-Rex. The LAA determination resulted from LOC exceedence based on T-Herps for noncrop (8 lb/A) to Christmas tree (2 lb/A). RQs range from 4.22 to 1.05.
	Direct chronic effects using the bird as a surrogate: NLAA for small herbivore mammal prey	The NLAA determination for alfalfa (1.5 lb/A) and pasture (1.1 lb/A) resulted from no LOC exceedence. RQs range from 0.79-0.58.
	Direct chronic effects using the bird as a surrogate: LAA for CRLF consuming small insect prey	T-Herps was used to refine the May Affect from T-REX to an "LAA" determination. All uses except Christmas tree, alfalfa and pasture resulted in LOC exceedences for broadleaf. RQs range from 3.6 (8 lb/A) to 1.35 (3 lb/A).
	Direct chronic effects using the bird as a surrogate: NLAA for CRLF consuming small insect prey	Christmas tree (2 lb/), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) did not result in LOC exceedences for small small insect prey resulting from T-Herps modeling. RQs for small herbivore mammals range from 0.90 (1.5 lb/A) to 0.50 (1.1 lb/A).
	Direct chronic effects using the bird as a surrogate: NLAA consuming large insect prey	Non-crop (8 lb/A), pineapple (3.6 lb/A), blueberry (3 lb/A) Christmas tree (2 lb/A), alfalfa (1.1 lb/A) and pasture (1.1 lb/A) did not result in LOC exceedences for large insect prey resulting from T-Herps modeling. RQs for broadleaf range from 0.90 (2 lb/A) to 0.5 (1.1 lb/A) based on T-Herps analysis.

	Direct chronic effects uaing the bird as a surrogate: NLAA consuming small insectivore mammals Direct chronic effects uaing the bird as a surrogate: NLAA consuming small insectivore mammals	An NLAA determination resulted from no LOC exceedence for small insectivore mammal,. RQs for small insectivore mammals range from 0.26 (8 lb/A) to 0.04 (1.1 lb/A). RQs for small amphibians range from for 0.12 (8 lb/A) to 0.02 (1.1 lb/A) based on T-Herps analysis.
	Chronic direct effects using the bird as a surrogate (Granular): May Affect LAA	Granular formulations for noncrop(12 lb/A), forest site preparation (5 lb/A) and rangeland (3 lb/A) resulted in LOC exceedences resulting from the T-REX analysis. RQs range from 1.35 for rangeland to 5.4 for noncrop.
Indirect Effect s of Hexazinone on the Terrestrial –Phase CRLF		
Survival, growth, and reproduction of CRLF individuals via effects on prey (*i.e.,* terrestrial invertebrates, small terrestrial vertebrates, including mammals and terrestrial phase amphibians)	Nongranualr Acute Indirect prey reduction for Terrestrial Invertebrates: May Affect LAA	An LAA determination for small insect prey is based on the uncertainty regarding the effects of hexazinone on terrestrial invertebrates due to related toxicity values reporting no mortality at the highest concentration tested for all uses.

	Nongranular Acute Indirect prey reduction for terrestrial invertebrates (Large Insect Prey): May Affect LAA	 An LAA determination for large insect prey is based on the uncertainty regarding the effects of hexazinone on terrestrial invertebrates due to related toxicity values reporting no mortality at the highest concentration tested for noncrop (8 lb/A), pineapple (3.6 lb/A) and blueberry/conifer release (3 lb/A).
	Nongranular Acute No Effect	A "No effect" determination for large insect prey for Christmas tree (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A) uses was based on no LOC exceedence from the T-REX analysis.
	Indirect mammal prey reduction: Non-granular Acute May Affect LAA	 The May Affect is based on T-Rex results for 15 g mammal with a diet of shortgrass for non-crop (8 lb/A) through pasture (1.1 lb/A) uses. The LAA determination is due to noncrop (8 lb/A) and blueberry (3 lb/A) LOC exceedence from the T-REX analysis.
	Indirect mammal prey reduction Nongranular Acute: May Affect NLAA	The May Effect is based on RQs falling between the endangered species and acute risk LOC. Due to RQs falling between the endangered species and acute risk LOC further refinement used the the probit analysis. The potential reduction in abundance of terrestrial mammals as food for Christmas tree, alfalfa and pasture uses would be < 1%; therefore a "not likely to adversely affect" determination can be made.

	Granular Acute Indirect mammal prey reduction: No Effect (granular uses)	A No effect determination is based on the LDft[2] results from the T-REX analysis for non-crop(12 lb/A), forest site preparation (5 lb/A) and rangeland (3 lb/A).
	Indirect prey reduction for mammal Nongranular Chronic: May Affect: LAA	Chronic reproductive effects are possible, based on non-granular uses of hexazinone. The May affect is due to LOC exceedences for all nongranular uses. All uses resulted in LOC exceedences for short grass. RQs range from 9.60 (8 lb/A) to 1.32 (1.1 lb/A) based on T-Rex.
	Indirect prey reduction for amphibian using the bird as a surrogate Nongranular Acute May Affect	The May affect is due to LOC exceedence using the bird as a surrogate for all crops resulting from the T-Rex analysis.

	LAA	An "LAA" determination using the bird as a surrogate is based on LOC exceedence for noncrop (8 lb/A) uses. RQ for noncrop = 0.76 resulted from the T-REX analysis.
	Indirect prey reduction for amphibian using the bird as a surrogate NLAA	Conifer release, blueberry, Christmas tree, alfalfa and pasture uses resulted in RQs falling between the listed and non-listed LOCs from the T-REX analysis. Based on this assessment, the probit analysis was used as a further refinement for the determination. The potential reduction in abundance of amphibians as food for these uses would be < 1% at most; therefore a "not likely to adversely affect" determination can be made.
	Indirect prey reduction for amphibian using the bird as a surrogate Nongranular Chronic May Affect: LAA	Chronic reproductive effects are possible, based on non-granular uses of hexazinone resulting from the T-REX analysis using the bird as a surrogate. All nongranular uses except Christmas tree, alfalfa and pasture resulted in LOC exceedences for broadleaf. RQs for broadleaf range from 1.35 (3 lb/A) to 3.60 (8 lb/A).

	Amphibian using the bird as a surrogate Nongranular Chronic No Effect	Christmas tree, alfalfa and pasture did not result in LOC exceedences broadleaf. RQs for broadleaf range from 0.5 (3 lb/A) to 0.90 (1.5 lb/A) resulting from the T-Rex analysis. .
Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.,* riparian vegetation)	Woody and grassy/herbaceous riparian vegetation: May Affect	Riparian woody and herbaceous vegetation may be affected because terrestrial plant RQs are above LOCs.
	LAA < 184 ft (ground)	Due to MOA, which interferes with photosynthesis and RNA, terrestrial-phase CRLFs may be indirectly affected by adverse effects to sensitive woody and herbaceous vegetation which provide habitat and cover for the CRLF and its prey.
	NLAA ≥ 184 ft (ground)	There was an "NLAA" determination for animals outside the aerial drift buffer of 184 ft resulting from the AGDISP model..
	LAA < 850 ft (aerial);	Due to MOA, which interferes with photosynthesis and RNA, terrestrial-phase CRLFs may be indirectly affected by adverse effects to sensitive woody and herbaceous vegetation which provide habitat and cover for the CRLF and its prey.
	NLAA ≥ 850 ft (aerial)	There was an "NLAA" determination for animals outside the aerial drift buffer of 850 ft resulting from the AGDISP model.

Table 7.2 Effects Determination Summary for the Critical Habitat Impact Analysis

Assessment Endpoint	Effects Determination	Basis for Determination
Aquatic-Phase PCEs (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)		
Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the	Habitat modification	Due to the MOA which interferes with photosynthesis and RNA ,sensitive herbaceous

stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.		riparian vegetation may be affected based on all modeled uses of hexazinone; therefore, critical habitat may be modified by an increase in sediment deposition and reduction in herbaceous riparian vegetation that provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult aquatic-phase CRLFs.
Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.[8]	Habitat modification	Sensitive non-vascular aquatic plants may be affected; therefore, critical habitat may be modified via turbidity and reduction in oxygen content necessary for normal growth and viability of juvenile and adult aquatic-phase CRLFs.
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	No effect to growth and viability Habitat modification based on alteration of food source	Direct effects to the aquatic-phase CRLF, via mortality, growth, and/or fecundity, are not expected. However, critical habitat of the CRLF may be modified via hexazinone-related impacts to non-vascular aquatic plants as food items for tadpoles. LOCs are exceeded for non-vascular uses for non-agricultural ROW (12 lb/A), noncrop (8 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas Trees (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A).
	No Habitat Modification	There was no LOC exceedence for blueberry uses for nonvascular plants.
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.,* algae)	Habitat modification No Habitat Modification	Based on the results of the effects determinations for aquatic plants, critical habitat of the CRLF may be modified via hexazinone-related impacts to non-vascular aquatic plants as food items for tadpoles. LOCs are exceeded for modeled uses for (12 lb/A), noncrop uses (8 lb/A), conifer release (3 lb/A), rangeland (3 lb/A), Christmas trees (2 lb/A), alfalfa (1.5 lb/A) and pasture (1.1 lb/A). Blueberry use resulted in no LOC exceedence.
Terrestrial-Phase PCEs (Upland Habitat and Dispersal Habitat)		

[8] Physico-chemical water quality parameters such as salinity, pH, and hardness are not evaluated because these processes are not biologically mediated and, therefore, are not relevant to the endpoints included in this assessment.

Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance	Habitat modification	Based on MOA, modification to critical habitat may occur via hexazinone-related impacts to sensitive woody and herbaceous vegetation, which provide habitat and cover for the terrestrial-phase CRLF and its prey, based on all assessed uses of hexazinone**.** Terrestial incident reports support a "habitat modification" determination.
Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal	Habitat modification	Based on the MOA for hexazione modification to dispersal habitat may occur via hexazinone-related impacts to sensitive woody and herbaceous vegetation, which provide habitat and cover for the terrestrial-phase CRLF and its prey, based on all assessed uses of hexazinone**.** Terrestrial incident reports support the "habitat modification' determination.
Reduction and/or modification of food sources for terrestrial phase juveniles and adults	Habitat modification	Based on the characterization of indirect effects to terrestrial-phase CRLFs via reduction in the prey base, critical habitat may be modified via a reduction in mammals and terrestrial-phase amphibians as food items.
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	Habitat modification	Direct acute effects, via mortality, are expected for the terrestrial-phase CRLF. Chronic reproductive effects are also possible. Therefore, hexazinone may modify critical habitat by altering chemical characteristics necessary for normal growth and viability of terrestrial-phase CRLFs and their mammalian and amphibian food sources.

Table 5.14 Hexazione Use-Specific Direct Effect Determinations

	Aquatic phase frogs				***Terrestrial-phase frogs***		
	Aquatic Animals		***Aquatic Plants***		***Terrestrial Animals***		***Terrestrial Plants***
Use	***Acute***	***Chronic***	***Non Vasular***	***Vascular***	***Acute***	***Chronic***	
Noncrop	NE	NE	LAA	LAA	LAA	LAA	LAA
Conifer Release	NE	NE	LAA	NE	NLAA	LAA	LAA
Blueberry	NE	NE	NE	NE	NLAA	LAA	LAA
Christmas Tree	NE	NE	LAA	NE	NLAA	NE	LAA
Alfalfa	NE	NE	NE	NE	NLAA	NE	LAA
Pasture	NE	NE	NE	NE	NE	NE	LAA

Table 5.15 Hexazinone Use-Specific Indirect Effects Determinations[1] Based on Effects to Prey											
Use	***Algae***	***Aquatic Invertebrates***		***Terrestrial Invertebrates (Acute)***	***Aquatic phase frogs and fish***		***Terrestrial-phase frogs***		***Small Mammals***	
		Acute	***Chronic***		***Acute***	***Chronic***	***Acute***	***Chronic***	***Acute***	***Chronic***
Noncrop	LAA	NE	NE	LAA	NE	NE	LAA	NE	LAA	LAA
Pineapple	LAA	NE	NE	LAA	NE	NE	NLAA	LAA	LAA	LAA
Conifer Release	LAA	NE	NE	LAA	NE	NE	NLAA	LAA	LAA	LAA
Blueberry	NE	NE	NE	LAA	NE	NE	NLAA	LAA	LAA	LAA
Christmas Tree	LAA	NE	NE	LAA	NE	NE	NLAA	NE	NLAA	LAA
Alfalfa	LAA	NE	NE	LAA	NE	NE	NLAA	NE	NLAA	LAA
Pasture	LAA	NE	NE	LAA	NE	NE	NE	NE	NLAA	LAA

[1]LAA = likely to adversely affect; NLAA = not likely to adversely affect; NE = no effect

Based on the conclusions of this assessment, a formal consultation with the U. S. Fish and Wildlife Service under Section 7 of the Endangered Species Act should be initiated to seek concurrence with the LAA determinations and to determine whether there are reasonable and prudent alternatives and/or measures to reduce and/or eliminate potential incidental take.

When evaluating the significance of this risk assessment's direct/indirect and habitat modification effects determinations, it is important to note that pesticide exposures and predicted risks to the species and its resources (i.e., food and habitat) are not expected to be uniform across the action area. In fact, given the assumptions of drift and downstream transport (i.e., attenuation with distance), pesticide exposure and associated risks to the species and its resources are expected to decrease with increasing distance away from the treated field or site of application. Evaluation of the implication of this non-uniform distribution of risk to the species would require information and assessment techniques that are not currently available. Examples of such information and methodology required for this type of analysis would include the following:

- Enhanced information on the density and distribution of CRLF life stages within specific recovery units and/or designated critical habitat within the action area. This information would allow for quantitative extrapolation of the present risk assessment's predictions of individual effects to the proportion of the population extant within geographical areas where those effects are predicted. Furthermore, such population information would allow for a more comprehensive evaluation of the significance of potential resource impairment to individuals of the species.
- Quantitative information on prey base requirements for individual aquatic- and terrestrial-phase frogs. While existing information provides a preliminary picture of the types of food sources utilized by the frog, it does not establish minimal requirements to sustain healthy individuals at varying life stages. Such information could be used to establish

biologically relevant thresholds of effects on the prey base, and ultimately establish geographical limits to those effects. This information could be used together with the density data discussed above to characterize the likelihood of adverse effects to individuals.

- Information on population responses of prey base organisms to the pesticide. Currently, methodologies are limited to predicting exposures and likely levels of direct mortality, growth or reproductive impairment immediately following exposure to the pesticide. The degree to which repeated exposure events and the inherent demographic characteristics of the prey population play into the extent to which prey resources may recover is not predictable. An enhanced understanding of long-term prey responses to pesticide exposure would allow for a more refined determination of the magnitude and duration of resource impairment, and together with the information described above, a more complete prediction of effects to individual frogs and potential modification to critical habitat.

8. References

Altig, R. and R.W. McDiarmid. 1999. Body Plan: Development and Morphology. In R.W. McDiarmid and R. Altig (Eds.), Tadpoles: The Biology of Anuran Larvae.University of Chicago Press, Chicago. pp. 24-51.

Alvarez, J. 2000. Letter to the U.S. Fish and Wildlife Service providing comments on the Draft California Red-legged Frog Recovery Plan.

Crawshaw, G.J. 2000. Diseases and Pathology of Amphibians and Reptiles *in:* Ecotoxicology of Amphibians and Reptiles; ed: Sparling, D.W., G. Linder, and C.A. Bishop. SETAC Publication Series, Columbia, MO.

Fellers, G. M., et al. 2001. Overwintering tadpoles in the California red-legged frog (*Rana aurora draytonii*). Herpetological Review, 32(3): 156-157.

Fellers, G.M, L.L. McConnell, D. Pratt, S. Datta. 2004. Pesticides in Mountain Yellow-Legged Frogs (Rana Mucosa) from the Sierra Nevada Mountains of California, USA. Environmental Toxicology & Chemistry 23 (9):2170-2177.

Fellers, Gary M. 2005a. *Rana draytonii* Baird and Girard 1852. California Red-legged Frog. Pages 552-554. *In*: M. Lannoo (ed.) Amphibian Declines: The Conservation Status of United States Species, Vol. 2: Species Accounts. University of California Press, Berkeley, California. xxi+1094 pp. (http://www.werc.usgs.gov/pt-reyes/pdfs/Rana%20draytonii.PDF)

Fellers, Gary M. 2005b. California red-legged frog, *Rana draytonii* Baird and Girard. Pages 198-201. *In*: L.L.C. Jones, et al (eds.) Amphibians of the Pacific Northwest. xxi+227.

Fletcher, J.S., J.E. Nellessen, and T.G. Pfleeger. 1994. Literature review and evaluation of the EPA food-chain (Kenaga) nomogram, and instrument for estimating pesticide residues on plants. Environmental Toxicology and Chemistry 13 (9):1383-1391.

Hayes, M.P. and M.M. Miyamoto. 1984. Biochemical, behavioral and body size differences between *Rana aurora aurora* and *R. a. draytonii*. Copeia 1984(4): 1018-22.

Hayes and Tennant. 1985. Diet and feeding behavior of the California red-legged frog. The Southwestern Naturalist 30(4): 601-605.

Hoerger, F., and E.E. Kenaga. 1972. Pesticide residues on plants: Correlation of representative data as a basis for estimation of their magnitude in the environment. In F. Coulston and F. Korte, *eds*., Environmental Quality and Safety: Chemistry, Toxicology, and Technology, Georg Thieme Publ, Stuttgart, West Germany, pp. 9-28.

Jennings, M.R. and M.P. Hayes. 1985. Pre-1900 overharvest of California red-legged frogs (*Rana aurora draytonii*): The inducement for bullfrog (*Rana catesbeiana*) introduction. Herpetological Review 31(1): 94-103.

Jennings, M.R. and M.P. Hayes. 1994. Amphibian and reptile species of special concern in California. Report prepared for the California Department of Fish and Game, Inland Fisheries Division, Rancho Cordova, California. 255 pp.

Karvonen, T., Koivusalo, H., Jauhiainen, M., Palko, J. and Weppling, K. 1999. A hydrological model for predicting runoff from different land use areas, Journal of Hydrology, 217(3-4): 253-265.

Kupferberg, S. 1997. Facilitation of periphyton production by tadpole grazing: Functional differences between species. Freshwater Biology 37:427-439.

Kupferberg, S.J., J.C. Marks and M.E. Power. 1994. Effects of variation in natural algal and detrital diets on larval anuran (*Hyla regilla*) life-history traits. Copeia 1994:446-457.

LeNoir, J.S., L.L. McConnell, G.M. Fellers, T.M. Cahill, J.N. Seiber. 1999. Summertime Transport of Current-use pesticides from California's Central Valley to the Sierra Nevada Mountain Range, USA. Environmental Toxicology & Chemistry 18(12): 2715-2722.

McConnell, L.L., J.S. LeNoir, S. Datta, J.N. Seiber. 1998. Wet deposition of current-use pesticides in the Sierra Nevada mountain range, California, USA. Environmental Toxicology & Chemistry 17(10):1908-1916.

McDonald M.A.1; Healey J.R.; Stevens P.A. 2002. The effects of secondary forest clearance and subsequent land-use on erosion losses and soil properties in the Blue Mountains of Jamaica. Agriculture, Ecosystems & Environment, Volume 92, Number 1: 1-19.

Modzelewski, E. H., Jr.; Culley, D. D., Jr. (1974) Growth responses of the bullfrog, Rana catesbeiana fed various live foods. Herpetologica 30: 396-405.

Okisaka S.; Murakami A.; Mizukawa A.; Ito J.; Vakulenko S.A.; Molotkov I.A.; Corbett C.W.; Wahl M.; Porter D.E.; Edwards D.; Moise C. 1997. Nonpoint source runoff modeling: A comparison of a forested watershed and an urban watershed on the South Carolina coast. Journal of Experimental Marine Biology and Ecology, Volume 213, Number 1: 133-149.

Phuong V.T. and van Dam J. Linkages between forests and water: A review of research evidence in Vietnam. ***In***: Forests, Water and Livelihoods European Tropical Forest Research Network. ETFRN NEWS (3pp).

Rathburn, G.B. 1998. *Rana aurora draytonii* egg predation. Herpetological Review, 29(3): 165.

Reis, D.K. Habitat characteristics of California red-legged frogs (*Rana aurora draytonii*): Ecological differences between eggs, tadpoles, and adults in a coastal brackish and freshwater system. M.S. Thesis. San Jose State University. 58 pp.

Seale, D.B. and N. Beckvar. 1980. The comparative ability of anuran larvae (genera: *Hyla*, *Bufo* and *Rana*) to ingest suspended blue-green algae. Copeia 1980:495-503.

Sparling, D.W.G.M. Fellers, L.L. McConnell. 2001. Pesticides and amphibian population declines in California, USA. Environmental Toxicology & Chemistry 20(7): 1591-1595.

Teske, Milton E., and Thomas B. Curbishley. 2003. *AgDisp ver 8.07 Users Manual*. USDA Forest Service, Morgantown, WV.

U.S. EPA (1993). Wildlife Exposure Factors Handbook. EPA/600/R-93/187. Office of Research and Development. December, 1993.

U.S. Environmental Protection Agency (U.S. EPA). 1998. Guidance for Ecological Risk Assessment. Risk Assessment Forum. EPA/630/R-95/002F, April 1998.

U.S. EPA. 2004. Overview of the Ecological Risk Assessment Process in the Office of Pesticide Programs. Office of Prevention, Pesticides, and Toxic Substances. Office of Pesticide Programs. Washington, D.C. January 23, 2004.

U.S. EPA (2006). User's Guide. T-REX Version 1.3.1. Environmental Fate and Effects Division. Office of Pesticide Programs. U.S. Environmental Protection Agency. Washington, D.C. December 07, 2006.

U.S. Fish and Wildlife Service (USFWS). 1996. Endangered and threatened wildlife and plants: determination of threatened status for the California red-legged frog. Federal Register 61(101):25813-25833.

USFWS. 2002. Recovery Plan for the California Red-legged Frog (*Rana aurora draytonii*). Region 1, USFWS, Portland, Oregon. (http://ecos.fws.gov/doc/recovery_plans/2002/020528.pdf)

USFWS. 2006. Endangered and threatened wildlife and plants: determination of critical habitat for the California red-legged frog. 71 FR 19244-19346.

USFWS. Website accessed: 30 December 2006. http://www.fws.gov/endangered/features/rl_frog/rlfrog.html#where

U.S. Fish and Wildlife Service (USFWS) and National Marine Fisheries Service (NMFS). 1998. Endangered Species Consultation Handbook: Procedures for Conducting Consultation and Conference Activities Under Section 7 of the Endangered Species Act. Final Draft. March 1998.

USFWS/NMFS. 2004. 50 CFR Part 402. Joint Counterpart Endangered Species Act Section 7 Consultation Regulations; Final Rule. FR 47732-47762.

Wassersug, R. 1984. Why tadpoles love fast food. Natural History 4/84.