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

Pesticide Effects Determination

Environmental Fate and Effects Division
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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 disulfoton 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.

Disulfoton is a systemic organophosphate insecticide, acaracide (miticide) registered for use to control aphids, thrips, mealybugs, other sucking insects, and spider mites on a variety of commodities including asparagus, beans, broccoli, Brussels sprouts, cabbage, cauliflower, Christmas trees, cotton, and lettuce. It is also registered for a number of residential uses including flowers, shrubs, and ornamentals. All registered uses are considered as part of the federal action evaluated in this assessment.

It is formulated as an emulsifiable concentrate for most agricultural uses, but is also formulated as a granular product for residential uses, Christmas trees, and cotton. Applications are generally soil applied: injection, in-furrow spray, or row treatment followed by soil incorporation. It can also be applied as a foliar treatment by ground or air. Disulfoton is typically applied 1 time per season, but may be applied multiple times per year for some crops. Application rates typically range from about 1 to 2 lb ai/A, although application to Christmas trees is allowed at up to 4.5 lb a.i./A.

Disulfoton itself is moderately mobile and generally non-persistent but its major degradates are more stable, so total residues are likely to be persistent. Disulfoton degrades through microbially-mediated degradation in aerobic soil and aquatic environments but appears to be more stable in anaerobic environments. Disulfoton is also subject to rapid aquatic and soil photolysis, but it is essentially stable to hydrolysis. In aerobic soil, disulfoton can be oxidized by chemical reaction and microbial metabolism to its corresponding disulfoton sulfoxide and disulfoton sulfone, which are also toxic. The only other major degradate is sulfonic acid. The oxon forms of disulfoton and d. sulfoxide are formed as minor degradates through hydrolysis and soil photolysis. The major degradates are more persistent and more mobile than the parent, and the toxicity of these degradates is similar to or greater than toxicity of the parent for several of the taxonomic groups included in this assessment. Therefore, a total toxic residues approach was used in this assessment. In aerobic soil, the half-life for total disulfoton residues ranged from 120 to 408 days.

Since CRLFs exist within aquatic and terrestrial habitats, exposure of the CRLF, its prey and its habitats to disulfoton are assessed separately for the two habitats. Tier-II aquatic exposure models are used to estimate high-end exposures of disulfoton in aquatic habitats resulting from runoff and spray drift from different uses. Peak model-estimated environmental concentrations for total toxic residues in surface water resulting from different disulfoton uses range from 0.6 µg/L to 67 µg/L. California surface water monitoring data for disulfoton are available from U.S.Geological Survey's National Water Quality Assessment (NAWQA) program and the California Department of Pesticide Regulation (CDPR). Most sampling was for disulfoton only, though, so the data are of limited utility in supplementing modeling analysis of total toxic residues. No disulfoton above detection limits of 0.02 ug/L to 1 ug/L was reported for any samples from the NAWQA or CDPR databases, which included 1920 and 2712 samples, respectively, collected statewide over a period of at least 10 years through 2005. NAWQA included sampling for d. sulfone as well as disulfoton at 6 sites, and 3 of these sites were also sampled for d. sulfoxide. D. sulfone was detected at 2 sites at levels up to 0.084 ug/L and there were no detections of d. sulfoxide.

Disulfoton residues also have the potential to reach groundwater. No California monitoring data report detections of disulfoton, but several targeted studies in other areas demonstrate that transport of disulfoton to groundwater can occur in some conditions. Available groundwater monitoring data are primarily for parent only. Given the greater persistence and mobility of the major degradates of disulfoton, total residues are more likely to leach; therefore, monitoring for parent only will not likely capture the highest exposures. Although groundwater *per se* is not evaluated herein, it could be significant nonetheless because discharging groundwater may support low-order streams, wetlands, and intermittent ponds – environments that are favorable to California Red-Legged Frogs (CRLFs). Long-term chronic concentrations derived from the PRZM-EXAMS model could reflect background concentrations that might be found in discharged groundwater/stream baseflow.

The T-REX model was used to estimate disulfoton exposures to the terrestrial-phase CRLF and its potential prey resulting from uses involving foliar and granular applications. The AgDRIFT model was also used to estimate deposition of disulfoton on terrestrial and aquatic habitats from spray drift. Exposure to terrestrial plants was estimated using Terrplant. The T-HERPS model was used to allow for further characterization of dietary exposures of terrestrial-phase CRLFs relative to birds.

The effects determination 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 disulfoton 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 disulfoton 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.

Based on the best available information, the Agency makes a Likely to Adversely Affect determination for the CRLF from the use of disulfoton. Additionally, the Agency has determined that there is the potential for modification of CRLF designated critical habitat from the labeled use of the chemical. A summary of the risk conclusions and effects determinations for the CRLF and its critical habitat is presented in **Tables 1.1 and 1.2**. Further information on the results of the effects determination is included as part of the Risk Description in Section 5.2.

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

Assessment Endpoint	Effects Determination[1]	Basis for Determination
	Aquatic-Phase CRLF (Eggs, Larvae, and Adults)	
Direct Effects: Survival, growth, and reproduction of CRLF individuals via direct effects on aquatic phases	**LAA**	Endangered species LOC was exceeded for all uses; chronic LOC was exceeded for all uses except cotton, beans, and residential uses. Potential effect was not considered discountable or insignificant.
Indirect Effects: Survival, growth, and reproduction of CRLF individuals via effects to food supply (*i.e.*, freshwater invertebrates, non-vascular plants, fish, and frogs)	Freshwater invertebrates: **LAA**	Acute and chronic RQs were exceeded for all uses. Acute RQs ranged from approximately 0.5 to 17 and chronic RQs ranged from 145 to 5600. The potential magnitude of effect could be sufficient to result in indirect effects to the CRLF.
	Non-vascular aquatic plants: **NE**	No aquatic plant toxicity data have been submitted or were located in the open literature. Disulfoton is an insecticide, and EC25s for terrestrial plants were greater than the maximum application rate.
	Fish and frogs: **LAA** for some uses	Magnitude of potential impacts to fish and aquatic phase amphibians could be sufficient to indirectly affect the CRLF for some uses. The highest RQs occurred for the lettuce, cabbage, and asparagus uses.
Indirect Effects: 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: **NE**	No aquatic plant toxicity data have been submitted or were located in the open literature. However, disulfoton is an insecticide, and EC25s for terrestrial plants were greater than the maximum application rate.
	Vascular aquatic plants: **NE**	
Indirect Effects: 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.	**NE**	The EC25 is greater than the highest labeled application rate for all uses except Christmas trees. The Christmas tree RQ is <0.5.

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

Table 1.1b Effects Determination Summary for Direct and Indirect Effects of Disulfoton on the CRLF		
Assessment Endpoint	**Effects Determination[1]**	**Basis for Determination**
Terrestrial-Phase CRLF (Juveniles and adults)		
Direct Effects: Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	**LAA**	Acute LOC (0.5) was exceeded for all uses for disulfoton and its degradates. Potential for reproductive effects also exists for all uses.
Indirect Effects: Survival, growth, and reproduction of CRLF individuals via effects on prey (*i.e.*, terrestrial invertebrates, small terrestrial vertebrates, including mammals and terrestrial phase amphibians)	Terrestrial invertebrates: **LAA**	The endangered species LOC of 0.05 was exceeded for all uses. Also, disulfoton is an insecticide, and the potential magnitude of effect could be sufficient to result in indirect effects to the CRLF.
	Mammals: **LAA**	Acute (0.5) and chronic (1.0) LOCs were exceeded for all uses. The potential magnitude of effect could be sufficient to result in indirect effects to the CRLF.
	Frogs: **LAA**	Acute (0.5) and chronic (1.0) LOCs were exceeded for all uses. The potential magnitude of effect could be sufficient to result in indirect effects to the CRLF.
Indirect Effects: Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.*, riparian vegetation)	**NE**	The EC25 is greater than the highest labeled application rate for all uses except Christmas trees. The Christmas tree RQ would be <0.5.
[1] NE = no effect; NLAA = may affect, but not likely to adversely affect; LAA = likely to adversely affect		

Table 1.2 Effects Determination Summary for the Critical Habitat Impact Analysis		
Assessment Endpoint	**Effects Determination[1]**	**Basis for Determination**
Aquatic-Phase CRLF 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.	**NE**	Effects determination for potential effects related to impacts on aquatic and terrestrial plants was No Effect.
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.[2]	**NE**	Effects determination for potential effects related to impacts on aquatic and terrestrial plants was No Effect.
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	**HM**	Effects determination for direct and indirect effects to the CRLF was LAA.
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.*, algae)	**NE**	Effects determination for potential effects related to impacts on aquatic plants was No Effect.
Terrestrial-Phase CRLF 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	**NE**	Effects determination for potential effects related to impacts on aquatic and terrestrial plants was No Effect.
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	**NE**	
Reduction and/or modification of food sources for terrestrial phase juveniles and adults	**HM**	Effects determination for indirect effects via reducing available food supply was LAA.
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	**HM**	Effects determination for direct and indirect effects was LAA.

[1] NE = No effect; HM = Habitat Modification
[2] 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.

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.

When evaluating the significance of this risk assessment's direct/indirect and adverse 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 disulfoton on all registered agricultural commodities and residential areas. In addition, this assessment evaluates whether registered uses are 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, and AgDRIFT, all of which are described at length in the Overview Document. Additional refinements include use of methodology that refines potential exposures to terrestrial phase CRLFs using food ingestion levels more specific to amphibians as described in Section 5.2. 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 disulfoton is based on an action area. The action area is the area directly or indirectly affected by the federal action, as indicated by the exceedance 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 disulfoton 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 disulfoton 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 disulfoton 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 disulfoton.

If a determination is made that use of disulfoton 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 disulfoton use sites) and further evaluation of the potential impact of disulfoton 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 disulfoton is expected to directly impact living organisms within the action area (defined in Section 2.7), critical habitat analysis for disulfoton 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 disulfoton 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

This assessment includes an evaluation of potential ecological risks of labeled uses of disulfoton. Labeled uses include asparagus, beans, broccoli, Brussels sprouts, cabbage, cauliflower, Christmas trees, cotton, and lettuce. It is also registered for a number of residential uses. Disulfoton is formulated as an emulsifiable concentrate or a granular product. Application methods include soil treatment via spray, injection, or chemigation, and foliar treatment via ground spray or air.

The end result of the EPA pesticide registration process (*i.e.*, 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 disulfoton in accordance with the approved product labels for California is "the action" relevant to this ecological risk assessment.

Although current registrations of disulfoton allow for use nationwide, this ecological risk assessment and effects determination addresses currently registered uses of disulfoton in portions of the action area that are reasonably assumed to be biologically relevant to the CRLF and its designated critical habitat. Labeled disulfoton uses not considered relevant in this assessment are on coffee, labeled only for Puerto Rico, and Easter lilies, which are grown in California, but not in areas that overlap the CRLF recovery areas. In California, Easter lilies are grown only in Del Norte county, which is out of the scope of the CRLF assessment. Further discussion of the action area for the CRLF and its critical habitat is provided in Section 2.7.

Chemicals included in this assessment include the parent compound, disulfoton, and a sulfone and sulfoxide degradate. These degradates have been shown to be of similar toxicity or more toxic to terrestrial animals compared with disulfoton. The degradates are also a concern for aquatic organisms; therefore, this assessment considers potential risks from exposure to disulfoton and the two degradates of concern.

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).

Disulfoton has registered products that contain multiple active ingredients. Analysis of the available open literature and acute oral mammalian LD_{50} 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 disulfoton is appropriate.

2.3 Previous Assessments

Disulfoton has been used for many years; therefore, it has a long regulatory history, and numerous ecological risk assessments have been conducted. Recently (2002), an interim Reregistration Eligibility Decision (IRED) for disulfoton was issued, which became a final RED after completion of the OP cumulative assessment in 2006. The following measures were identified in the IRED as necessary to mitigate ecological risks. These measures have since been incorporated into the currently approved labels.

- A precautionary bee statement is added to all product labels for liquid formulations of disulfoton.

- Use is prohibited within a level, well maintained 25 foot vegetative buffer between treated fields and all permanent water bodies.

- No more than one application of disulfoton per calendar year for all crops, except for asparagus, barley, coffee, peanuts (North Carolina only), and potatoes, for which no more than two applications of disulfoton per calendar year are permitted.

- The maximum application rate for Christmas trees is reduced from 78 to 4.5 lbs ai/A nationally, the use is limited to fir species only, and disulfoton is soil incorporated, watered in, or applied to areas with permanent groundcover.

- Use on barley, wheat, potatoes, and commercially grown ornamentals (field or nursery stock) is phased out by June 2005.

- Several other uses were phased out and others were not eligible for re-registration.

This assessment incorporates all mitigations that were instituted in the RED, which have been included in all current labels of disulfoton.

2.4 Stressor Source and Distribution

2.4.1 Physical and Chemical Properties

Disulfoton is a water soluble organophosphate insecticide; some physical and chemical properties are listed below in **Table 2.1**. Structures of disulfoton and its major degradates, disulfoton sulfoxide and disulfoton sulfone, are presented in **Figures 2-1 to 2-3**. Note that the oxon forms of the compounds in Figures 2-1 to 2-3 would be represented by replacing the marked (*) sulfur with an oxygen.

Table 2.1. Physical and Chemical Properties of Disulfoton

Property	**Value**
Chemical Name (common)	Disulfoton
Chemical Name (CAS)	O,O'-diethyl-S-[2-(ethylthio)ethyl]phosphorothioate
CAS Number	298-04-4
Chemical Formula	$C_8H_{18}0_2PS_3$
Molecular Weight	274.39
Chemical Class	Organophosphate
Physical State	Colorless liquid
Specific Gravity (20°C)	1.144
Boiling point (at 0.01 mmHg)	62°C
Aqueous Solubility (25°C)	15 mg/L
Vapor Pressure (20°C)	1.8×10^{-4} mm Hg
Henry's Law Constant (*°C)	2.6×10^{-6} atm·m³/mol

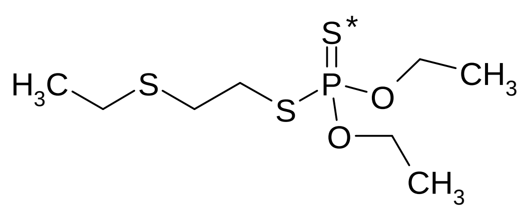


Figure 2-1. Chemical Structure of Disulfoton

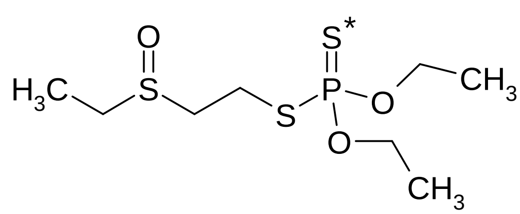


Figure 2-2. Chemical Structure of Disulfoton Sulfoxide
O,O-Diethyl S-[2-(ethylsulfinyl)ethyl] phosphorodithioate

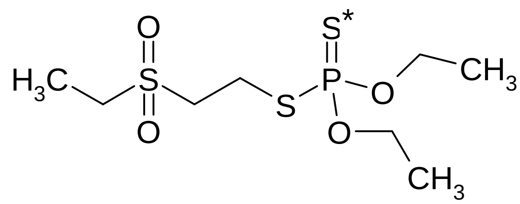


Figure 2-3. Chemical Structure of Disulfoton Sulfone
O,O-diethyl S-[2-(ethylsulfonyl)ethyl]-phosphorodithioate

2.4.2 Environmental Fate Properties

Disulfoton itself is moderately mobile and generally non-persistent, but its major degradates are more stable than the parent, so total residues are likely to be persistent. Disulfoton degrades rapidly through microbially-mediated degradation in aerobic soil and aquatic environments ($t_{1/2}$ = 2-17 d) but appears to be more stable in anaerobic environments ($t_{1/2}$ = 275 d). Disulfoton is also subject to aquatic and soil photolysis ($t_{1/2}$ = 3-4 d), but it is essentially stable to hydrolysis (pH 7, $t_{1/2}$ = 323 d). Based on physical properties, disulfoton has some potential to volatilize. A field volatility study shows dissipation to non-detectable levels at 5 ft within 6 hours of application.

Given the rapid transformation of disulfoton, exposure to its degradates is an important factor in assessing ecological risk. The primary degradates detected in environmental fate studies were disulfoton sulfoxide and disulfoton sulfone, both of which are of similar or greater toxicity than the parent compound. D. sulfoxide is formed at maximum levels of 15% to 95% through all microbial and abiotic processes excluding hydrolysis. D. sulfone is the major product of aerobic metabolism in soil and aquatic environments, reaching maximum levels of 19% to 72%. Sulfonic acid is the only other major degradate, formed at up to 16% through aerobic aquatic metabolism. Carbon dioxide and bound residues are other end products of metabolism.

Oxygen analogs (oxons) are potential degradates of OP pesticides that are often toxic and so are important to consider in assessing disulfoton's ecological risk. Transformation of disulfoton could lead to formation of three oxons: disulfoton oxon, d. sulfoxide oxon, and d. sulfone oxon. All were tested for in environmental fate studies and found only as minor degradates: d. sulfone oxon was not detected in any study, disulfoton oxon formed through hydrolysis at up to 3%, and d. sulfoxide oxon was formed through aquatic and soil photolysis at up to 0.3% and 4%, respectively. This dataset may not fully represent potential exposure to oxons, however. In all cases, maximum levels were reached at study termination, so it is possible that further increases could occur. Additionally, no study is available that considers photooxidation in air of volatilized disulfoton to its oxon forms, which could be an important route of transformation. Based on a field volatility study for disulfoton, if all of the volatilized disulfoton were oxidized, the maximum amount of disulfoton oxon that could be formed in air at 1 ft above the field would be 20.9 ng/L.

Both major degradates are more persistent and more mobile than the parent. In aerobic soil environments, half-lives for d. sulfoxide and d. sulfone are 63 to 77 days and 133 to 257 days, respectively. Total toxic residues (TTR) therefore dissipate more slowly and have a greater potential to leach. Terrestrial field studies indicate that disulfoton dissipates rapidly following application and does not leach below surface soil layers, but that d. sulfoxide and d. sulfone are more persistent and have greater leaching potential. D. sulfone was detected in the 0-6 in layer as late as 271 days following application, and was found at depths up to 18 inches.

Because of their toxicity and persistence, exposure to the degradates of disulfoton must be considered as well as exposure to the parent. In this assessment, aquatic exposure is estimated for the parent and its major toxic degradates as a group. “Total toxic residues” refers to the sum of three major forms of disulfoton (parent + sulfoxide + sulfone). Oxon degradates are not considered explicitly in this estimation. Inclusion of oxon data from available laboratory studies would not affect exposure estimates quantitatively because the TTR half-lives for the processes for which oxons are found as minor degradates are already represented as essentially stable. The potential for oxon exposure remains an uncertainty, however, because not all routes of transformation are accounted for in the guideline studies.

Tables 2.2 and 2.3 list the environmental fate properties of disulfoton and the degradates d. sulfoxide and d. sulfone and **Table 2.4** lists the major and minor degradates detected in the submitted environmental fate and transport studies. Results of the fate and transport studies on which these values are based are briefly described below. TTR half-lives reported in **Table 2.2** were estimated based on studies conducted on the parent compound. For each study, first order log linear TTR half-lives were calculated using, for each sampling point, the sum of the percent active ingredient recovered for all three species. Degradate half-lives reported in **Table 2.3** were estimated based on the pattern of decline observed in studies of the parent compound, rather than measured directly.

Table 2.2. Environmental fate and transport data for disulfoton			
Study	**Parent Value**	**Total Toxic Residues Value**	**Source**
Hydrolysis Half-life (at 20° C)	pH 4: 1174 d pH 7: 323 d pH 9: 231 d	Stable.	MRID 00143405
Aqueous Photolysis Half-life	3.9 d	141 d	MRID 40471102
Soil Photolysis Half -life	2.8 d	385 d	MRID 40471103
Aerobic Soil Metabolism Half-life	2.4 d 16.6 d 15.6 d	408 d 120 d 257 d	MRID 40042001 MRID 41585101 MRID 43900101
Anaerobic Soil Metabolism Half-life	No data	No data	n/a
Aerobic Aquatic Metabolism Half-life	10.7 d	51 d	MRID 46961201
Anaerobic Aquatic Metabolism Half-life	275 d	385 d	MRID 46316901
Foliar Dissipation Half-life	No data.	3.3 d	MRID 41201801
Soil Water Partition Coefficient (K_d)	sand: 4.67 L/kg_{soil} sandy loam: 9.66 L/kg_{soil} silt loam: 6.85 L/kg_{soil} clay loam: 4.47 L/kg_{soil}	See Table 2.3.	MRID 44373103
Organic Carbon Water Partition Coefficient (K_{oc})	sand: 888 L/kg_{oc} sandy loam: 483 L/kg_{oc} silt loam: 449 L/kg_{oc} clay loam: 386 L/kg_{oc}	See Table 2.3.	MRID 44373103
Field Dissipation DT_{50}	4 lb a.i./A x 1: 2 d, 3.7 d 4 lb a.i./A x 2: <3 d, <3 d No detections beneath 6” Sand/sandy loam soils	TTR persisted up to 271 days. Half-lives could not be calculated.	MRID 43042502
Laboratory Volatility Flux	25% field cap.: 0.026 $\mu g/cm^2/hr$ 75% field cap.: 0.096 $\mu g/cm^2/hr$	No data	MRID 42585802

a TTR half-lives were estimated based on studies conducted on the parent compound. For each study, first order log linear TTR half-lives were calculated using the sum of all three species detected at each sampling point.

Table 2.3. Environmental fate and transport data for d. sulfoxide and d. sulfone			
Study	**D. Sulfoxide**	**D. Sulfone**	**Source [a]**
Hydrolysis Half-life (at 20° C)	No data	No data	n/a
Aqueous Photolysis Half-life	As degradation product, reached 88% on day 10	No data	MRID 40471102
Soil Photolysis Half -life	As degradation product, appeared stable between days 15 and 30	No data	MRID 40471103
Aerobic Soil Metabolism Half-life	77 d 68 d 63 d	-- 133 d 257 d	MRID 40042001 MRID 41585101 MRID 43900101
Anaerobic Soil Metabolism Half-life	No data	No data	n/a
Aerobic Aquatic Metabolism Half-life	46 d	76 d	MRID 46961201
Anaerobic Aquatic Metabolism Half-life	39 d [b] --	-- 120 d [b]	MRID 46766603 MRID 46766604
Foliar Dissipation Half-life	No data	No data	n/a
Soil Water Partition Coefficient (K_d)	sandy loam: 0.6 L/kg_{soil} sandy loam: 1.7 L/kg_{soil} silt loam: 0.3 L/kg_{soil} loam: 3.5 L/kg_{soil}	sandy loam: 1.36 L/kg_{soil} sandy loam: 2.49 L/kg_{soil} silt loam: 0.43 L/kg_{soil} loam: 5.90 L/kg_{soil}	MRID 46766601 MRID 46766602
Organic Carbon Water Partition Coefficient (K_{oc})	sand: 63 L/kg_{oc} sandy loam: 94 L/kg_{oc} silt loam: 61 L/kg_{oc} clay loam: 62 L/kg_{oc}	sand: 136 L/kg_{oc} sandy loam: 138 L/kg_{oc} silt loam: 87 L/kg_{oc} clay loam: 104 L/kg_{oc}	MRID 46766601 MRID 46766602
Field Dissipation (DT_{50}s could not be calc'd)	In 3 of 4 sites, was detected up to 90 days, the final sampling period.	Persisted up to 178 to 364 days. Detected at 6-12".	MRID 43042502

a Unless otherwise noted, degradation rates were estimated based on the pattern of decline observed in studies of the parent compound, rather than measured directly.
b Measured directly.

Table 2.4. Maximum degradate amounts in environmental fate studies of disulfoton						
Degradate/ Metabolite	**Max. Degradate % of Applied (Time of peak)[1]**					
	Hydrolysis (pH 4,7,9)	**Aquatic Photolysis**	**Soil Photolysis**	**Anaerobic Aquatic Metabolism**	**Aerobic Aquatic Metabolism[2]**	**Aerobic Soil Metabolism** (n=3)
Disulfoton Oxon	1-3% (30 d*)					
Disulfoton Sulfoxide	4-6% (7 d)	89% (9.6 d*)	95% (30 d*)	15% (364 d*)	63% (3 d)	37-62% (3-7d)
Disulfoton Oxon Sulfoxide		0.7% (9.6*)	4% (30 d*)			
Disulfoton Sulfone			1% (20 d)	2% (270 d)	19% (28)	53-72% (14-90 d)
Sulfonic Acid				2% (364 d*)	16% (59 d)	
CO_2*				0.6%	29%	0-39%
Bound Residues*				34%	32%	24-39%

* Maximum reached at study termination.
[1] Unless otherwise noted, unidentified degradates were <3.7% and volatile organic carbon compounds were not detected.
[2] Unidentified degradates reached 11.1% at study termination (168 d).

Abiotic Degradation
Disulfoton is essentially stable to hydrolysis at 20°C, with reported half-lives of 1,174 days, 323 days, and 231 days in sterile aqueous buffered solutions at pH 4, 7, and 9, respectively. At 40°C, the half-lives were 30, 23.2, and 22.7 days at pH 4, 7, and 9, respectively. There were no major degradates. Minor degradates included disulfoton oxon and d. sulfoxide.

In both aqueous and soil environments, disulfoton is photolyzed primarily to d. sulfoxide with half-lives of 3.9 d in water and 2.8 d on soil (both dark corrected). By study termination (aqueous, 9.6 d; soil, 30 d), d. sulfoxide had reached 89% and 95% of the applied respectively. Disulfoton oxon sulfoxide was also formed at low levels (aqueous, 0.7%; soil, 4%). Total toxic residues of disulfoton are therefore essentially stable.

Microbial metabolism in soil
In three aerobic soil metabolism studies in sandy loam soils, disulfoton degraded rapidly, with DT_{50}s of <3 days in all cases and calculated first-order half-lives of 2.4 to 16.6 days. In the one study with a sterile control, degradation occurred with a half-life of less than one month, so the degradation may be due in part to abiotic processes rather than microbial metabolism.

The primary transformation products are d. sulfoxide and d. sulfone, so total toxic residues are much more persistent than the parent alone, with 20% to 58% of the applied compound remaining as total toxic residues by day 270 and TTR half-lives of 120 to 408 days. In all three studies, the parent compound readily oxidizes to d. sulfoxide which then undergoes further oxidation to the more stable d. sulfone, leading to carbon dioxide and bound residues as the end products. At study termination, d.sulfone is the primary compound remaining (17% to 51%), with parent at <1% and d. sulfoxide at 2% to 6%.

In the two studies for which the pattern of decline for d. sulfoxide and d. sulfone could be determined, estimated first-order half-lives were 63 to 68 d and 133 to 257 d, respectively. In two of the three studies, transformation of d.sulfone slowed at the end, remaining nearly stable in the final sampling periods, so the calculated half-lives may underestimate its persistence.

No acceptable studies of disulfoton metabolism in anaerobic soil conditions have been submitted.

Microbial metabolism in water-sediment systems
Acceptable aquatic metabolism studies are available for disulfoton in aerobic and anaerobic conditions and for the degradates d. sulfoxide and d. sulfone in anaerobic conditions.

In aerobic conditions, disulfoton was observed to degrade rapidly with a total system first-order half-life of 10.9 d. It was undetectable in water by day 14 and in sediment by day 90. Toxic residues d. sulfoxide and d. sulfone were formed as major degradates, reaching total system maximum levels of 63% (day 3) and 19% (day 28), respectively, and declining with half-lives of 46 d and 76 d. Another major degradate was sulfonic acid, formed at up to 15.8% (day 59). At study termination (168 d), each major degradate was at 5-7%, and the remaining residues were present as bound residues (32%) and carbon dioxide (29%). The total toxic residues half-life was 51 d.

In anaerobic conditions, disulfoton is much more persistent than in aerobic conditions. With a total system half-life of 227 days, 35.7% remained as parent at day 364. D. sulfoxide was the only major degradate and reached a maximum of 14.6% in the final sample. The total toxic residues half-life was 385 d.

A study of d. sulfoxide applied directly in an anaerobic system also demonstrates that disulfoton is more stable. Over the first 30 days, d. sulfoxide transformed to disulfoton with a half-life of 12.6 days. Through the final 3 months of the study (to 120 days), the system remained relatively stable with disulfoton at 75-77%, d. sulfoxide at 10-12%, and bound residues at 4-6%. During this period, 83-87% of the residues were found in the sediment. No d. sulfone was detected throughout the study.

D. sulfone applied directly to an anaerobic water-sediment system partitioned to sediment at a steady rate without any other degradates formed. By the end of the study (120 days), 20% of the applied sulfone was in the water, 39.7% was extractable as parent in the sediment, and the remaining was present as bound residue.

Field and Foliar Dissipation
Disulfoton dissipation has been measured in terrestrial field environments and on foliage. In the field, disulfoton was applied at sites in Fresno, CA and Watsonville, CA as one and

two applications of 4 lbs a.i./A each. In all 4 tests, disulfoton DT_{50}s of 2 to 4 days were observed in the upper 6 inches of soil and there were no detections below 6 inches. D. sulfoxide and d. sulfone were detected as well and both were more persistent than the parent and showed greater tendency to leach. Oxons were not analyzed for. D. sulfoxide had peak levels within the first week of sampling and in all but one of the sites was still detected at low levels at 90 days, the final sample date. Two sites had single detections of d. sulfoxide at low levels in the 6 to 12 inch layer. D. sulfone was more persistent and showed more tendency to leach. It was detected at low levels for most of the study period. In the four sites, peak concentrations were recovered in the first two weeks and the first samples with no detections were found between 178 and 364 days. Both Fresno plots had detections of d. sulfone at depths of up to 18 inches. In one plot, d. sulfone was detected in the 6 to 12 inch layer on sampling periods between 14 and 271 days in the other, there were detections between 28 days and 180 days. In both plots, the final detection was also the highest concentration. In the 12 to 18 inch layer, several detections at low levels were found between 90 and 180 days. Application rates were not confirmed and recoveries at some sampling points exceeded initial recoveries, so concentrations cannot be reported as percent applied.

A foliar dissipation rate of 3.3 days was calculated based on field monitoring data from a study in Michigan in which disulfoton was aerially applied to potatoes 3 times at 1 lb ai/A. Foliar dissipation rate estimates are based on potato foliage samples which were collected up to 14 days after the third treatment.

2.4.3 Environmental Transport Mechanisms

Potential transport mechanisms include pesticide surface water runoff, spray drift, and secondary drift of volatilized or soil-bound residues leading to deposition onto nearby or more distant ecosystems. Surface water runoff and spray drift are expected to be the major routes of exposure for disulfoton. The toxic degradates (d. sulfoxide and d. sulfone) have the potential to move vertically down through the soil profile, and potentially into groundwater, as they form primarily in the shallow subsurface. Groundwater that contains disulfoton residues may then be discharged into surface waters as baseflow. Soil adsorption and volatility data are discussed below.

Mobility
Disulfoton is moderately mobile in soil. The major degradates d. sulfoxide and d. sulfone are more mobile than the parent. Considering the greater persistence of the degradates as well, their higher mobility indicates that they are more likely to runoff and/or leach. Based on batch equilibrium studies in four soils, the mean K_F for disulfoton was 6.4 mL/g, while for d. sulfoxide and d. sulfone, mean K_F values were 1.5 mL/g and 2.5 mL/g, respectively. The K_{oc} model appears to be appropriate to describe disulfoton adsorption. Normalized for organic carbon content, K_{oc} values for the parent and the sulfoxide and sulfone degradates were 551, 70, and 161 mL/g_{oc}, respectively.

Volatility
Disulfoton has been classified as slightly volatile (Burkhard and Guth, 1981, and EPA, 1975). The vapor pressure (1.8×10^{-4} mm Hg) and Henry's Law Constant (2.6×10^{-6}

atm-m^3/mole) values for disulfoton indicate some degree of volatility from both soil and water. Measured vapor pressure values and Henry's law constants are not available for the major degradates, but estimates using EPI-SUITE suggest that the degradates are less likely to volatilize than the parent (D. sulfoxide: v.p. = 3.65 x 10^{-5}, K_H = 1.32 x 10^{-10} atm-m^3/mole; D. sulfone: v.p. = 1.12 x 10^{-5} mm Hg, K_H = 1.67 x 10^{-8} atm-m^3/mole). Given the rapid degradation of the parent compound, lower volatility of the degradates would lead to lower potential for volatilization of total toxic residues.

Disulfoton was not observed to volatilize in any of the aerobic soil metabolism studies, but in a laboratory volatilization study, maximum volatilization of 0.026 and 0.096 µg/cm^2/hr was seen in the first 24 hours from sand soil adjusted to 25% and 75% of field capacity. The study was conducted at 25°C with an air flow of approximately 300 ml/minute (MRID 42585802). Volatility was measured in the field as well (MRID 40471105). The maximum concentration observed in air at 1 foot above ground was 22.2 ng/L. After 6 hours, disulfoton concentrations at the 5 foot level were not detectable.

Photooxidation of OP pesticides in air can be an important route of oxon formation but no studies have been conducted to test this in disulfoton. Based on the field volatility study, the maximum amount of oxon that could be formed at the 1 ft level would be 20.9 ng/L, assuming that all volatilized disulfoton were oxidized.

Long Range Atmospheric Transport
Based on the measured physical properties of disulfoton and its degradates, as well as on field volatility studies, long range atmospheric transport is not expected to be an important exposure pathway. Available air monitoring studies in the Central Valley and Sierra Nevada do not include disulfoton as an analyte (http://www.cdpr.ca.gov/docs/empm/pubs/tac/tacstdys.htm; http://www.nature.nps.gov/air/Studies/air_toxics/wacap.cfm; Majewski, 1995). One national study was found that tested for disulfoton at 10 sites, finding it in only 1 out of 123 samples at a concentration of 0.0047 ng/L (Carey and Kutz, 1985).

2.4.4 Mechanism of Action

Organophosphate toxicity is based on the inhibition of the enzyme acetylcholinesterase which cleaves the neurotransmitter acetylcholine. Inhibition of acetylcholinesterase by organophosphate insecticides, such as disulfoton, interferes with proper neurotransmission in cholinergic synapses and neuromuscular junctions (USEPA 2002).

2.4.5 Use Characterization

Analysis of labeled use information is the critical first step in evaluating the federal action. The current labels for disulfoton represent 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.

Currently approved agricultural uses of disulfoton include asparagus, beans, broccoli, Brussels sprouts, cabbage, cauliflower, Christmas trees, cotton, and lettuce. Residential uses are allowed as well, including uses on vegetable gardens and flower beds. All agricultural uses require a 25 ft vegetated buffer strip around water bodies. Disulfoton is also registered for some residential uses. **Table 2.5** presents the uses and corresponding application rates and methods of application considered in this assessment.

Table 2.5 Disulfoton Uses Assessed for the CRLF

Use	Application Method	Form	Max. Annual App Rate[a] (lb a.i./A)
Asparagus	Ground spray Aerial spray	EC	2 (1 lb a.i./A/app, 2 apps/yr)
Beans	Soil injection (side of furrow)	EC	1
Broccoli	Soil injection [b]	EC	1
Brussels sprouts	Soil injection [b]	EC	1
Cabbage	Soil injection [b] Ground spray (broadcast) [c]	EC	1 (spray) 2 (soil injection)
Cauliflower	Soil injection [b]	EC	1
Christmas trees	Broadcast; wetted in	G	4.5
Cotton	Ground spray (in furrow) Soil injection (side of furrow)	EC	1
	Drill planting (in furrow)	G	0.975
	Hill-drop planting (in furrow)	G	0.375
Lettuce	Chemigation (drip or trickle) Soil injection [c]	EC	2
Residential Uses (Vegetables, Flowers)	Hand application, broadcast or per plant	G	1.6 [e]

[a] Unless otherwise specified, labels allow only a single application per year.
[b] Pre-seeding in transplant beds; 2-3 inch incorporation.
[c] Side of furrow (at planting) or side-dress (post-emergent).
[d] Not specified.
[e] The highest labeled application rate is 0.2 lb a.i./1000 ft2. Assuming a flower bed/garden size per lot of 200 ft x 10 ft and 4 lots per acre, this is equivalent to 1.6 lb a.i./A.

The Agency's Biological and Economic Analysis Division (BEAD) provides an analysis of both national- and county-level usage information (Kaul and Jones, 2006) using state-level usage data obtained from USDA-NASS[1], Doane (www.doane.com; the full dataset is not provided due to its proprietary nature) and the California's Department of Pesticide

[1] United States Depart 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.

Regulation Pesticide Use Reporting (CDPR PUR) database[2]. CDPR PUR is considered a more comprehensive source of usage data than USDA-NASS or proprietary databases, and thus the usage data reported for disulfoton by county in this California-specific assessment were generated using CDPR PUR data. Four years (2002-2005) of usage data were included in this analysis. Data from CDPR PUR were obtained for every pesticide application made by professional applicators on every use site at the section level (approximately one square mile) of the public land survey system. BEAD summarized these data to the county level by site, pesticide, and unit treated. Calculating county-level usage involved summarizing across all applications made within a section and then across all sections within a county for each use site and for each pesticide. The county level usage data that were calculated include: average annual pounds applied, average annual area treated, and average and maximum application rate across all five years. The units of area treated are also provided where available. According to the CDPR PUR database, a total of 54,554 lbs of disulfoton were used in California in 2002, dropping to 31,512 lbs in 2005. **Figures 2.4 and 2.5** below show the reported average annual number of pounds used in each county and for each crop between 2002 and 2005. These data indicate that the predominant use of disulfoton was on asparagus in San Joaquin county, representing a quarter of total average annual use over this time period. Another quarter of total average annual use was in Monterey county, primarily on broccoli, lettuce, and asparagus. This analysis is not entirely representative of current use patterns because residential uses are not included in these data, and because labeled uses have changed since these data were collected. Uses on peppers and wheat, which make up substantial portions of total use in this analysis, have been cancelled and phased out over the last few years. Data from the CDPR PUR database are presented in Appendix B.

[2] 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.

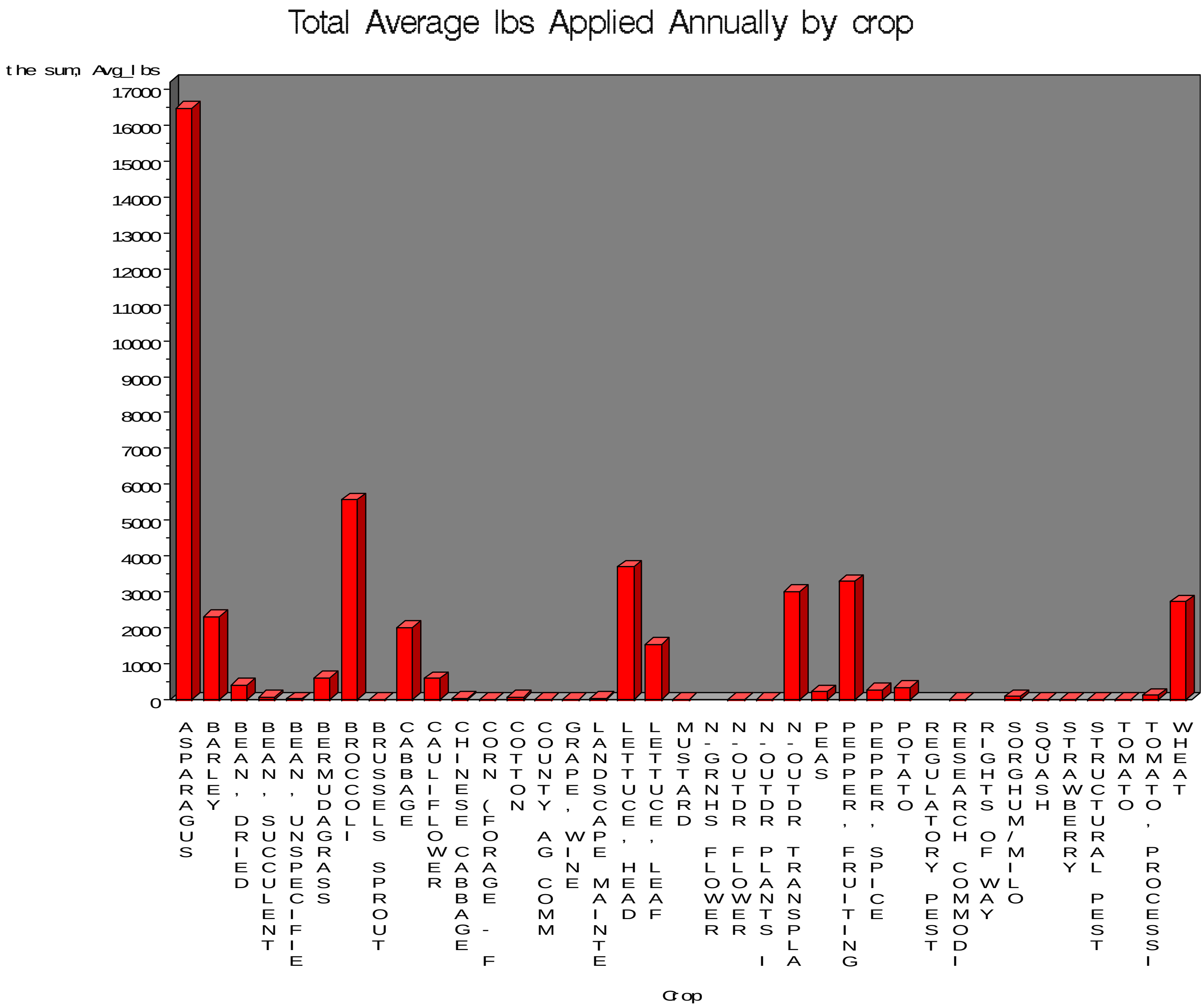


Figure 2.4. Summary of Disulfoton Use by Crop in California from 2002 to 2005

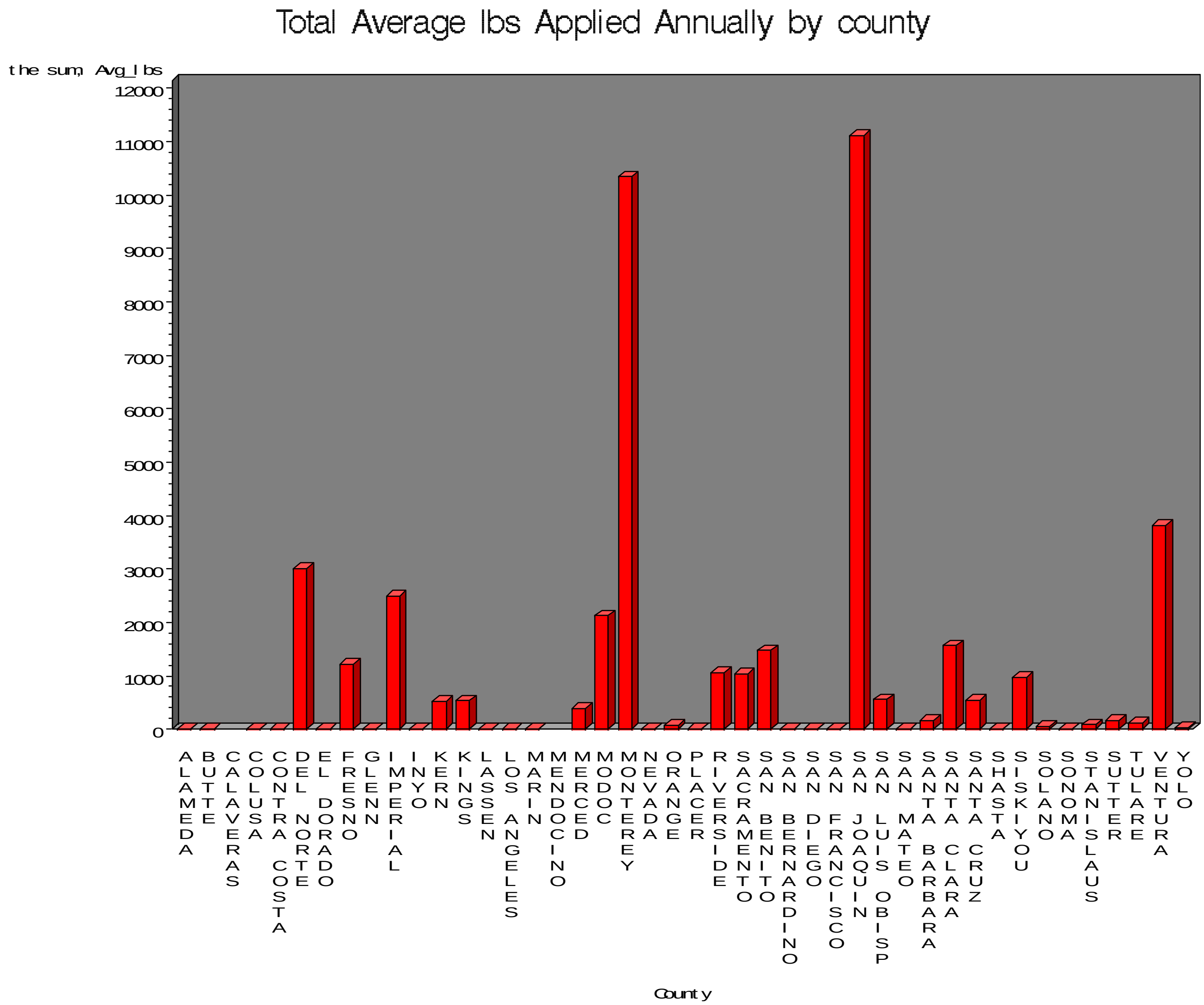


Figure 2.5. Summary of Disulfoton Use by County in California from 2002 to 2005

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 elevation 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 2.6**). 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 elevation maximum for the species of 1,500 m above sea level. The eight recovery units for the CRLF are listed in **Table 2.6** and shown in **Figure 2-6**.

Core Areas

USFWS has designated 35 core areas across the eight recovery units to focus their recovery efforts for the CRLF (see **Figure 2-6**). **Table 2.6** 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.

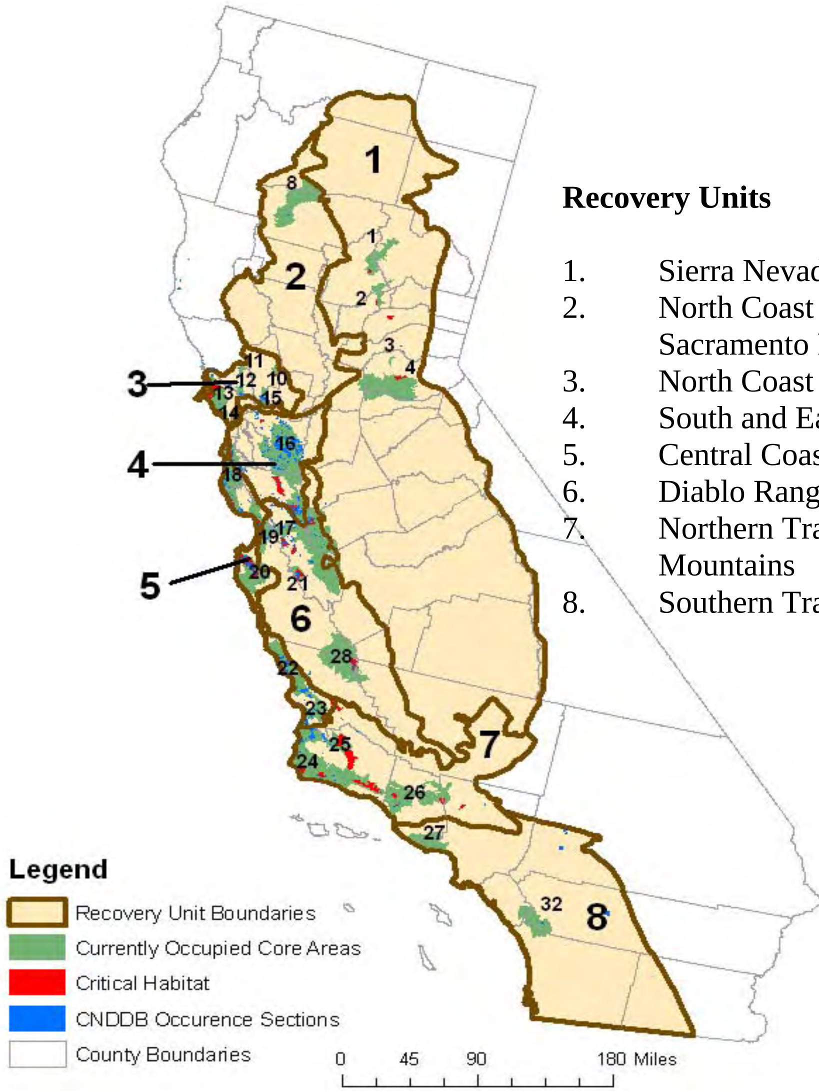
For purposes of this assessment, designated critical habitat, currently occupied (post-1985) core areas, and additional known occurrences of the CRLF from the CNDDB are considered. Historically occupied sections of the core areas are not evaluated as part of this assessment because the USFWS Recovery Plan (USFWS 2002) indicates that CRLFs are extirpated from these areas. A summary of currently and historically occupied core areas is provided in **Table 2.6** (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 2.6 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.a)	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).
[2] Core areas designated by the USFWS (USFWS 2000, pg 51).
[3] Critical habitat units designated by the USFWS on April 13, 2006 (USFWS 2006, 71 FR 19244-19346).
[4] Currently occupied (post-1985) and historically occupied core areas as designated by the USFWS (USFWS 2002, pg 54).
[5] Critical habitat unit where identified threats specifically included pesticides or agricultural runoff (USFWS 2002).
[6] 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

Figure 2.2 Recovery Unit, Core Area, Critical Habitat, and Occurrence Designations for CRLF

Core Areas

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

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

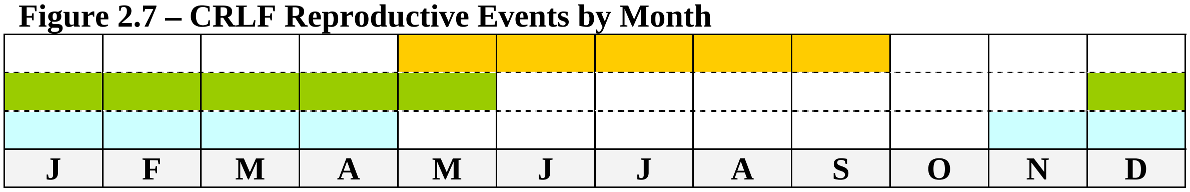
Other Known Occurrences from the CNDBB

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 2.7 depicts CRLF annual reproductive timing.

Figure 2.7 – 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 (USFWS 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 2.6**.

'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 adverse 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.

Further 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 adverse modification standards for designated critical habitat (USFWS 2006). Activities that may destroy or adversely modify critical habitat are those that alter the PCEs and jeopardize the continued existence of the species. Evaluation of actions related to use of disulfoton 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 disulfoton is expected to directly impact living organisms within the action area, critical habitat analysis for disulfoton 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 disulfoton is likely to encompass considerable portions of the United States based on the large array of 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. The Agency's approach to defining the action area under the provisions of the Overview Document (USEPA 2004) considers the results of the risk assessment process to establish boundaries for that action area with the understanding that exposures below the Agency's defined Levels of Concern (LOCs) constitute a no-effect threshold. For the purposes of this assessment, attention will be focused on the footprint of the action (i.e., the area where pesticide application occurs), plus all areas where offsite transport (i.e., spray drift, downstream dilution, etc.) may result in potential exposure within the state of California that exceeds the Agency's LOCs.

Deriving the geographical extent of this portion of the action area is based on consideration of the types of effects that disulfoton may be expected to have on the environment, the exposure levels to disulfoton that are associated with those effects, and the best available information concerning the use of disulfoton and its fate and transport within the state of California. Specific measures of ecological effect for the CRLF that define the action area include any direct and indirect toxic effect to the CRLF and any potential modification of its critical habitat, including reduction in survival, growth, and fecundity as well as the full suite of sublethal effects available in the effects literature. Therefore, the action area extends to a point where environmental exposures are below any measured lethal or sublethal effect threshold for any biological entity at the whole organism, organ, tissue, and cellular level of organization. In situations where it is not possible to determine the threshold for an observed effect, the action area is not spatially limited and is assumed to be the entire state of California.

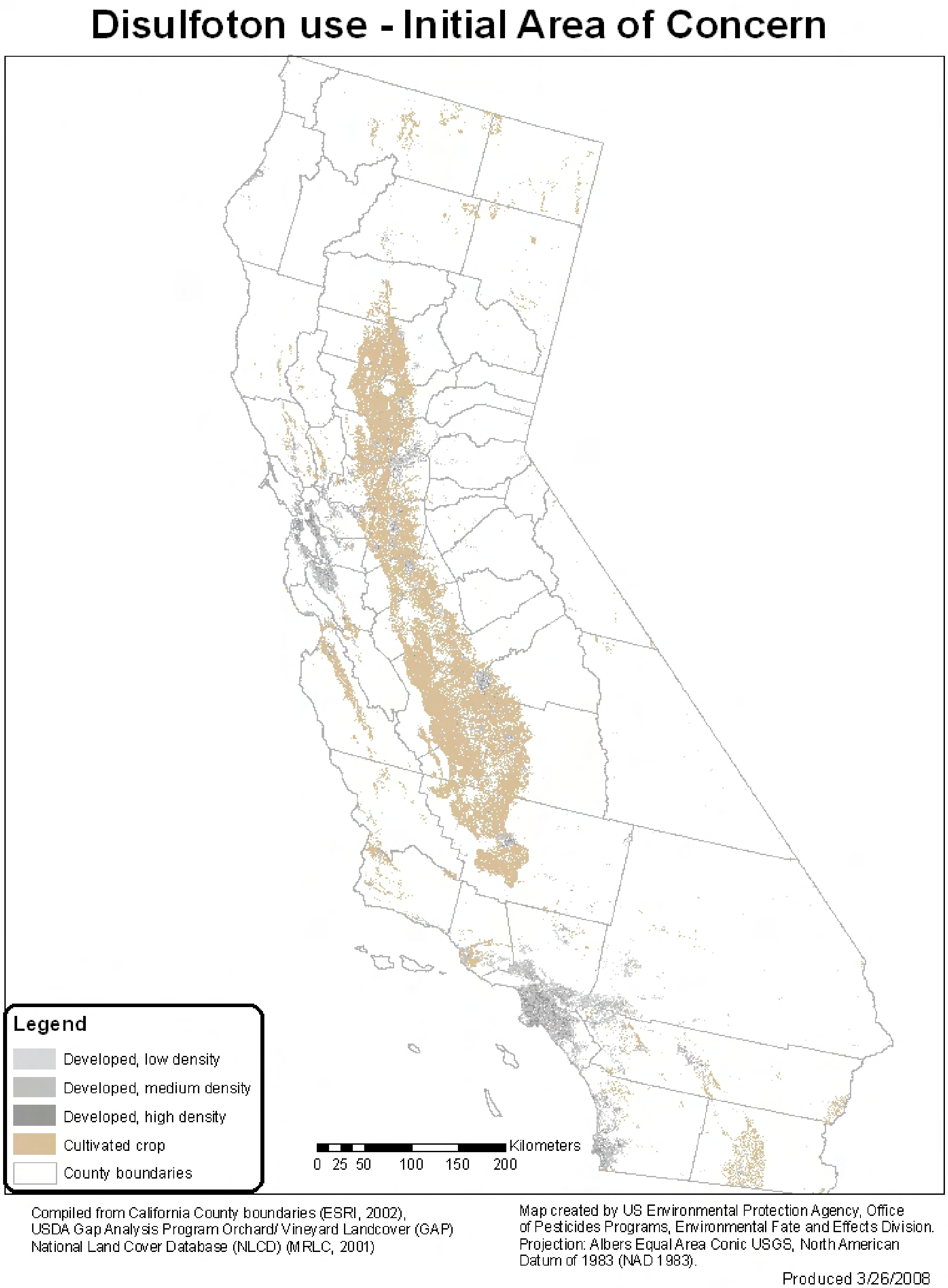
The definition of action area requires a stepwise approach that begins with an understanding of the federal action. The federal action is defined by the currently labeled uses for disulfoton. An analysis of labeled uses and review of available product labels was completed. Several of the currently labeled uses are special local needs (SLN) uses or are restricted to specific states and are excluded from this assessment. In addition, a distinction has been made between food use crops and those that are non-food/non-agricultural uses. For those uses relevant to the CRLF, the analysis indicates that, for disulfoton, the following agricultural uses are considered as part of the federal action evaluated in this assessment: asparagus, beans, broccoli, Brussels sprouts, cabbage, cauliflower, Christmas trees, cotton, and lettuce. In addition, residential uses on flowers, shrubs, and ornamentals are also considered.

Following a determination of the assessed uses, an evaluation of the potential “footprint” of disulfoton use patterns (i.e., the area where pesticide application occurs) is determined. This “footprint” represents the initial area of concern, based on an analysis of available land cover data for the state of California. The initial area of concern is defined as all land cover types and the stream reaches within the land cover areas that represent the labeled uses described above. A map representing all the land cover types that make up the initial area of concern for disulfoton is presented in **Figure 2.8**. The development of this map is described in Appendix C.

Once the initial area of concern is defined, the next step is to define the potential boundaries of the action area by determining the extent of offsite transport via spray drift and runoff where exposure of one or more taxonomic groups to the pesticide exceeds the listed species LOCs.

As previously discussed, the action area is defined by the most sensitive measure of direct and indirect ecological effects including reduction in survival, growth, reproduction, and the entire suite of sublethal effects from valid, peer-reviewed studies. Due to the lack of a defined no adverse effect concentration (NOAEC) for several reported effects (see Appendix D, ECOTOX summary), the spatial extent of the action area (i.e., the boundary where exposures and potential effects to some component of the ecosystem are less than the Agency’s LOC) for disulfoton cannot be determined. Therefore, it is assumed that the action area encompasses the entire state of California, regardless of the spatial extent (i.e., initial area of concern or footprint) of the pesticide use(s). This does not mean that there is no level below which effects are not expected to occur, but only that the available data do not allow for a determination of such a threshold.

Figure 2.8 Initial area of concern, or "footprint" of potential use, for disulfoton



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."[3] 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 disulfoton (*e.g.,* runoff, spray drift, etc.), and the routes by which ecological receptors are exposed to disulfoton *(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 or modification of its habitat. In addition, potential 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 (Appendix I) that are performed on a limited number of organisms. Additional ecological effects data from the open literature are also considered. It should be noted that assessment endpoints are limited to direct and indirect effects associated with survival, growth, and fecundity, and do not include the full suite of sublethal effects used to define the action area. According the Overview Document (USEPA 2004), the Agency relies on acute and chronic effects endpoints that are either direct measures of impairment of survival, growth, or fecundity or endpoints for which there is a scientifically robust, peer reviewed relationship that can quantify the impact of the measured effect endpoint on the assessment endpoints of survival, growth, and fecundity.

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 disulfoton is provided in **Table 2.7**.

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

Table 2.7 Assessment Endpoints and Measures of Ecological Effects

Assessment Endpoint	Measures of Ecological Effects Typically Used for Risk Assessment[4]
Aquatic-Phase CRLF *(Eggs, larvae, juveniles, and adults)*[a]	
Direct Effects	
1. Survival, growth, and reproduction of CRLF	1a. Amphibian acute LC_{50} (ECOTOX) or most sensitive fish acute LC_{50} (guideline or ECOTOX) if no suitable amphibian data are available 1b. Amphibian chronic NOAEC (ECOTOX) or most sensitive fish chronic NOAEC (guideline or ECOTOX) 1c. Amphibian early-life stage data (ECOTOX) or most sensitive fish early-life stage NOAEC (guideline or ECOTOX)
Indirect Effects and Critical Habitat Effects	
2. Survival, growth, and reproduction of CRLF individuals via indirect effects on aquatic prey food supply (*i.e.*, fish, freshwater invertebrates, non-vascular plants)	2a. Most sensitive fish, aquatic invertebrate, and aquatic plant EC_{50} or LC_{50} (guideline or ECOTOX) 2b. Most sensitive aquatic invertebrate and fish chronic NOAEC (guideline or ECOTOX)
3. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat, cover, food supply, and/or primary productivity (*i.e.*, aquatic plant community)	3a. Vascular plant acute EC_{50} (duckweed guideline test or ECOTOX vascular plant) 3b. Non-vascular plant acute EC_{50} (freshwater algae or diatom, or ECOTOX non-vascular)
4. Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation	4a. Distribution of EC_{25} values for monocots (seedling emergence, vegetative vigor, or ECOTOX) 4b. Distribution of EC_{25} values for dicots (seedling emergence, vegetative vigor, or ECOTOX)
Terrestrial-Phase CRLF *(Juveniles and adults)*	
Direct Effects	
5. Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	5a. Most sensitive bird[b] or terrestrial-phase amphibian acute LC_{50} or LD_{50} (guideline or ECOTOX) 5b. Most sensitive bird[b] or terrestrial-phase amphibian chronic NOAEC (guideline or ECOTOX)
Indirect Effects and Critical Habitat Effects	
6. Survival, growth, and reproduction of CRLF individuals via effects on terrestrial prey (*i.e.*,terrestrial invertebrates, small mammals , and frogs)	6a. Most sensitive terrestrial invertebrate and vertebrate acute EC_{50} or LC_{50} (guideline or ECOTOX)[c] 6b. Most sensitive terrestrial invertebrate and vertebrate chronic NOAEC (guideline or ECOTOX)
7. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.*, riparian and upland vegetation)	7a. Distribution of EC_{25} for monocots (seedling emergence, vegetative vigor, or ECOTOX 7b. Distribution of EC_{25} for dicots (seedling emergence, vegetative vigor, or ECOTOX)

[a] 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 that exposure pathways on land.
[b] Birds are used as surrogates for terrestrial phase amphibians.

[4] All registrant-submitted and open literature toxicity data reviewed for this assessment are included in Appendix A.

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 disulfoton 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 modify critical habitat are those that alter the PCEs and jeopardize the continued existence of the CRLF. 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 disulfoton effects data are available.

Adverse modification to the critical habitat of the CRLF includes, but is not limited to, the following, as specified by USFWS (2006):

- Alteration of water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs.
- Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs.
- Significant increase in sediment deposition within the stream channel or pond or disturbance of upland foraging and dispersal habitat.
- Significant alteration of channel/pond morphology or geometry.
- Elimination of upland foraging and/or aestivating habitat, as well as dispersal habitat.
- Introduction, spread, or augmentation of non-native aquatic species in stream segments or ponds used by the CRLF.
- Alteration or elimination of the CRLF's food sources or prey base.

Measures of such possible effects by labeled use of disulfoton on critical habitat of the CRLF are described in **Table 2.8**. 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 adverse modification standard established by USFWS (2006).

Table 2.8 Summary of Assessment Endpoints and Measures of Ecological Effect for Primary Constituent Elements of Designated Critical Habitat

Assessment Endpoint	Measures of Ecological Effect
Aquatic-Phase CRLF 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} (guideline or ECOTOX) b. Distribution of EC_{25} values for terrestrial monocots (seedling emergence, vegetative vigor, or ECOTOX) c. Distribution of EC_{25} values for terrestrial dicots (seedling emergence, vegetative vigor, or ECOTOX)
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.	a. Most sensitive EC_{50} values for aquatic plants (guideline or ECOTOX) b. Distribution of EC_{25} values for terrestrial monocots (seedling emergence or vegetative vigor, or ECOTOX) c. Distribution of EC_{25} values for terrestrial dicots (seedling emergence, vegetative vigor, or ECOTOX)
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 fish or aquatic-phase amphibians and aquatic invertebrates (guideline or ECOTOX) b. Most sensitive NOAEC values for fish or aquatic-phase amphibians and aquatic invertebrates (guideline or ECOTOX)
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.*, algae)	a. Most sensitive aquatic plant EC_{50} (guideline or ECOTOX)
Terrestrial-Phase CRLF 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 EC_{25} values for monocots (seedling emergence, vegetative vigor, or ECOTOX) b. Distribution of EC_{25} values for dicots (seedling emergence, vegetative vigor, or ECOTOX) c. Most sensitive food source acute EC_{50}/LC_{50} and NOAEC values for terrestrial vertebrates (mammals) and invertebrates, birds or terrestrial-phase amphibians, and freshwater fish.
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	
Reduction and/or modification of food sources for terrestrial phase juveniles and adults	
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	

[a] 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.

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 disulfoton to the environment. The following risk hypotheses are presumed for this endangered species assessment:

The labeled use of disulfoton within the action area may:

- directly affect the CRLF by causing mortality or by adversely affecting growth or fecundity;
- indirectly affect the CRLF by reducing or changing the composition of food supply;
- indirectly affect the CRLF 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;
- indirectly affect the CRLF 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;
- 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);
- modify the designated critical habitat of the CRLF by reducing the food supply required for normal growth and viability of juvenile and adult CRLFs;
- 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.
- 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.
- 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 disulfoton 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 2.9 and 2.10**, respectively, and the conceptual models for the aquatic and terrestrial PCE components of critical habitat are shown in **Figures**

2.11 and 2.12, respectively. Exposure routes shown in dashed lines 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.

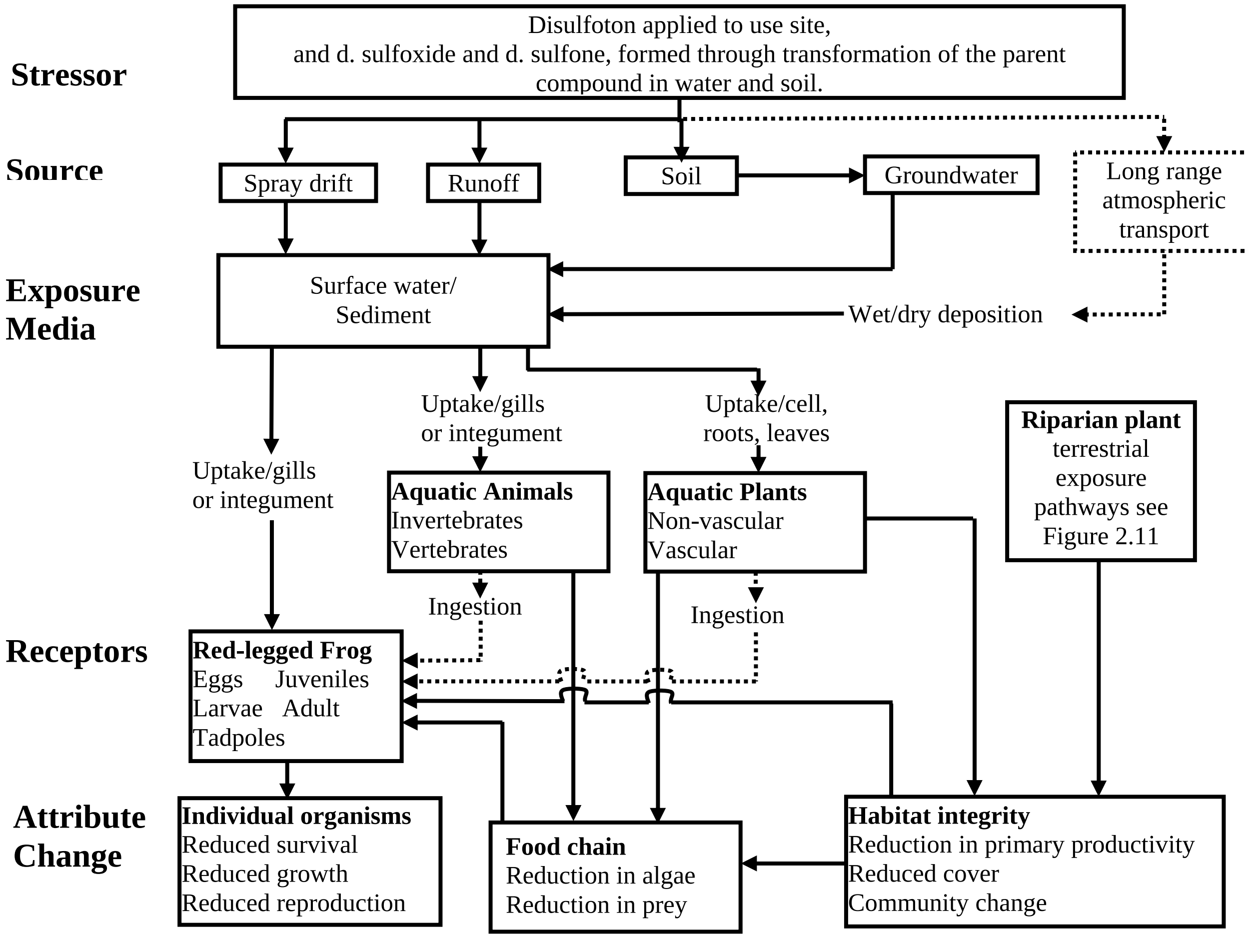


Figure 2.9. Conceptual model for disulfoton effects on aquatic phase of the red-legged frog.

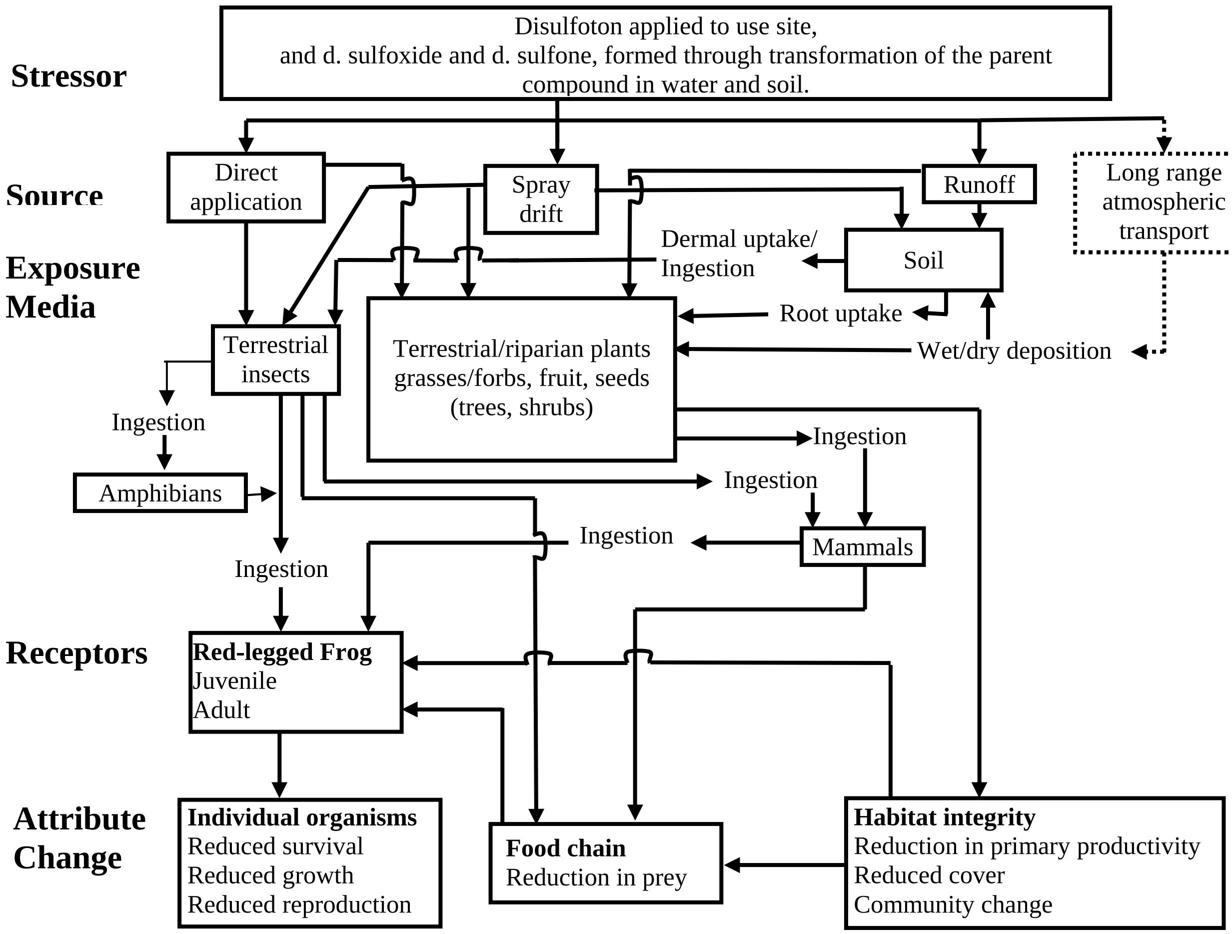


Figure 2.10. Conceptual model for disulfoton effects on terrestrial phase of the red-legged frog.

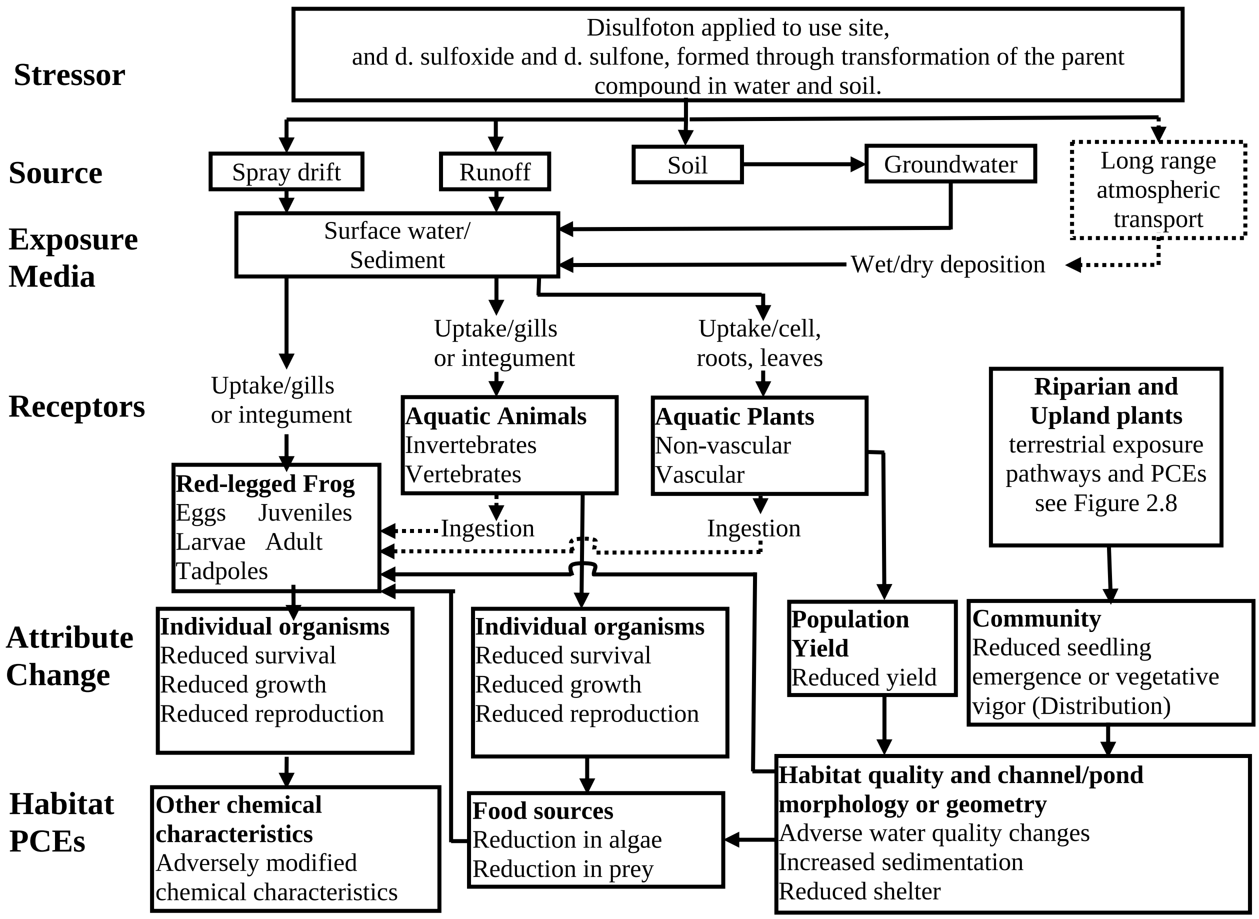


Figure 2.11. Conceptual Model for disulfoton Effects on Aquatic Component of Red-Legged Frog Critical Habitat.

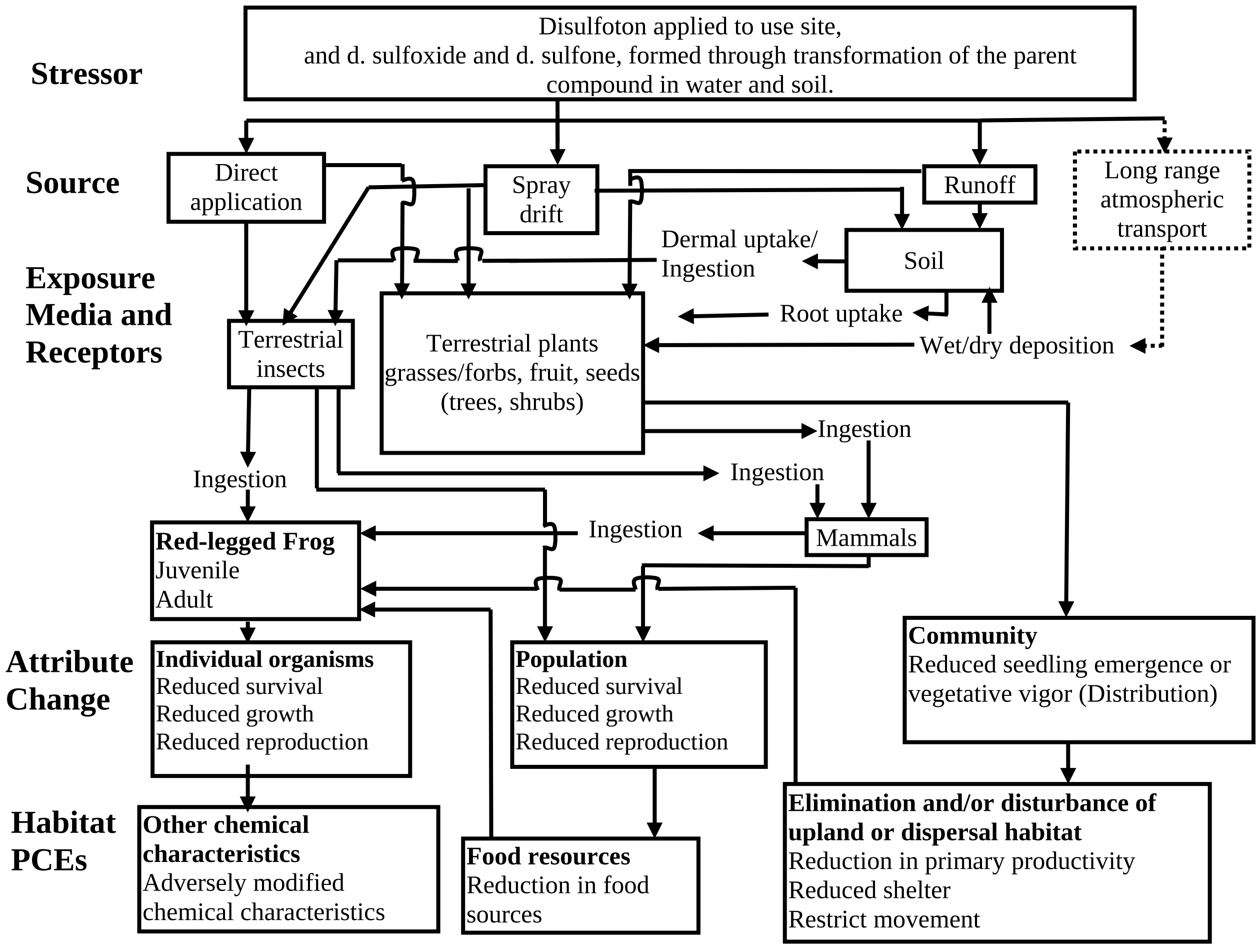


Figure 2.12. Conceptual Model for disulfoton Effects on Terrestrial Component of the Red-Legged Frog Critical Habitat.

2.10 Analysis Plan

In order to address the risk hypothesis, the potential for direct and indirect effects to the CRLF, its prey, and its habitat is estimated. In the following sections, the use, environmental fate, and ecological effects of disulfoton and its toxic major degradates are characterized and integrated to assess the risks. This is accomplished using a risk quotient (ratio of exposure concentration to effects concentration) approach. Exposure concentrations used in calculating risk quotients account for toxic major degradates of disulfoton as well as to the parent, either as "total toxic residues" (parent + sulfoxide + sulfone) or as individual species. 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 disulfoton 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 to Evaluate the Risk Hypothesis and Conceptual Model

2.10.1.1 Measures of Exposure

The environmental fate properties of disulfoton along with available monitoring data indicate that runoff and spray drift are the principle potential transport mechanisms of disulfoton to the aquatic and terrestrial habitats of the CRLF. Disulfoton has a limited potential for long-range transport. In this assessment, transport of disulfoton through runoff and spray drift is considered in deriving quantitative estimates of disulfoton exposure to CRLF, its prey and its habitats.

Measures of exposure are based on aquatic and terrestrial models that predict estimated environmental concentrations (EECs) of disulfoton and/or its degradates using maximum labeled application rates and methods of application. The models used to predict aquatic EECs are the Pesticide Root Zone Model coupled with the Exposure Analysis Model System (PRZM/EXAMS). The model used to predict terrestrial EECs on food items is T-REX. The model used to derive EECs relevant to terrestrial and wetland plants is TerrPlant. These models are parameterized using relevant reviewed registrant-submitted environmental fate data (Appendix I).

Exposure to disulfoton and two degradates were quantified (sulfone and sulfoxide degradates). The aquatic assessment utilized a total toxic residue approach to quantify potential risks from exposure to degradates. Potential risks to terrestrial organisms from exposure to each degradate were quantified. Oxon degradates of disulfoton and its sulfone and sulfoxide degradates are also a concern, but exposure to these degradates is considered qualitatively because transformation pathways and routes of exposure are not accounted for in guideline environmental fate data and because toxicity data are not available for quantitative comparison.

Aquatic Exposures

PRZM (v3.12.2, May 2005) and EXAMS (v2.98.4.6, April 2005) are screening simulation models coupled with the input shell pe5.pl (Aug 2007) to generate daily exposures and 1-in-10 year EECs of disulfoton total toxic residues that may occur in surface water bodies adjacent to application sites receiving disulfoton 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, 2-meters deep (20,000 m^3 volume) with no outlet. PRZM/EXAMS was used to estimate screening-level exposure of aquatic organisms to disulfoton.

PRZM/EXAMS modeling was conducted using a total toxic residues approach to account for exposure to disulfoton as well as its toxic degradates d. sulfoxide and d. sulfone.

Model inputs were selected based on available environmental fate studies. Half-lives were calculated from these data to represent the rate of degradation of the total toxic residues of disulfoton, including the toxic degradates d. sulfoxide and d. sulfone as well as the parent compound and adsorption inputs were based on the most mobile species.

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 for aquatic invertebrates, which are also potential prey items.

Terrestrial Exposures

Terrestrial exposure to disulfoton and two degradates were quantified (sulfone and sulfoxide degradates). Toxicity data are not available for other degradates to allow for a risk estimation. However, oxon degradates of disulfoton and its sulfone and sulfoxide degradates are also a concern. The oxon degradates form at low levels (<4%). Nonetheless, if the oxon degradates are considerably more toxic than disulfoton, then the EECs used in this assessment could underestimate potential risks.

Exposure to each degradate with available toxicity data was estimated separately by considering the highest amount of degradate that formed in available laboratory studies. The sulfoxide degradate has been shown to form up to 95% of parent and the sulfone degradate has been shown to form up to 72% of parent. It is acknowledged that exposure will likely occur to parent and degradates concurrently. The RQs for parent and each degradate are expected to encompass the potential risks for the total residues that may be found on food items of the CRLF and its prey.

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. For modeling purposes, direct exposures of the CRLF to disulfoton 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 disulfoton are bound by using the dietary based EECs for small insects and large insects.

Birds are currently used as surrogates for terrestrial-phase CRLF. However, amphibians are poikilotherms (body temperature varies with environmental temperature) while birds are homeotherms (temperature is regulated, constant, and largely independent of environmental temperatures). Therefore, amphibians 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 on a daily dietary intake basis, assuming similar caloric content of the food items. Therefore, the use of avian food intake allometric equation as a surrogate to amphibians is likely to result in an over-estimation of exposure and risk for reptiles and terrestrial-phase amphibians. Therefore, T-REX (version 1.3.1) has been refined to the T-HERPS model (v. 1.0), which allows for an estimation of food intake for poikilotherms using the same basic procedure as T-REX to estimate avian food intake.

EECs for terrestrial plants inhabiting dry and wetland areas were derived for Christmas trees using Terrplant (v. 1.2.2.). EECs were only derived for the Christmas tree use because the toxicity data indicate that the EC25 is higher than the highest labeled application rate for all disulfoton uses except Christmas trees.

AgDRIFT, a spray drift model was used to assess exposures of terrestrial phase CRLF and its prey to disulfoton 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. In addition to the buffered area from the spray drift analysis, the downstream extent of disulfoton that exceeds the LOC for the effects determination is also considered.

2.10.1.2 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 USEPA, Office of Research and Development, and the National Health and Environmental Effects Research Laboratory's Mid-Continent Ecology Division.

The assessment of risk for direct effects to the terrestrial-phase CRLF makes the assumption that potential risks to birds is similar to or less than potential risks to 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.

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 NOEC. 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.

The measures of effect for direct and indirect effects to the CRLF and its designated critical habitat are associated with impacts to survival, growth, and fecundity, and do not include the full suite of sublethal effects used to define the action area. According the Overview Document (USEPA 2004), the Agency relies on effects endpoints that are either direct measures of impairment of survival, growth, or fecundity or endpoints for which there is a scientifically robust, peer reviewed relationship that can quantify the impact of the measured effect endpoint on the assessment endpoints of survival, growth, and fecundity.

2.10.1.3 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 disulfoton, 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 disulfoton 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).

This assessment estimated potential risk from exposure to total toxic residues (TTR) or to parent disulfoton and specific degradates for which toxicity data exists. Toxicity data are not available for other degradates to allow for a risk estimation. Of particular concern are oxon degradates of disulfoton and its sulfone and sulfoxide degradates. The oxon degradates form at low levels (<4%); however, if the oxon degradates are considerably more toxic than disulfoton, then the EECs used in this assessment could underestimate potential risks.

For this endangered species assessment, listed species LOCs are used for comparing RQ values for acute and chronic exposures of disulfoton directly to the CRLF. If estimated exposures directly to the CRLF of disulfoton 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 disulfoton 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 RQ being considered also exceeds the non-listed species acute risk LOC, then the effects determination is a LAA. If the acute RQ is between the listed species LOC and the non-listed acute risk 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 "may affect".

3. Exposure Assessment

Disulfoton is formulated as an emulsifiable concentrate and granular formulation. For most crops, it is applied directly to and incorporated in soil. However, for others, it may be applied by air or ground spray.

Risks from ground boom and aerial applications are expected to result in the highest off-target levels of disulfoton 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. Disulfoton is also labeled for application through soil injection. With soil injection, runoff is the primary route of off-target transport.

Exposure to major toxic degradates of disulfoton is considered in this assessment as well as exposure to the parent. The aquatic exposure assessment estimates concentrations of total toxic residues, or parent + d. sulfoxide + d. sulfone. The terrestrial exposure assessment estimates exposure to each compound individually.

3.1 Label Application Rates and Intervals

Disulfoton labels may be categorized into two types: labels for manufacturing uses (including technical grade disulfoton and its formulated products) and end-use products. While technical products, which contain disulfoton of high purity, are not used directly in the environment, they are used to make formulated products, which can be applied in specific areas to control aphids, mites, thrips, and other insect pests. The formulated product labels legally limit disulfoton's potential use to only those sites that are specified on the labels.

Currently registered agricultural uses of disulfoton relevant to CRLF critical habitat in California include use on asparagus, beans, broccoli, Brussels sprouts, cabbage, cauliflower, Christmas trees, and cotton. Non-agricultural uses include application to residential flower beds and vegetable gardens. The uses being assessed are summarized in **Table 3.1**. More detail about use patterns is provided in Section 2.4. Presence of a 25-foot well maintained vegetative buffer strip between application sites and all permanent water bodies is also specified on the labels for all agricultural uses. For residential uses, determining application rates in lb a.i./A, as necessary for modeling purposes, required making assumptions about treated area. The highest residential application rate of 0.02 lb a.i./1000 ft^2, labeled for broadcast granular application to beds prior to planting, was estimated to be equivalent to 1.6 lb a.i./A, based on the assumption of 0.25 acre lots with 20 x 100 ft gardens on each lot, leading to 8,000 ft^2 treated per acre.

Table 3.1. Labeled use pattern for each crop, used in assessing disulfoton environmental exposure.

Uses	Application Methods	Application Rate (*lb a.i./A*)	No. of Apps.	Application Interval (*days*)	PHI (*days*)
Asparagus	Ground spray, Aerial spray	1	2	7[1]	180
Beans	Soil injection	1	1	n/a	60
Broccoli	Soil injection	1	1	n/a	14
Brussels sprouts	Soil injection	1	1	n/a	30
Cabbage	Soil injection	2	1	n/a	42
	Ground spray	1	1	n/a	40
Cauliflower	Soil injection	1	1	n/a	NS [d]
Christmas trees	Broadcast granular	4.5	1	n/a	NS
Cotton	Soil injection, Ground spray (in furrow)	1	1	n/a	NS
	Drill planting	0.975	1	n/a	NS
	Hill drop planting	0.325	1	n/a	60
Lettuce	Soil injection, Chemigation (drip or trickle)	2	1	n/a	NS
Residential Uses	Broadcast granular	1.6[2]	1[3]	n/a	NS

[1] Label does not specify application interval. Modeling assumed that a 7 day interval would be conservative.

[2] The highest labeled application rate is 0.2 lb a.i./1000 ft^2 (Reg. No. 432-1286, BEAD LUIS report 8/22/07) Based on an assumption of 4 lots per acre, each lot with a flower bed/garden of 200 ft x 10 ft, this is equivalent to 1.6 lb a.i./A.

[3] Label does not specify the number of applications. The broadcast applications are intended for planting time, so modeling assumed a single application.

3.2 Aquatic Exposure Assessment

3.2.1 Modeling Approach

Surface water aquatic exposures for all assessed uses are quantitatively estimated using the Pesticide Root Zone Model coupled with the Exposure Analysis Model System (PRZM/EXAMS). These screening level models are operated based on scenarios that represent high exposure sites for disulfoton 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.

Additionally, PRZM/EXAMS modeling does not account for transport to groundwater followed by discharge to surface water as a possible route of aquatic exposure. Discharging groundwater is likely to support low-order streams, wetlands, and intermittent ponds, environments that are favorable to California Red-Legged Frogs (CRLFs). Groundwater specific modeling is not conducted in this assessment. Long-term chronic concentrations derived from the PRZM-EXAMS model should reflect background concentrations that might be found in discharged groundwater/stream baseflow.

Because the disulfoton degradates d. sulfoxide and d. sulfone are also toxic, modeling was conducted to estimate exposure to total toxic residues (TTR) of all three compounds. Crop-specific management practices for all of the assessed uses of disulfoton were used for modeling, including application rates, number of applications per year, application intervals, buffer widths and resulting spray drift values modeled from AgDRIFT, and the first application date for each crop.

3.2.2 Model Inputs

3.2.2.1 Physical Properties and Environmental Fate Inputs

Disulfoton environmental fate data were discussed previously and are listed in **Table 2.2.** Chemical-specific model input parameters for PRZM and EXAMS are based on these data and are listed in **Table 3.2**. Environmental fate inputs represent properties of the total toxic residues (TTR) of disulfoton, including the toxic degradates d. sulfoxide and d. sulfone as well as the parent compound. Oxon degradates are not considered explicitly in this estimation. TTR transformation rates were estimated based on studies conducted on the parent compound. For each study, first order log linear TTR half-lives were calculated using the sum of all three species detected at each sampling point. Soil-water partitioning coefficients (Kd) were measured directly for each toxic species. In order to provide a conservative exposure estimate, the Kd from the most mobile compound, d. sulfoxide, was chosen as a modeling input.

Table 3.2. PRZM/EXAMS Environmental Fate Inputs for Aquatic Exposure to Total Toxic Residues of Disulfoton

Fate Property	Value[1]	Comment	Source
Molecular Weight	274.39		MRID 150088
Henry's constant	2.6×10^{-6} atm m^3/mol		EFED One-liner 5/21/97
Vapor Pressure	1.8×10^{-4} mm Hg		MRID 150088
Solubility in Water	150 mg/L	Measured value x 10	EFED One-liner 5/21/97
Photolysis in Water	141 days		MRID 40471102
Aerobic Soil Metabolism Half-lives	418 days	Upper 90% confidence bound on the mean of three values	MRID 40042201, 41585101, 43800101
Hydrolysis at pH 7	Stable	TTR remain at 97% at end of study.	MRID 00143405
Aerobic Aquatic Metabolism (water column)	181 days	Single value x 3, corrected for hydrolysis	MRID 49691201
Anaerobic Aquatic Metabolism (benthic)	Stable	Single value x 3 = 1100 days	MRID 46316901
K_{oc}	70 mL/g_{OC}	Mean K_{oc} for d. sulfoxide, the most mobile component	MRID 46766601

1 – 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

3.2.2.2 Use-specific Management Practices Inputs

Use specific management practices for all of the assessed uses are also considered in modeling. Application rates, intervals, and methods were previously listed in **Table 3.1**. Scenarios and other crop-specific model input parameters for PRZM and EXAMS are included in **Table 3.3**, with justification for these inputs discussed below.

Table 3.3. PRZM/EXAMS Use-Specific Aquatic Exposure Inputs for Total Toxic Residues of Disulfoton

Use	PRZM Scenario	App. date	Application Method	CAM [a]	App. Efficiency (%)	Drift (%)
Asparagus	CA Row Crop	Sep. 15	Ground spray	2	99	2.7
			Aerial spray	2	95	9
Beans	CA Row Crop	Jan. 15	Soil injection	5	100	0
Broccoli, Cauliflower	CA Cole Crop	Feb. 8	Soil injection	5	100	0
Brussels sprouts	CA Lettuce	Feb. 1	Soil injection	5	100	0
Cabbage	CA Cole Crop	Feb. 8	Soil injection	5	100	0
			Ground spray	1	99	2.7
Christmas trees	CA Forestry	Aug. 28	Broadcast (granular)	1	100	0
Cotton	CA Cotton	Apr. 25	Soil injection	5	100	0
			Ground spray (in furrow)	1	99	1
			Drill planting	5	100	0
			Hill-drop planting	5	100	0
Lettuce	CA Lettuce	Feb. 1	Ground spray	1	99	2.7
			Chemigation (drip or trickle)	1	100	0
Residential	CA Residential	Apr. 1	Broadcast (granular)	1	100	0

[a] CAM 1 = Soil applied, default incorporation depth of 4 cm, linearly decreasing with depth
CAM 2 = Foliar applied
CAM 5 = Soil applied, incorporation depth of 4 cm, linearly increasing with depth.

3.2.2.3 PRZM/EXAMS Scenarios and Application Dates

Use-specific parameters are input into modeling scenarios which have been developed to represent locally specific soil and climatic conditions in vulnerable use sites. Scenarios also include crop specific agronomic data and management practices such as planting and harvest date. A group of scenarios are available to represent crops grown in California, including standard scenarios as well as scenarios developed specifically for CRLF assessments. For most disulfoton uses, scenarios are available which were developed specifically for that use. In cases where a scenario does not exist for a specific use, it is necessary to assign a surrogate scenario. Asparagus and beans are both included as crops for which the California Row Crop scenario was developed. The California Cole Crop scenario was developed based on broccoli and is appropriate for cauliflower and cabbage as well, because these are also cole crops with similar cultivation requirements as broccoli. The California Lettuce scenario was developed for lettuce and is also an appropriate surrogate for Brussels sprouts, which are leafy vegetables with similar cultural practices as lettuce. The California Forestry scenario was developed to represent Northern California forests, which include Christmas tree farms. The California Residential scenario was developed specifically for residential uses on lawns and gardens.

Application dates are not specified on product labels. For modeling, the dates of first application were developed based on label instructions, crop profiles maintained by the USDA and University of California (http://www.ipm.ucdavis.edu/PDF/PESTNOTES/index.html, http://www.ipmcenters.org/cropprofiles/GetCropProfiles.cfm; accessed 5/08), historical use data from the California PUR dataset, and planting dates and precipitation data specific to each scenario (scenario metadata: http://www.epa.gov/oppefed1/models/water/state_crop.htm). Several of the labeled use patterns specify application at time of planting while others also allow application after plants are established, as necessary. Crop profiles indicate that, due to the long pre-harvest intervals required by labels, disulfoton is typically applied early in the growing season. Many of these crops can be planted and harvested year-round, so for modeling purposes the early season was defined by the crop emergence, maturity and harvest dates specified in each scenario. Modeled application dates were selected to be between the planting date and the limit of the pre-harvest interval, as defined in the scenario. Within this window, dates were selected to have a high potential for runoff, as indicated by precipitation data from local weather stations, in order to provide conservative exposure estimates. Historical data were considered as well to verify that the chosen application date adequately represented typical use pattern. For residential uses and application to Christmas trees, emergence and harvesting dates are less relevant and historical use data were not available, so other assumptions had to be made. For each disulfoton use, the selected application date and its justification are defined in **Table 3.4**.

Table 3.4. PRZM/EXAMS Disulfoton Application Dates		
Use	**App. date**	**Application date comments**
Asparagus	Sep. 15	PUR data 2001-2005 indicate that most applications to asparagus occur in August – October. Within this time period, Sep. 15 had the highest precipitation.
Beans	Jan. 15	CA ROW CROP SCENARIO (*Crop emergence*: Jan 1, *Crop harvest*: Apr 8). Label specifies apply at seeding. Near the scenario planting time, Jan 15 had the highest precipitation.
Broccoli, Cabbage, Cauliflower	Feb. 8	CA COLE CROP SCENARIO (*Crop emergence*: Jan 1, *Crop harvest*: Mar 1). Between planting and the limit of the PHI, Feb. 8 had the highest precipitation.
Brussels sprouts, Lettuce	Feb. 1	CA LETTUCE SCENARIO (*Crop emergence*: Feb 16, *Crop harvest*: May 12). Between planting and the limit of the PHI, Feb. 1 had the highest precipitation.
Cotton	Apr. 25	CA COTTON SCENARIO (*Crop emergence*: May 1, *Crop harvest*: Nov. 11) Label specifies apply at seeding. Near the scenario planting time, Apr 25 had the highest precipitation.
Christmas Trees	Jun. 28	No PUR historical use data are available for Christmas trees. UC crop profiles indicate that pest pressure from spider mites and aphids, the primary targets of disulfoton on Christmas trees, is highest between June and September. Multi-run modeling found that within this season, Jun 28 provided conservative EECs.
Residential	Apr. 1	Most residential uses intend that disulfoton be applied at planting. For modeling, EFED assumed that residential planting occurs in the spring. Apr. 1 was the highest precipitation date in this season.

3.2.2.5 Application Methods

Modeling parameters for which inputs are based on the application method include Chemical Application Method (CAM) as well as application efficiency and spray drift. CAM 1 represents application directly to soil and was used for ground spray prior to crop emergence, chemigation, and broadcast granular application methods. This CAM assumes maximum active ingredient on the soil surface with concentrations decreasing linearly with depth to a default incorporation depth of 4 cm. CAM 2 represents foliar applications and was input for the asparagus use for both ground and aerial spray methods because the asparagus label indicates the application is made after emergence. CAM 5 represents application beneath the soil, with concentrations increasing linearly with depth to a maximum concentrations at a user defined depth. In this case, the incorporation depth was defined as 4 cm. Disulfoton is intended to be applied near plant roots, and crop profiles indicate that most of these crops are planted to depths of less than 1 inch. Therefore a 4 cm injection depth was used in order to be consistent with the default incorporation assumptions for CAM 1.

Application efficiency inputs were set to the default values corresponding to each application method: 100% efficiency for all soil injection, granular, and drip irrigation applications, 99% for ground spray applications, and 95% for aerial spray applications. Spray drift is set to 0% for granular and soil injection application methods, because it is assumed that using these methods, all active ingredient remains on the treated field and that drift is not a factor. For spray applications, spray drift inputs were estimated using the AgDRIFT model in order to account for the effect of the label requirement for 25-ft buffer zones between cropped areas and water bodies. For aerial applications, the AgDRIFT default is to assume a droplet size distribution that is fine to medium. This leads to a spray drift estimate of 9% of the applied active ingredient. For broadcast ground spray applications, the estimated drift was 2.7% of the applied, based on the conservative assumptions that a high boom height is used and that the droplet size distribution is very fine to fine. Some ground spray applications of disulfoton are specified as in-furrow spray which has a lower potential for drift than broadcast applications. In these cases, use of a low boom was assumed to be more appropriate, leading to an estimated drift of 1% of the applied. The low boom height considered by AgDRIFT is 4 ft, which is likely conservative for an in-furrow spray application.

3.2.3 Results

PRZM/EXAMS EECs representing 1-in-10 year peak, 21-day, and 60-day concentrations of total toxic residues of disulfoton in the aquatic environment are located in **Table 3.5**. All model output are included in Appendix E. Estimated aquatic exposures are highest for disulfoton use on lettuce with a peak EEC of 67 ug/L. Peak EECs for other agricultural crops ranged from 1.8 ug/L for beans to 24 ug/L for cabbage. Use on residential gardens and flowerbeds resulted in a peak EEC of 3.7 ug/L.

Table 3.5. Aquatic EECs (µg/L) for total toxic residues of disulfoton uses in California

Crops Represented	Application Method	Peak EEC	21-day average EEC	60-day average EEC
Asparagus	Ground spray	18.8	17.3	14.6
	Aerial spray	22.6	20.7	17.7
Beans	Soil injection	1.8	1.6	1.3
Broccoli	Soil injection	3.6	3.3	2.8
Brussels sprouts	Soil injection	5.8	5.4	4.6
Cabbage	Ground spray	23.6	23.2	21.8
	Soil injection	7.1	6.6	5.5
Cauliflower	Soil injection	3.6	3.3	2.8
Christmas trees	Broadcast granular	15.1	14.1	12.3
Cotton	Soil injection	0.6	0.5	0.4
	Hilldrop	0.8	0.7	0.6
	Drill Planting	2.3	2.1	1.7
	Ground spray (in furrow)	4.8	4.3	3.5
Lettuce	Soil injection	11.6	10.8	9.2
	Chemigation	66.7	61.8	53.7
Residential Uses	Broadcast granular	3.7	3.5	3.2

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. In the case of disulfoton, however, available monitoring data are primarily for parent alone and so are of limited utility in validating model estimates for total toxic residues. Disulfoton is less persistent than its other toxic degradates, so impacts from disulfoton use may not be identified through disulfoton sampling alone. Included in this assessment are California-specific disulfoton data for both surface and groundwater from the USGS NAWQA program (http://water.usgs.gov/nawqa), which include limited sampling for degradates d. sulfoxide and d. sulfone, and surface water data from the California Department of Pesticide Regulation (CDPR). Additionally, this discussion includes national surface water and groundwater data presented in the previous RED, from NAWQA, from the STORET database, and from several individual local studies.

3.2.4.1 Previous Assessment Surface Water Data

The disulfoton RED published in 2002 considered national monitoring data available as of that time. Although these data do not represent California-specific environmental conditions, they are useful to provide insight into the potential for transport of disulfoton to surface water and groundwater. On a national scale, NAWQA data collected through 1998 had detections in 0.27% of all samples at levels from 0.01 to 0.06 ug/L, with detections in 0.20% of samples from agricultural streams and 0.61% of samples from urban streams. A separate monitoring study in Virginia targeted to one watershed (50% agricultural/50% forested) detected disulfoton in 3 samples at 2 sites at concentrations from 0.37 to 6.11 ug/L. The low detection was at the same site as the high, but collected three hours later.

3.2.4.2 Previous Assessment Ground Water Data

The previous RED discusses groundwater data from studies found in the Pesticides in Ground Water database. Disulfoton was tested for with no detections in a number of studies nationally, including 974 wells in California. No details are reported about the studies, although the RED notes that detection limits as high as 6 ug/L reduce the certainty of the data. The RED also discusses 3 groundwater monitoring studies which targeted vulnerable wells in agricultural areas. One study in North Carolina had no detections while two studies, in Virginia and Wisconsin, had detections from 0.01 ug/L to 100 ug/L. In Virginia, monthly sampling for 4 years detected disulfoton at 5 of 8 wells. Six detections ranged from 0.04 to 2.87 ug/L. In Wisconsin, individual samples from 29 wells were tested for disulfoton with detections ranging from 4.0 to 100 ug/L. The samples were from the Central Sands area of Wisconsin where environmental conditions are conducive to preferential flow, and at least 14 other pesticides were detected in the samples, so the observed leaching may have been due to local environmental conditions that may not be typical in California. These studies demonstrate that in some conditions, disulfoton has the potential to reach groundwater, but Application practices in these watersheds are not reported and the environmental conditions may differ from those typical in the region of concern for the CRLF.

3.2.4.3 USGS NAWQA Surface Water Data

NAWQA monitoring data are available for disulfoton in California surface waters, although this monitoring does not target specific chemicals or uses. Between 1993 and 2006, 1920 samples were taken at 74 sites with no detections of disulfoton above the detection limit of 0.02 ug/L. Sites included those with land cover classified as agriculture, urban, mixed, and other, and the majority were in Stanislaus, San Bernardino, and Merced counties. Six sites were sampled for d. sulfone as well as disulfoton and 3 of these were also sampled for d. sulfoxide. D. sulfone was detected at 2 sites, one agricultural site in Stanislaus county and one urban site in Sacramento county (DL = 0.006 - 0.016 ug/L). In Stanislaus county, d. sulfone was detected in 1 out of 28 samples at 0.01 ug/L. At the Sacramento county site, d. sulfone was detected in 14 out of 14 samples taken over 15 months. The peak level of 0.084 ug/L was detected in June and a steady decline to 0.018 ug/L was observed in biweekly samples through October. Bimonthly samples through the following August remained below 0.036 ug/L except for one spike to 0.069 ug/L in June. There were no detections for d. sulfoxide.

3.2.4.4 USGS NAWQA Groundwater Data

The NAWQA groundwater California dataset included 672 samples from 374 wells analyzed for disulfoton between 1993 and 2006. 90 of these samples were also tested for d. sulfoxide and 171 for d. sulfone. There were no detections of either parent or degradates in any of the analyzed samples. Detection limits for all species were $\leq$ 0.02 ug/L. Samples represented all NAWQA study areas in California. 47% of the parent samples were from sites with agricultural landcover, 40% with mixed and/or other, and 13% with urban land cover.

3.2.4.5 California Department of Pesticide Regulation (CDPR) Data

CDPR maintains a database of monitoring data of pesticides in CA surface waters. The sampled water bodies include rivers, creeks, urban streams, agricultural drains, the San Francisco Bay delta region and storm water runoff from urban areas. The database contains data from 51 different studies by federal state and local agencies as well as groups from private industry and environmental interests. Some data reported in this database are also reported by USGS in NAWQA; therefore, there is some overlap between these two data sets (http://www.cdpr.ca.gov/docs/emon/surfwtr/surfdes.htm).

From 1991-2005, 2712 samples from 173 CA surface water sites were analyzed for disulfoton. About 60% of the samples are from the San Joaquin Valley region and 25% from the Sacramento Valley, with the remaining samples dispersed throughout the state. There were no disulfoton detections above the detection limits of 0.01 to 1 ug/L. D. sulfoxide and d. sulfone were not included in the database.

3.2.4.6 Atmospheric Monitoring Data

Available studies monitoring atmospheric transport in the Central Valley and Sierra Nevada do not include disulfoton as an analyte (http://www.cdpr.ca.gov/docs/empm/pubs/tac/tacstdys.htm; http://www.nature.nps.gov/air/Studies/air_toxics/wacap.cfm; Majewski, 1995). One national study is available which tested for disulfoton at 10 sites, finding it in only 1 out of 123 samples at a concentration of 0.0047 ng/L, suggesting that disulfoton is not likely to be present in ambient air (Carey and Kutz, 1985).

3.3. Terrestrial Animal Exposure Assessment

T-REX (Version 1.3.1) is used to calculate dietary and dose-based EECs of disulfoton for the CRLF and its potential prey (*e.g*. small mammals and terrestrial insects) inhabiting terrestrial areas. EECs used to represent the CRLF are also used to represent exposure values for frogs serving as potential prey of CRLF adults. T-REX simulates a 1-year time period.

For assessing potential risk to the **terrestrial**-phase CRLF and its prey (*e.g*. terrestrial insects, small mammals and terrestrial-phase frogs), exposures to disulfoton resulting from spray applications were modeled, which include applications to asparagus via ground and aerial spray. Also, ground broadcast spray is allowed for additional crops including broccoli, Brussels sprouts, cabbage, cauliflower, Christmas trees, cotton, and Easter lilies. These are pre-plant applications followed by incorporation into soil. Foliar residues on the field from these uses are expected to be lower than those resulting from direct foliar applications from the asparagus use. However, drift potential off the field remains equivalent to foliar ground spray applications, and insect EECs are presumed to be comparable for soil and foliar sprays. Therefore, the T-REX estimates of pesticide residues on insects will be used for all uses with ground spray, regardless of foliar or soil application.

Disulfoton may also be applied to soil in furrow (incorporated), via injection, or as a granular that is subsequently wetted into the soil. Potential risks from these applications were estimated using the LD50 per square foot method. In addition, disulfoton is a systemic insecticide that is taken up into the plant, and insects are killed when they consume contaminated plants. EFED's current methodologies do not allow for quantification of potential exposures and risks from consumption of systemic pesticides that have been taken up through the root system and distributed throughout the plant. Therefore, potential risks from this exposure route will be qualitatively discussed.

Given that no data on interception and subsequent dissipation from foliar surfaces suitable for estimating foliar dissipation half-lives is available for disulfoton, a foliar dissipation half-life of 35 days (default) was used. However, foliar dissipation half life was only used in the derivation of EECs for asparagus because multiple applications are not allowed for crops other than asparagus.

T-REX is also used to calculate EECs for terrestrial insects exposed to disulfoton. Dietary-based EECs calculated by T-REX for small and large insects (units of a.i./g) are used to bound an estimate of exposure to bees. Available acute contact toxicity data for bees exposed to disulfoton (in units of µg a.i./bee), are converted to µg a.i./g (of bee) by multiplying by 1 bee/0.128 g. The EECs are later compared to the adjusted acute contact toxicity data for bees in order to derive RQs.

For modeling purposes, exposures of the CRLF to disulfoton 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 are assessed using the small mammal (15 g) which consumes short grass. Upper-bound Kenega nomogram values reported by T-REX for these two organism types are used for derivation of EECs for the CRLF and its potential prey (**Table 3.6**). EECs used to estimate potential exposures to insects are presented in **Table 3.7**. An example output from T-REX v. 1.3.1 is available in Appendix F.

EECs for the sulfoxide and sulfone degradates were estimated by multiplying the maximum amount of degradate formed in available laboratory studies by the disulfoton EEC. For example, the sulfoxide degradate has been shown to form up to 95% of parent disulfoton. Therefore, the sulfoxide EEC would equal the disulfoton EEC x 0.95 x (MW degradate / MW disulfoton).

Table 3.6 Upper-bound Kenega Nomogram EECs for Dietary- and Dose-based Exposures of the CRLF and its Prey to disulfoton

Use	Application Rate (lbs a.i./Acre)	EECs for CRLF		EECs for Prey (small mammals)	
		Dose-based EEC	Dietary-based EEC	Dose-based EEC	Dietary-based EEC
Cabbage , cotton	1 Single application	154 mg/kg-bw	135 mg/kg-diet	228 mg/kg-bw	240 mg/kg-diet
Asparagus	1 (2 apps, 14-day interval)	270 mg/kg-bw	240 mg/kg-diet	402 mg/kg-bw	420 mg/kg-diet

LD50/square foot analysis[a]

Use	Application Rate (lbs a.i./Acre)	Application Method	EEC Assumptions	EEC (mg a.i./f^{t2})
Beans, broccoli, Brussels sprouts, cabbage, cauliflower, cotton	1	Soil injection; Incorporated ground spray (cotton)	99% incorporation; assumed a 6-inch furrow and 12-inch spacing between furrows/rows	0.33
Christmas trees	4.5	Granular broadcast, wetted in	85% Incorporated	7.03
Cabbage and Lettuce	2	Soil injection	99% incorporation; assumed injection occurs within a 6-inch space and 12-inch spacing between rows	0.42
Residential	1.6	Granular broadcast, wetted in	85% Incorporated	2.5

[a] LD50 per square foot analysis does not include potential exposures from consumption of treated plants or contaminated insects that have consumed treated plants.

Table 3.7 EECs (ppm) Used to Estimate Indirect Effects to the Terrestrial-Phase CRLF via Effects to Terrestrial Invertebrate Prey Items

Use	Small Insect	Large Insect
Asparagus (1 lb a.i./Acre, 2 applications, 14-day interval)	240	26
Cabbage , cotton (1 lb a.i./Acre, 1 application)	135	15

3.4. Terrestrial Plant Exposure Assessment

Potential risks to terrestrial plants were quantified for Christmas trees (granular formulation). The EC25 and NOAEC in plants was greater than the maximum application rate for spray applications for all uses except Christmas trees. Inputs use for Terrplant (v. 1.2.2) are in Table 3.8, and results are listed in Table 3.9.

Table 3.8. Input Parameters Used to Derive Terrestrial Plant EECs.

Input Parameter	Symbol	Value	Units
Application Rate	A	4.5	lbs a.i./acre
Incorporation	I	1	none
Runoff Fraction	R	0.02	none
Drift Fraction	D	0	none
Seedling Emergence EC25	--	1.9	lbs a.i./acre
Vegetative Vigor EC25	--	2.4	lbs a.i./acre

Table 3.9. Terrestrial Plant EECs for Disulfoton. Units in lbs a.i./acre.

Description	EEC
Runoff to dry areas	0.09
Runoff to semi-aquatic areas	0.9
Spray drift	0 (granular formulation)
Total for dry areas	0.09
Total for semi-aquatic areas	0.9

4. Effects Assessment

This assessment evaluates the potential for disulfoton 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 effects determination 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, given that birds are generally used as a surrogate for terrestrial-phase amphibians. 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 disulfoton.

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 ECOTOX 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 for the effects determination 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 available. Although the effects determination relies on endpoints that are relevant to the assessment endpoints of survival, growth, or reproduction, it is important to note that the full suite of sublethal endpoints potentially available in the effects literature (regardless of their significance to the assessment endpoints) are considered to define the action area for disulfoton.

Additional information on the sublethal effects available in the open literature and evaluated by the Health Effects Division (HED) is included in Appendix G.

Citations of all open literature, including those 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), are included in Appendix H. Appendix H 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. A detailed spreadsheet of the available ECOTOX open literature data, including the full suite of lethal and sublethal endpoints is presented in Appendix D.

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 disulfoton. A summary of the available aquatic and terrestrial ecotoxicity information, use of the probit dose response relationship, and the incident information for disulfoton are provided in Sections 4.1 through 4.4, respectively.

This assessment evaluates the potential for disulfoton to adversely affect the CRLF. Two degradates are also included in this assessment. The available data suggests that the sulfone and sulfoxide degradates may also be a concern to both aquatic and terrestrial phase CRLFs. Toxicity data for disulfoton and the two degradates of concern are also discussed in Sections 4.1 to 4.4.

4.1 Toxicity of Disulfoton to Aquatic Organisms

Table 4.1 summarizes the most sensitive aquatic toxicity endpoints used for this assessment, based on an evaluation of both the submitted studies and the open literature, as previously discussed. A brief summary of submitted and open literature data considered relevant to this ecological risk assessment for the CRLF is presented below. Additional discussion of the data was presented in the Interim Reregistration Eligibility Decision (IRED, 2002), which may be referenced for additional information and may be found at the following url:

http://www.epa.gov/pesticides/reregistration/REDs/disulfoton_ired.pdf

Table 4.1 Freshwater Aquatic Toxicity Profile for Disulfoton

Assessment Endpoint	Test Chemical	Species	Toxicity Value Used in Risk Assessment	Citation MRID # (Author & Date)	Comment
Acute Direct Toxicity to Aquatic-Phase CRLF	Disulfoton	Bluegill	39 ppb	MRID 00068268	Very highly toxic.
	Sulfone	Bluegill	112 ppb	MRID 42585108	Highly toxic
	Sulfoxide	Bluegill	188 ppb	MRID 42585107	Highly toxic
Chronic Direct Toxicity to Aquatic-Phase CRLF	Disulfoton (degradates not tested)	Bluegill	4 ppb	MRID 41935801	Value extrapolated using acute to chronic ratio derived from other fish species and applied to the acute bluegill LC50 (IRED: U.S. EPA, 2002).
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Freshwater Invertebrates (i.e. prey items)	Disulfoton	*Glass shrimp*	3.9 ppb	MRID 40094602	Very highly toxic.
	Sulfone degradate	*Daphnid*	35 ppb	MRID 42585112	Very highly toxic
	Sulfoxide degradate	*Daphnid*	64 ppb	MRID 42585109	Very highly toxic
Indirect Toxicity to Aquatic-Phase CRLF via Chronic Toxicity to Freshwater Invertebrates (i.e. prey items)	Disulfoton	*Glass shrimp*	0.01 ppb	MRID 41935802	Value extrapolated using acute to chronic ratio derived from daphnids and applied to the acute glass shrimp LC50.
	Sulfone	Daphnids	0.14 ppb	MRID 43738001	--
	Sulfoxide	Daphnids	1.5 ppb	MRID 43738002	--
Indirect Toxicity to	Disulfoton	No data			

Aquatic-Phase CRLF via Acute Toxicity to Non-vascular Aquatic Plants	and degradates	
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Vascular Aquatic Plants	Disulfoton and degradates	No data

Toxicity to fish and aquatic invertebrates is categorized using the system shown in **Table 4.2** (U.S. EPA, 2004). Toxicity categories for aquatic plants have not been defined.

Table 4.2 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

4.1.1 Toxicity to Freshwater Fish

Given that no disulfoton 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 disulfoton to the CRLF. Effects to freshwater fish resulting from exposure to disulfoton may indirectly 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, including data from the open literature, is provided below in Sections 4.1.1.1 through 4.1.1.3.

4.1.1.1 Freshwater Fish: Acute Exposure (Mortality) Studies

The most sensitive acute freshwater LC50s are summarized in **Table 4.3**. Disulfoton is very highly toxic to fish on an acute exposure basis. The most sensitive available LC50 is 39 ug/L (MRID 00068268) in bluegill sunfish. The sulfone and sulfoxide degradates are less toxic than disulfoton to fish, but are highly toxic to fish. The bluegill LC50 for the sulfone and sulfoxide degradates are 112 ppb and 188 ppb, respectively.

Table 4.3. Acute Fish Toxicity Values for Disulfoton and Degradates

Freshwater Species	Results (ppb ai)	Toxicity Category	Source of Data MRID
Bluegill	LC50=39 **Probit slope:** 4.5 (2 – 9)[a]	very highly toxic	00068268
Bluegill	LC50 (sulfone metabolite)	highly toxic	42585108

	=112 **Probit slope:** 5.4 (3.4 – 7.5)		
Bluegill	LC50 (sulfoxide metabolite) =188 **Probit slope:** 4.5 (2 – 9)[a]	highly toxic	42585107

[a] 4.5 is the default slope with 2 and 9 representing reasonable lower and upper bounds (U.S. EPA, 2004).

Bluegill sunfish was the most sensitive species tested as shown in **Table 4.3**. LC50s for other species are summarized in **Table 4.4**.

Table 4.4. Range of Acute Fish Toxicity Values for Disulfoton

Freshwater Species	Results (ppb ai)	Toxicity Category	Source of Data (MRID)
Bluegill	39 – 300	Highly to very highly toxic	40098001, 0068268
Rainbow trout	1850 to 3000	Moderately toxic	40098001 and 68268
	60,000 (sulfoxide) Probit slope: 11 (6.4 – 16)	Slightly toxic	42585110
	>9,200 (sulfone)	Moderately toxic	42565111
Channel Catfish	4700	Moderately toxic	40098001
Goldfish	7200	Moderately toxic	229299
Largemouth Bass	60 - 120	Very highly toxic	0003503, 40098001,
Fathead minnow	59 - 4300	Very highly toxic	0003503
Guppy	280	Highly Toxic	229299

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

Available early life stage toxicity studies are summarized in Table 4.5. The NOAEC in rainbow trout was 220 ppb, which is approximately 8.4 fold lower than the most sensitive acute LC50. Rainbow trout were considerably less sensitive to disulfoton than bluegill sunfish, and no chronic studies in bluegill have been submitted. The most sensitive NOAEC in rainbow trout from an early life stage study (MRID 41935801) was 220 ppb, which is considerably higher than the most sensitive acute LC50 reported in bluegill of 37 ppb. No chronic study in bluegill has been submitted or was located in the open literature. Therefore, an acute to chronic ratio was used to estimate a chronic NOAEC in bluegill using the following equation.

Bluegill NOAEC = Bluegill LC50 / (Trout LC50 / Trout NOAEC)
= 37 ppb / (1850 ppb / 220 ppb)
= 37 ppb / 8.4
= 4 ppb

Table 4.5. Freshwater Fish Early Life-Stage Toxicity

Species	NOAEC/LOAEC (ppb ai)	Endpoints Affected	MRID No. Author/Year	Comments
Rainbow trout *(Oncorhynchus mykiss)*	220/420	Growth	41935801 1991	Acceptable study.
Bluegill	4 ppb	N/A	N/A	NOAEC in bluegill estimated using acute to chronic ratio based on rainbow trout data.

4.1.1.3 Freshwater Fish: Sublethal Effects and Additional Open Literature Information

In the available submitted acute toxicity studies, sublethal effects were not observed at levels that did not also induce mortality. In the submitted chronic studies, sublethal effects were not observed at levels below the NOAEC.

In the open literature, Arnold et al. (1996) reported cytologic effects in the liver at levels as low as 0.1 ug/L, which is below the acute and chronic toxicity values used in this assessment. These effects were not chosen for use in this effects determination because they could not be directly linked to the assessment endpoints of survival, growth, and reproduction. No other sublethal effects were reported at levels lower than the NOAEC used to calculate RQs for this assessment in the open literature.

4.1.1.4 Aquatic-phase Amphibian: Acute and Chronic Studies

No useful studies in amphibians were located in the open literature or were submitted to the Agency.

4.1.2 Toxicity to Freshwater Invertebrates

Freshwater aquatic invertebrate toxicity data were used to assess potential indirect effects of disulfoton to the CRLF. Effects to freshwater invertebrates resulting from exposure to disulfoton may 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 sowbugs, larval alderflies, and water striders.

A summary of acute and chronic freshwater invertebrate data, including data published in the open literature, is provided below in Sections 4.1.2.1 through 4.1.2.3.

4.1.2.1 Freshwater Invertebrates: Acute Exposure Studies

The available data indicates that disulfoton and its degradates of concern are very highly toxic to aquatic invertebrates. Results of the available freshwater invertebrate toxicity data are summarized in **Table 4.6**.

Table 4.6. Acute Aquatic Invertebrate Toxicity Data for Disulfoton and its Major Degradates.

Freshwater Species	Test Material	Results (ppb ai)	Toxicity Category	Source of Data
Daphnia	Disulfoton	13 **Probit slope**: reliable slope not available	Very highly toxic	MRID 00143401
	Sulfone metabolite	35 **Probit slope**: 3.5 (2.3 - 4.7)	Very highly toxic	MRID 42585112
	Sulfoxide metabolite	64 **Probit slope**: 4.6 (3.1 – 6.1)	Very highly toxic	MRID 42585109
Scud	Disulfoton	27 to 52	Very highly toxic	MRID 05017538; 40098001
Glass shrimp	Disulfoton	3.9	Very highly toxic	MRID 40094602
Stonefly	Disulfoton	5 to <8.2	Very highly toxic	MRID 229299, 40098001

4.1.2.2 Freshwater Invertebrates: Chronic Exposure Studies

Freshwater invertebrate life-cycle tests are summarized in **Table 4.7**. The most sensitive NOAEC in daphnids was 0.037 ug/L for disulfoton. The available NOAECs for the sulfone and sulfoxide degradates were 0.14 and 1.5 ug/L, respectively. The most sensitive species in acute studies was the glass shrimp. No chronic studies in glass shrimp were available. Therefore, an acute to chronic ratio was used to estimate a NOAEC for glass shrimp using the following equation:

Glass shrimp NOAEC = Glass shrimp LC50 / (Daphnid EC50 / Daphnid NOAEC)
= 3.9 ppb / (13 ppb / 0.037 ppb)
= 3.9 ppb / 351
= 0.01 ppb

Table 4.7. Freshwater Aquatic Invertebrate Life-Cycle Toxicity

Species	NOAEC/ LOAEC (ppb)	Endpoints Affected	MRID No. Author/Year	Study Classification
Waterflea *(Daphnia magna)*	Disulfoton: 0.037/0.070	survival, length, and # young/adult	41935802 Blakemore/1991	core
Waterflea *(Daphnia magna)*	Sulfone degradate: 0.14/0.27	length	43738001 Bowers/1995	core
Waterflea *(Daphnia magna)*	Sulfoxide degradate: 1.53/2.97	Weight & length	43738002 Bowers/1995	core
Glass shrimp	0.01	N/A – Estimated value		

4.1.2.3 Freshwater Invertebrates: Open Literature Data

No studies were located in the open literature that reported toxicity values that were more sensitive than studies used to calculate RQs in this assessment.

4.1.3 Toxicity to Aquatic Plants

Aquatic plant toxicity studies may be used as one of the measures of effect to evaluate whether disulfoton 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.

Two types of studies may be used to evaluate the potential of disulfoton to affect aquatic plants. Laboratory and field studies were used to determine whether disulfoton may cause direct effects to aquatic plants. However, no freshwater aquatic plant studies have been submitted to the Agency or were located in the open literature. Therefore, the potential toxicity of disulfoton to aquatic plants was not quantified.

4.2 Toxicity of Disulfoton to Terrestrial Organisms

Table 4.8 summarizes the most sensitive terrestrial toxicity endpoints used to assess potential risks to the CRLF based on an evaluation of both the 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. Additional information is presented in Sections 4.2.1. to 4.2.4.

Table 4.8 Terrestrial Toxicity Profile for Disulfoton and its Degradates of Concern

Endpoint	Test Material	Species	Toxicity Value Used in Risk Assessment	Citation MRID# (Author & Date)	Comment
Acute Direct Effects to Terrestrial-Phase CRLF (LD50)	Disulfoton	Mallard Duck	LD50: 6.5 mg/kg-bw	MRID 00160000	--
	Sulfone Degradate	Bobwhite Quail	LD50 = 18 mg/kg-bw Probit slope: not calculated	42585103	--
	Sulfoxide Degradate	Bobwhite Quail	LD50 = 9.2 mg/kg-bw Probit slope: 6.2 (2.8 – 9.5	42585102	--
Acute Direct Toxicity to Terrestrial-Phase CRLF (LC_{50})	Disulfoton	Japanese quail	LC50: 333 ppm	0034769	LC50 in bobwhite quail was 544 mg/kg-diet (MRID 0094233).
	Sulfone Degradate	Bobwhite quail	LC50: 558 ppm	42585106	Data in other species have not been submitted.
	Sulfoxide Degradate	Bobwhite quail	LC50: 456 ppm	42585105	Data in other species have not been submitted.
Chronic Direct Effects to Terrestrial-Phase CRLF	Disulfoton	Mallard duck	NOAEC=37 LOAEC=80 (decreased adult and hatchling body weight)	43032502	--
Indirect Effects to Terrestrial-Phase CRLF (via acute toxicity to mammalian prey items)	Disulfoton	Laboratory Rat	LD50=1.9 mg ai/kg	072293	--
	Sulfone Degradate	Laboratory Rat	LD50 (sulfone metabolite) =11.24 mg /kg	0071873	--
Indirect Effects to Terrestrial-Phase CRLF (via chronic toxicity to mammalian prey items)	Disulfoton	Laboratory Rat	NOAEL=0.04 mg/gk-bw LOAEL= 1.2 mg/kg-bw (decreased litter size and pup survival)	00157511	--
Indirect Effects to Terrestrial-Phase CRLF (via acute	Disulfoton	Honey bee	LD50: 4.1 ug ai/bee	05004151	--
	Sulfone			42582902	--

Endpoint	Test Material	Species	Toxicity Value Used in Risk Assessment	Citation MRID# (Author & Date)	Comment
toxicity to terrestrial invertebrate prey items)	metabolite	Honey bee	LD50: 0.96 ug/bee		
	Sulfoxide metabolite	Honey bee	LD50: 1.1 ug /bee	42582901	--
Indirect Effects to Terrestrial- and Aquatic-Phase CRLF (via toxicity to terrestrial plants)	Disulfoton	Seedling Emergence Monocots	EC25: >1.9 lbs a.i./Acre	46526601	--
		Seedling Emergence Dicots	EC25: >1.9 lbs a.i./Acre	46526601	--
		Vegetative Vigor Monocots	EC25: >2.4 lbs a.i./Acre	46526602	--
		Vegetative Vigor Dicots	EC25: >2.4 lbs a.i./Acre	46526602	--

Acute toxicity to terrestrial animals is categorized using the classification system shown in **Table 4.9** (U.S. EPA, 2004). Toxicity categories for terrestrial plants have not been defined.

Table 4.9 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

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 data are not available (U.S. EPA, 2004). No terrestrial-phase amphibian data are available for disulfoton; therefore, acute and chronic avian toxicity data are used to assess the potential direct effects of disulfoton to terrestrial-phase CRLFs.

4.2.1.1 Birds: Acute Exposure (Mortality) Studies

Available acute oral and subacute dietary studies are summarized in **Table 4.10** below. Disulfoton is very highly toxic to birds on an acute oral basis and moderately toxic on a subacute dietary basis. In addition, the sulfone and sulfoxide degradates were shown to be approximately as toxic to birds as disulfoton on a subacute dietary basis and more toxic than disulfoton on an acute oral basis.

Table 4.10. Toxicity Endpoints Used to Estimate Potential Risk of Direct Effects to Terrestrial Phase CRLFs

Species	Test Type / chemical	Results (ppm ai)	Toxicity Classification	Source of Data
Northern bobwhite quail	Subacute dietary Disulfoton	LC50 = 544	moderately toxic	0094233
	Subacute dietary Sulfone degradate	LC50 = 558 Probit slope: 5.4 (2.8 – 7.9)	moderately toxic	42585106
	Sub acute dietary Sulfoxide degradate	LC50 = 456 mg/kg Probit slope: 3.0 (1.7 – 4.3)	highly toxic	42585105
Mallard Duck	Subacute dietary Sulfoxide degradate	LC50 = 823 ppm Probit slope 6.2 (2.6 – 9.7)	moderately toxic	42585104
	Subacute dietary Sulfone degradate	LC50 = 622 ppm Probit slope: 5.8 (2.8 – 8.9)	moderately toxic	42585101
Japanese quail	Subacute dietary Disulfoton	LC50=333	highly toxic	0034769
Mallard duck	Acute oral Disulfoton	LD50=6.54 mg ai/kg	very highly toxic	00160000
Bobwhite quail	Acute oral Disulfoton	LD50: 39 mg/gk-bw Probit slope: 4.8 (0.9 – 8.6)	highly toxic	42585803
	Acute oral Sulfoxide degradate	LD50 = 9.2 mg/kg-bw Probit slope: 6.2 (2.8 – 9.5)	very highly toxic	42585102
	Acute oral Sulfone degradate	LD50 = 18 mg/kg-bw Probit slope: not calculated	highly toxic	42585103

4.2.1.2 Birds: Chronic Exposure (Growth, Reproduction) Studies

Available reproduction toxicity studies in birds are summarized in **Table 4.11** below. Data were not submitted on the degradates of concern. The NOAEC in both mallard ducks and bobwhite quail was 37 ppm based on reduced body weight.

Table 4.11. Summary of Available Avian Reproduction Toxicity Studies for Disulfoton.

Species	Endpoint Tested	Results	Source of Data
Mallard duck	reproduction	NOAEC=37 mg/kg-diet LOAEC=80 mg/kg-diet (decreased adult and hatchling body weight)	43032502
Bobwhite quail	reproduction	NOAEC=37 mg/kg-diet LOAEC=74 mg/kg-diet (decreased adult body	43032501

		weight)	

4.2.2 Toxicity to Mammals

Mammalian toxicity data are used to assess potential indirect effects of disulfoton to the terrestrial-phase CRLF. Effects to small mammals resulting from exposure to disulfoton may indirectly 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).

4.2.2.1 Mammals: Acute Exposure (Mortality) Studies

Available acute oral toxicity studies are summarized in **Table 4.12** below. Disulfoton is very highly toxic to mammals on an acute oral basis. In addition, the sulfone metabolite is also highly toxic to mammals on an acute oral basis, although it was approximately 10-fold less toxic than disulfoton to mammals on an acute basis. Data on the sulfoxide degradate have not been submitted.

Table 4.12. Summary of Available Mammalian Acute Toxicity Studies for Disulfoton and its Degradates of Concern

Test Species	Study Type	Toxicity Value	Toxicity Category	MRID
Laboratory rat	acute oral	LD50=1.9 mg ai/kg Adj. LD50: 4.2 mg/kg-bw	very highly toxic	072293
Laboratory rat sulfone metabolite	acute oral	LD50 =11.24 mg /kg Adj. LD50: 25 mg/kg-bw	highly toxic	0071873

4.2.2.2 Mammals: Chronic Exposure (Growth, Reproduction) Studies

Available reproduction toxicity studies in mammals are summarized in **Table 4.13** below. Disulfoton affected reproductive success (defined as decreased litter size and pup survival) at 2.4 mg/kg-bw with a NOAEC of 0.8 mg/kg-bw. Neither the sulfone nor the sulfoxide degradate have been tested for reproductive effects to mammals.

Table 4.13. Summary of Available Mammalian Reproduction Toxicity Studies for Disulfoton

Test Species	Study Type	Toxicity Value	Toxicity Category	MRID
Laboratory rat	2-generation reproduction	NOAEL=0.04 mg/gk-bw LOAEL= 1.2 mg/kg-bw (decreased litter size and pup survival)	N/A	00157511

4.2.3 Toxicity to Terrestrial Invertebrates

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

4.2.3.1 Terrestrial Invertebrates: Acute Exposure (Mortality) Studies

Submitted acute exposure studies in terrestrial invertebrates are summarized in **Table 4.14**. A number of studies have also been conducted that evaluated the efficacy of disulfoton with respect to insecticidal activity. Although these studies evaluated effects to terrestrial invertebrates, the study designs do not allow for an estimate of a dose or application rate associated with a toxicity value that can be used in risk assessment.

Table 4.14. Summary of Available Mammalian Acute Toxicity Studies for Disulfoton

Test Species	Test Type	Toxicity Value	Reference (MRID) / Comment
Honey bee	acute contact	LD50: 4.1 ug ai/bee	05004151
Honey bee	acute contact	LD50 (sulfone metabolite): 0.96 ug/bee	42582902
Honey bee	acute contact	LD50 (sulfoxide metabolite): 1.11 ug /bee	42582901
Honey bee	acute foliar residue	RT25 (8 EC) < 3hrs at 1.0 lb ai/A	0163423 / RT 25 is the residual time required to reduce mortality of caged bees to field weathered spray deposits.

4.2.4 Toxicity to Terrestrial Plants

Terrestrial plant toxicity data are used to evaluate the potential for disulfoton to affect riparian zone and upland vegetation within the action area for the CRLF. Impacts to riparian and upland (i.e., grassland, woodland) vegetation may 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 disulfoton, 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.

A Tier I seedling emergence test and a Tier II vegetative vigor test was submitted. The EC25 for seedling emergence and vegetative vigor was >1.9 lbs a.i./Acre and >2.4 lbs a.i./Acre, respectively.

4.3 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 aquatic animals that may indirectly 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 disulfoton 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. Results of this analysis are presented in the Risk Description (Section 5.2).

4.4 Incident Database Review

A review of the EIIS database for ecological incidents involving disulfoton was completed on March 10, 2008. Several reports of wildlife poisonings are associated with disulfoton. These poisoning incidents are summarized in **Table 4.15** below. Some of these incident reports support EPA's concerns for acute risk. In particular, one incident reported that birds consuming insects that fed on plants treated with disulfoton were killed. This incident further emphasizes the potential importance of an exposure pathway that is not quantified in this assessment. Consumption of insects that have consumed plant material contaminated with disulfoton via systemic uptake and translocation in the plant resulted in effects to higher trophic level organisms that fed on such insects.

Table 4.15. Chronological List of Ecological Incidents

Start Date	Misuse? (yes/no/unknown)	Incident Description
6/12/95	unknown	Johnston County, NC: Fish kill occurred in commercial fish pond. Crop fields nearby treated with pesticides. Water, soil and vegetation samples analyzed for a variety of pesticides. Disulfoton, as well as several other pesticides, was found at 0.2-2.5 ppm in vegetation samples. Possible certainty index for disulfoton. (Incident Report No. I003826-002).
1/24/94	unknown	Puerto Rico: 6 grackles fell dead from tree in yard of private residence. Dead heron and owl also found in vicinity. Use site and method not reported. Birds had depressed acetyl cholinesterase. Analysis of GI contents of a grackles showed disulfoton at 2.37 ppm wet weight. Highly probable certainty index for disulfoton. (Incident Report No. I003966-004).
6/11/94	unknown	Arapahoe CO: Fish kill following application of Di-Syston EC. to wheat just before heavy rain. Water samples contained disulfoton sulfoxide at 29.5-48.7 ppb and disulfoton sulfone at 0.0199-0.214 ppb. (Incident Report No. I001167-001).
6/18/93	No	Young County,TX: 18 Swainson's hawks dead, 1severely disabled in a cotton field. Cotton seed had been treated with disulfoton prior to planting, ~10 days before the birds were discovered. No additional applications of OP or carbamate pesticides made in vicinity of field. Autopsies showed no trauma or disease. Lab analysis showed insect material in GI tracts; this material contained disulfoton (~7 ppm); no other OP or carbamate insecticides were present. Hawks fed on insects, which had been feeding on the young cotton plants, which contained disulfoton residues. (L.Lyon, Div. of Environmental Contaminants, U.S. Fish and Wildlife Service, Arlington, VA.)
6/22/91	unknown	Onslow County, NC: Fish kill in pond at private residence. Pond received runoff from neighboring tobacco field; pondwater analysis showed disulfoton and several other pesticides, including endosulfan. Disulfoton sulfoxide found in water at 0.32 ppb. Endosulfan had highest concentration (1.2 µg/L), and is toxic to fish, but disulfoton cannot be ruled out as a possible cause of death. No tissue analysis. Possible certainty index for disulfoton. (Incident Report No. B0000216-025).
4/26/91	unknown	Sussex County, DE: 9 American robins dead following application of granular disulfoton at tree nursery. Corn and soybeans also in vicinity. No laboratory analysis. Probable certainty index for disulfoton. (Incident Report No. I000116-003).

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 disulfoton 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 (U.S. EPA, 2004). 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 disulfoton usage scenarios summarized in **Section 3** and the appropriate aquatic toxicity endpoint reported in Section 4. 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 disulfoton (Section 3) and the appropriate toxicity endpoint from Section 4.

5.1.1 Exposures in the Aquatic Habitat

5.1.1.1 Direct Effects to Aquatic-Phase CRLF

RQs used to estimate potential risks to aquatic phase CRLFs are summarized in **Table 5.1**. Direct effects to the aquatic-phase CRLF are based on peak EECs 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. RQs exceeded the LOC for either acute or chronic effects for all uses. Therefore, disulfoton may directly affect the CRLF. Additional analysis on the potential for disulfoton to adversely affect the CRLF is in Section 5.2.

Table 5.1 Summary of Direct Effect RQs for Aquatic-phase CRLFs Based on an LC50 of 37 ug/L and a NOAEC of 4 ug/L in Fish

Use	Peak EEC (μg/L)	Acute RQ (LC50: 37 ug/L)	Probability of Individual Effect (acute effects only)	60-Day EEC (μg/L)	RQ (NOAEC: 4 ug/L)	LOC Exceedance and Risk Interpretation
Beans, Broccoli, Cauliflower	1.8 – 3.6	0.05 – 0.097	1 in 4E8 to 1 in 4E5	1.3 – 2.8	0.33 – 0.70	The endangered species acute LOC is exceeded.
Residential, Cotton, Brussels sprouts, Christmas trees, lettuce	3.7 - 15	0.10 – 0.41	1 in 3E5 to 1 in 25	3.2 - 12	0.8 (residential), 0.88 (cotton), 1.1 – 3 (other uses)	Restricted use LOC is exceeded for these uses, and the chronic LOC is exceeded for Brussels sprouts, Christmas trees, and lettuce.
Asparagus, cabbage	23 - 24	0.62 – 0.65	1 in 6 to 1 in 5	18 - 22	3.5 – 8.8	Acute and chronic LOCs are exceeded for these uses.
Lettuce (drip irrigation)	67	1.8	1 in 1.1	54	14	Acute and chronic LOC exceeded

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

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. A summary of the acute and chronic RQ values for exposure to aquatic invertebrates (as prey items of aquatic-phase CRLFs) is provided in **Table 5.2**. RQs exceeded the LOC for acute and chronic effects for all uses. Therefore, disulfoton may affect the CRLF. Additional analysis on the potential for disulfoton to adversely affect the CRLF by reducing aquatic invertebrate prey base is in Section 5.2.

Table 5.2 Summary of Acute and Chronic RQs Used to Estimate Indirect Effects to the CRLF via Effects on Aquatic Invertebrates as Dietary Food Items (prey of CRLF juveniles and adults in aquatic habitats) Based on an LC50 of 3.9 ppb and a NOAEC of 0.011 ppb

Use	Peak EEC (μg/L)	Acute RQ	Probability of Individual Effect (acute effects only)	21-Day EEC (μg/L)	Chronic RQ	LOC Exceedance and Risk Interpretation
All uses	1.8 - 67	0.46 to 17	1 in 16 to 1 in 1	1.6 - 23	145 – 5600	Acute and chronic LOCs were exceeded for all uses.

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. RQs exceeded the LOC for acute and chronic effects for most uses. Therefore, disulfoton may affect the CRLF. Additional analysis on the potential for disulfoton to adversely affect the CRLF is in Section 5.2.

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

No aquatic plant toxicity data are available for derivation of RQs. The effects determination for potential indirect effects to the CRLF by affecting aquatic plants is presented in Section 5.2.

5.1.2 Exposures in the Terrestrial Habitat

5.1.2.1 Direct Effects to Terrestrial-phase CRLF

As previously discussed in Section 3.3, potential direct effects to terrestrial-phase CRLFs are based on spray applications of disulfoton either to soil or foliage. 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 and acute oral and subacute dietary toxicity endpoints for avian species. RQs used to estimate potential risks to terrestrial phase CRLFs are in **Tables 5.3a and 5.3.b**. Based on exceedance of the acute and reproduction EECs, a preliminary “may effect” determination is made. Additional analysis and the effects determination is presented in Section 5.2.

Table 5.3a. Avian RQs Used To Estimate Potential Risk of Direct Effects to Terrestrial Phase CRLFs for Spray Applications[a]

Use	Assessed Effect and Species	Chemical	EEC[a]	Toxicity Value	RQ
Asparagus (1 lbs a.i./Acre, 2 apps, 14-day invert)	Direct Acute Effect (dose)	Disulfoton	270	**LD50:** 6.5 mg/kg-bw **Adj LD50:** 3.4 mg/kg-bw	80
		Sulfoxide Degradate	217	**LD50:** 9.2 mg/kg-bw **LD50Adj:** 6.6 mg/kg-bw	33
		Sulfone Degradate	216	**LD50:** 18 mg/kg-bw **LD50Adj:** 13 mg/kg-bw	16
	Direct Acute Effect (dietary)	Disulfoton	240	**LC50:** 330 ppm	0.72
		Sulfone Degradate	192	**LC50:** 558 ppm	0.42
		Sulfoxide Degradate	240	**LC50**: 456 ppm	0.43
Cabbage and cotton (1 lb a.i./Acre, single application) (1 lb a.i./Acre, single application)	Direct Acute Effect (dose)	Disulfoton	150	**LD50:** 6.5 mg/kg-bw **Adj LD50:** 3.4 mg/kg-bw	44
		Sulfoxide Degradate	150	**LD50:** 9.2 mg/kg-bw **LD50Adj:** 6.6 mg/kg-bw	23
		Sulfone Degradate	120	**LD50:** 18 mg/kg-bw **LD50Adj:** 13 mg/kg-bw	6.7
	Direct Acute Effect (dietary)	Disulfoton	140	**LC50:** 330 ppm	0.4
		Sulfone Degradate	110	**LC50:** 558 ppm	0.20
		Sulfoxide Degradate	140	**LC50**: 456 ppm	0.31

[a] EECs for the sulfoxide and sulfone degradates were estimated assuming a 95% and 72% formation rate from parent, respectively (Section 2). RQs are based on small insect EECs

Table 5.3b. Avian LD50/Square Foot Analysis Used to Estimate Potential Direct Effects to the CRLF from Granular and Soil Incorporated Applications[a]

Use	Application Rate **(lbs a.i./Acre)**	**Application Method (% incorporated)**	**EEC (mg a.i./f^{t2})**	**LD50/ft^2**
Beans, broccoli, Brussels sprouts, cabbage, cauliflower, cotton	1	Soil incorporated, spray or injection (99%)	0.33	3.1
Christmas trees	4.5	Granular broadcast, wetted in (85%)	7.0	104
Cabbage and Lettuce	2	Soil injection (99%)	0.42	6.2
Residential	1.6	Granular broadcast, wetted in (85%)	2.5	37

[a] LD50 per square foot analysis does not include exposures from consumption of contaminated plants that have taken up and translocated the chemical throughout the plant. However, risk to CRLFs that consume insects that have fed on treated foliage presumably exceed LOCs based on the incident data.

Potential direct reproduction effects from exposure to disulfoton to the terrestrial-phase CRLF are derived by considering dietary-based exposures modeled in T-REX for a small bird (20g) consuming small invertebrates. Reproduction effects are estimated using the lowest available toxicity data for birds. EECs are divided by toxicity values to estimate chronic dietary-based RQs. RQs used to estimate potential direct reproduction effects are summarized in **Table 5.4**. RQs were only estimated for chronic exposures for spray applications. Although potential risks to reproduction of CRLFs and their prey from soil injection and in-furrow applications was not quantified, risks may be above concern levels for these types of applications as discussed in Section 5.2 (Risk Description).

Table 5.4. Reproduction RQs for Birds Used to Estimate Potential Direct Effects to CRLFs from Spray Uses[a]

Use	Assessed Effect and Species	Chemical	Toxicity Value	EEC (ppm)	RQ
Asparagus 2 applications of 1 lb a.i./acre, 14-day interval	Direct Reproduction Effect	Disulfoton	NOAEC: 37 ppm	240	6.4
Other uses Single application of 1 lb a.i./Acre				211	5.7

a RQs are based on small insect EECs

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

5.1.2.2.1 Terrestrial Invertebrates

In order to assess the potential risks of disulfoton to terrestrial invertebrates, which are considered prey of CRLF in terrestrial habitats, 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 4 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 31 µg a.i./g of bee. The resulting RQs were 7.7 for asparagus and 4.4 for other uses. Based on LOC exceedances for the surrogate terrestrial invertebrate, a preliminary "may effect" determination was made. Additional analysis is presented in Section 5.2.

Table 5.5. 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 EEC	Small Insect RQ	Large Insect EEC	Large Insect RQ
Asparagus	240	7.7	26	0.84
All other uses	135	4.4	15	0.48

5.1.2.2.2 Mammals

Risk quotients used to evaluate potential indirect effects resulting from impacts to mammalian prey are presented in **Table 5.6**. RQs were 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 reproduction dose-based RQs as well as reproduction dietary-based RQs.

Table 5.6a. RQs used to Estimate Potential Acute Risks to Mammalian Prey of CRLFs From Spray Applications

Use	Chemical	EEC (mg/kg-bw)	Toxicity Value (mg/kg-bw)	RQ
Asparagus 2 applications of 1 lb a.i./acre, 14-day interval	Disulfoton	400	LD50: 1.9 Adj. LD50: 4.2	96
	Sulfone Degradate	320	LD50: 11 Adj. LD50: 25	13
	Sulfoxide Degradate	Not calculated due to lack of toxicity data		
Other uses Single application of 1 lb a.i./Acre	Disulfoton	229	LD50: 1.9 Adj. LD50: 4.2	55
	Sulfone Degradate	180	LD50: 11 Adj. LD50: 25	7.3
	Sulfoxide Degradate	Not calculated due to lack of toxicity data		

Table 5.6b. LD50/Square Foot Analysis Used to Estimate Potential Effects to Mammal Prey Items of the CRLF (soil incorporated applications)[a]

Use	Application Rate **(lbs a.i./Acre)**	**Application Method (% incorporated)**	**EEC (**mg a.i./f^{t2}**)**	**LD50/ft²**
Beans, broccoli, Brussels sprouts, cabbage, cauliflower, cotton	1	Soil incorporated, spray or injection (99%)	0.33	3.3
Christmas trees	4.5	Granular broadcast, wetted in (85%)	7.0	104
Cabbage and Lettuce	2	Soil injection (99%)	0.42	6.6
Residential	1.6	Granular broadcast, wetted in (85%)	2.5	37

a LD50 per square foot analysis does not specifically evaluate exposures from consumption of plants that have taken up the material through the roots and translocated the material throughout the plant. However, risk to CRLFs that consume insects that have fed on treated foliage presumably exceed LOCs based on the incident data.

Table 5.7. Summary of Reproduction RQs used to Estimate Potential Risk to Mammalian Prey of CRLFs from Spray Applications of Disulfoton

Use	EEC	Toxicity Value	RQ
Asparagus 2 applications of 1 lb a.i./acre, 14-day interval	400 mg/kg-bw	AdjNOAEL: 0.09 mg/kg-bw	4600
	420 mg/kg-diet	NOAEC: 0.8 mg/kg-diet	530
Other uses Single application of 1 lb a.i./Acre	228 mg/kg-bw	AdjNOAEL: 0.09 mg/kg-bw	2600
	240 ppm	NOAEC: 0.8 ppm	300

5.1.2.2.3 Terrestrial Amphibians

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.

No amphibian toxicity data were located that evaluated potential effects from exposure to disulfoton or its degradates of concern. Avian RQs used to evaluate potential effects to amphibians were summarized in **Table 5.3**. Acute and reproduction RQs exceeded LOCs for birds. Because birds serve as a surrogate for terrestrial phase amphibians and reptiles, LOC exceedances for birds suggest that amphibian prey could be impacted. Additional analysis of the potential impacts to terrestrial amphibians as they relate to the effects determination is in Section 5.2 (Risk Description).

5.1.2.3 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 were not quantified for any use except Christmas trees because the EC25 was higher than the highest labeled application rate for all uses except Christmas trees. Non-endangered terrestrial plant RQs were <0.5 for Christmas trees, which is below the terrestrial plant LOC that is used for indirect effects determinations. The effects determination is presented in Section 5.2.

5.1.3 Primary Constituent Elements of Designated Critical Habitat

For disulfoton use, the assessment endpoints for designated critical habitat PCEs involve a reduction and/or modification of food sources necessary for normal growth and viability of aquatic-phase CRLFs, and/or a reduction and/or modification of food sources for terrestrial-phase juveniles and adults. Because these endpoints are also being assessed relative to the potential for indirect effects to aquatic- and terrestrial-phase CRLF, the effects determinations for indirect effects from the potential loss of food items are used as the basis of the effects determination for potential modification to designated critical habitat.

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 disulfoton's use within the action area. However, if direct or indirect effect LOCs are exceeded or effects may modify the

PCEs of the CRLF's critical habitat, the Agency concludes a preliminary "may affect" determination for the FIFRA regulatory action regarding disulfoton. A summary of the results of the risk estimation (*i.e.*, "no effect" or "may affect" finding) is provided in **Table 5.8** for direct and indirect effects to the CRLF.

Table 5.8. Preliminary Effects Determination Summary for disulfoton - Direct and Indirect Effects to CRLF

Assessment Endpoint	Preliminary Effects Determination	Basis For Preliminary Determination
Aquatic Phase (eggs, larvae, tadpoles, juveniles, and adults)		
Survival, growth, and reproduction of CRLF individuals via direct effects on aquatic phases	May affect	Acute LOC exceedance for all uses. Chronic RQs exceeded the LOC for all uses except beans, broccoli, cotton, residential, and cauliflower.
Survival, growth, and reproduction of CRLF individuals via effects to food supply (*i.e.*, freshwater invertebrates, non-vascular plants)	May affect	Acute and chronic LOCs were exceeded for all uses. Acute RQs were 0.5 to 12 and chronic RQs were 150 to 3800.
Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat, cover, and/or primary productivity (*i.e.*, aquatic plant community)	No effect	No LOC exceedance for terrestrial or aquatic plants
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.	No effect	No LOC exceedance for terrestrial or aquatic plants
Terrestrial Phase (Juveniles and adults)		
Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	May affect	LOCs were exceeded for disulfoton and two degradates of concern.
Survival, growth, and reproduction of CRLF individuals via effects on prey (*i.e.*, terrestrial invertebrates, small terrestrial mammals and terrestrial phase amphibians)	May affect	LOCs were exceeded for all taxonomic groups of prey items.
Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.*, riparian vegetation)	No effect	Potential risks to terrestrial plants were lower than the concern level.

For disulfoton use, the assessment endpoints for designated critical habitat PCEs involve a reduction and/or modification of food sources necessary for normal growth and viability of aquatic-phase CRLFs, and/or a reduction and/or modification of food sources for terrestrial-phase juveniles and adults. Because these endpoints are also being assessed relative to the potential for indirect effects to aquatic- and terrestrial-phase CRLF, the effects determinations for indirect effects from the potential loss of food items are used as the basis of the effects determination for potential modification to designated critical habitat. The following PCEs may be adversely impacted by disulfoton; other PCEs are related to potential adverse impacts to aquatic or terrestrial plants, which are not expected to be adversely impacted by labeled use of disulfoton to an extent that is expected to indirectly affect the CRLF.

- Alteration of chemical characteristics necessary for normal growth and viability of CRLFs and their food source.

- Reduction and/or modification of food sources for terrestrial phase juveniles and adults

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:

- <u>Significance of Effect</u>: 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.
- <u>Likelihood of the Effect Occurring</u>: Discountable effects are those that are extremely unlikely to occur.
- <u>Adverse Nature of Effect</u>: 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**.

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 disulfoton.

Acute RQs exceeded the endangered species LOC for all uses and ranged from 0.05 to 1.8. Based on the assumptions of a probit slope and a default slope of 4.5 (lower and upper bounds of 2 to 9) the probability of an individual mortality ranges from 1 in 4E8 (lowest RQ, beans) to approximately 1 in 1 (lettuce). Although the magnitude of some of the estimated probabilities of individual effects may be considered discountable, the slope of the dose-response curve is uncertain. The default slope of 4.5 was used for this analysis because slopes could not be obtained from the available studies. A more shallow or steep dose-response curve would result in a higher or lower estimated probability of an effect at RQs lower than 1. For example, use of lower and upper reasonable bounds for probit slopes of 2 to 9 (U.S. EPA, 2004) results in an estimated probability of an individual mortality of 1 in 26 to 1 in 1E15. Based on the exceedance of the endangered species LOC for all uses and uncertainty in the dose-response curve, it was concluded that disulfoton is likely to adversely affect the CRLF for all uses.

For the residential use, this conclusion of acute adverse effects to the CRLF is based on assumptions about application rates that are conservative and may lead to overestimates of potential exposure. The labels for residential uses do not define maximum application rates in terms of lb a.i/A and depend instead on the size of a garden or the number of plants treated. Extrapolation of these rates to a lb a.i./A value, as is necessary for the aquatic models used in this assessment, requires assumptions about the area that will be treated. The EEC of 3.7 ppb, which leads to an LOC exceedance with an RQ of 0.10, assumes that there are 4 lots per acre and that each one has a flower bed/garden of 2000 ft^2. If less area is treated, EECs would be lower and RQs may not exceed the LOC. With an endpoint of 37 ug/L, then any EEC less than 1.85 ug/L would not exceed the endangered species LOC. If actual application rates are half of those assumed, i.e. if the garden sizes are only 1000 ft^2 or if only 2 lots out of 4 have gardens treated with disulfoton, then the LOC would not be exceeded. Additionally, some of the residential labels define application rates by number of plants, rather than by area treated. At a labeled application rate of 0.0013 lb a.i./plant, 650 plants per acre could be treated without exceeding the LOC. Many typical residential applications, then, would not lead to LOC exceedances and conclusions of risk.

The chronic RQs also exceed the LOC of 1 for all uses except for beans, residential, and cotton. Chronic RQs for uses that did not exceed LOCs ranged from 0.33 (beans) to 0.88 (cotton). RQs for other uses exceeded the chronic LOC of 1.0 and ranged from 1.1 to 14 (lettuce).

Disulfoton is not expected to remain in the terrestrial environment for very long with a half life of several days. Therefore, after several days post application, runoff to aquatic systems will consist primarily of degradates. The EECs used in this assessment included total toxic residues (parent disulfoton and its degradates of concern). The most toxic degradate was the sulfone degradate, which is approximately 3-fold less toxic than disulfoton to fish. Therefore, assuming the toxicity of the residue in water is similar to the most toxic degradate tested, then RQs would be approximately 3-fold lower after several days post application. Therefore, assuming toxicity of the sulfone would still result in LOC exceedance for several uses.

The effects determination was based on the most sensitive species tested (bluegill). However, a number of fish species have been tested including rainbow trout, catfish, goldfish, largemouth bass, fathead minnows, and guppies. Some species have shown similar sensitivity to bluegill; however, other species tested have shown lower sensitivity (**Table 5.9**). RQs would remain above LOCs for several uses if the CRLF is as sensitive as largemouth bass, fathead minnows, guppies, or bluegill. However, RQs would be lower than the endangered species LOC of 0.05 for rainbow trout, catfish, and goldfish for all uses.

Table 5.9. Range of Acute Fish LC50s for Disulfoton.

Freshwater Species	Results (ppb ai)	Toxicity Category	Source of Data
Bluegill	39 – 300	Highly to very highly toxic	40098001, 0068268
Rainbow trout	1850 to 3000	Moderately toxic	MRIDs 40098001 and 68268
Channel Catfish	4700	Moderately toxic	40098001
Goldfish	7200	Moderately toxic	229299
Largemouth Bass	60 - 120	Very highly toxic	0003503, 40098001
Fathead minnow	59 - 4300	Very highly toxic	0003503
Guppy	280	Highly Toxic	229299

In addition, several incidents involving freshwater fish have been reported as summarized below:

6/12/95 Johnston County, NC: Fish kill occurred in commercial fish pond. Crop fields nearby treated with pesticides. Water, soil and vegetation samples analyzed for a variety of pesticides. Disulfoton, as well as several other pesticides, was found at 0.2-2.5 ppm in vegetation samples. Possible certainty index for disulfoton. (Incident Report No. I003826-002).

6/11/94 Arapahoe CO: Fish kill following application of Di-Syston EC. to wheat just before heavy rain. Water samples contained disulfoton sulfoxide at 29.5-48.7 ppb and disulfoton sulfone at 0.0199-0.214 ppb. (Incident Report No. I001167-001).

6/22/91 Onslow County, NC: Fish kill in pond at private residence. Pond received runoff from neighboring tobacco field; pondwater analysis showed disulfoton and several other pesticides, including endosulfan. Disulfoton sulfoxide found in water at 0.32 ppb. Endosulfan had highest concentration (1.2 µg/L), and is toxic to fish, but disulfoton cannot be ruled out as a possible cause of death. No tissue analysis. Possible certainty index for disulfoton. (Incident Report No. B0000216-025).

The incidences support the conclusion that freshwater fish (and aquatic phase amphibians as a surrogate) may be affected by labeled uses of disulfoton. Therefore, the RQ analysis together with the presence of several incidences that associated fish mortality with disulfoton use support the conclusion that the labeled uses of disulfoton are likely to adversely affect aquatic phase CRLFs.

5.2.1.2 Terrestrial-Phase CRLF, Direct Effects

Acute and chronic RQs exceeded the LOC for endangered birds for all uses. The highest dose-based acute RQ for disulfoton was 80 based on an adjusted LD50 of 3.4 mg/kg-bw and EEC derived assuming 2 applications of 1 lb a.i./Acre with a 14 day application interval. Assuming a single application of 1 lb a.i./Acre results in an RQ of 46 for disulfoton. The associated probability of an individual effect at an RQ of 46 or 80 approaches 100% for reasonable lower and upper bound probit slopes of 2 to 9 (U.S. EPA, 2004).

LD50s for the degradates are similar (within a factor of 3) to those of disulfoton, and EECs were similar to those of disulfoton. RQs for the sulfoxide and sulfone degradates also exceeded the endangered species LOC and the LD50 (RQ >1). The amount of degradate that may form in the environment is likely variable. This assessment assumed that the amount of degradate that formed was equivalent to the highest observed degradate level from the available degradation studies, which was 94% of parent for the sulfoxide (photolysis) and 72% of parent for the sulfone degradate (aerobic metabolism).

The RQ analysis was based on an evaluation of potential risks to birds. However, terrestrial amphibians are poikilotherms (body temperature varies with environmental temperature) while birds are homeotherms (temperature is regulated, constant, and largely independent of environmental temperatures). As a consequence, the caloric requirements of amphibians are markedly lower than birds. Therefore, on a daily dietary intake basis, birds consume more food than amphibians. This can be seen when comparing the caloric requirements for free living iguanid lizards to Passeriformes (song birds) (U.S. EPA, 1993):

iguanid FMR (kcal/day)= 0.0535 (bw g)^0.799

passerine FMR (kcal/day) = 2.123 (bw g)^0.749

With relatively comparable slopes to the allometric functions, one can see that, given a comparable body weight, the free living metabolic rate of birds can be 40 times higher than reptiles, though the requirement differences narrow with high body weights.

To quantify the potential differences in food intake and resulting potential differences in pesticide exposure between birds and terrestrial amphibians, RQs were calculated based on food intake estimates considered to be more representative of CRLFs. These results are in **Table 5.10**. Consideration of the different dietary behaviors of the CRLF compared with birds does not alter conclusions of this assessment. The highest acute RQ was 23 based on a single application of 1 lb a.i./Acre, and the acute RQ was exceeded for all food items except other terrestrial phase amphibians. Modeling of two applications would also result in RQs that exceed LOCs.

Table 5.10. Upper Bound Kenaga, Acute Terrestrial Herpetofauna Dose-Based Risk Quotients Based on a Single Application of 1 lb a.i./Acre

Size Class (grams)	Adjusted LD50	EECs and RQs									
		Broadleaf Plants/ Small Insects		Fruits/Pods/ Seeds/ Large Insects		Small Herbivore Mammals		Small Insectivore Mammal		Small Amphibians	
		EEC	RQ	EEC	RQ	EEC	RQ	EEC	RQ	EEC	RQ
1.4	6.50	5.24	**0.81**	0.58	0.09	N/A	N/A	N/A	N/A	N/A	N/A
37	6.50	5.15	**0.79**	0.57	0.09	N/A	N/A	N/A	N/A	0.18	0.03
238	6.50	3.38	**0.52**	0.38	0.06	23.26	**3.58**	1.45	**0.22**	0.12	0.02

In addition to LOC exceedances, several incidences have associated disulfoton exposure to bird mortality as summarized below. No incidences involving terrestrial amphibians have been reported.

One incident (L.Lyon, Div. of Environmental Contaminants, U.S. Fish and Wildlife Service, Arlington, VA) reported that birds consuming insects that fed on plants treated with disulfoton were killed. This incident further emphasizes the potential importance of an exposure pathway that is not quantified in this assessment. Consumption of insects that have consumed plant material contaminated with disulfoton via system uptake and translocation in the plant resulted in effects to higher trophic level organisms that fed on insects.

1/24/94 Puerto Rico: 6 grackles fell dead from tree in yard of private residence. Dead heron and owl also found in vicinity. Use site and method not reported. Birds had depressed acetyl cholinesterase. Analysis of GI contents of a grackles showed disulfoton at 2.37 ppm wet weight. Highly probable certainty index for disulfoton. (Incident Report No. I003966-004).

6/18/93 Young County,TX: 18 Swainson’s hawks dead, 1severely disabled in a cotton field. Cotton seed had been treated with disulfoton prior to planting, ~10 days before the birds were discovered. No additional applications of OP or carbamate pesticides made in vicinity of field. Autopsies showed no trauma or disease. Lab analysis showed insect material in GI tracts; this material contained disulfoton (~7 ppm); no other OP or carbamate insecticides were present. Hawks fed on insects, which had been feeding on the young cotton plants, which contained disulfoton residues. (L.Lyon, Div. of Environmental Contaminants, U.S. Fish and Wildlife Service, Arlington, VA.)

4/26/91 Sussex County, DE: 9 American robins dead following application of granular disulfoton at tree nursery. Corn and soybeans also in vicinity. No laboratory analysis. Probable certainty index for disulfoton. (Incident Report No. I000116-003).

The incidences support the conclusion that birds (and terrestrial phase amphibians as a surrogate) may be affected by labeled uses of disulfoton. Therefore, the RQ analysis together with the presence of several incidences that associated bird mortality with disulfoton exposure support the conclusion that the labeled uses of disulfoton are likely to adversely affect terrestrial phase CRLFs.

5.2.2 Indirect Effects (via Reductions in Prey Base), Aquatic Phase CRLFs

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. No to toxicity data are currently available for aquatic plants; therefore, EC50s cannot be derived for use in risk assessment. However, disulfoton is an insecticide with low toxicity to terrestrial plants. No terrestrial plant incidents have been reported for disulfoton with a certainty index of “probably” or higher. Therefore, there is no compelling evidence that aquatic or terrestrial plants will be impacted to a degree that would affect CRLFs by labeled uses.

5.2.2.2 Aquatic Invertebrates

The potential for disulfoton 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.

The acute RQs for aquatic invertebrates ranged from 0.5 – 17 based on the most sensitive species tested (glass shrimp). Therefore, the acute LOCs were exceeded for all uses. The associated estimated probability of an individual mortality at these RQs is greater than 1 in 10 based on a probit dose-response slope of 4.5 (default). The most sensitive species tested was the glass shrimp. Toxicity of disulfoton to other aquatic invertebrate species is in **Table 5.11**.

Table 5.11. Acute RQs for Various Aquatic Invertebrates for Disulfoton.

Freshwater Species	Test Material	Most Sensitive Toxicity Value (ppb ai)	RQ	Source of Data
Daphnia	Disulfoton	13	0.14 – 5.2	MRID 00143401
Scud	Disulfoton	27	0.07 – 2.5	MRID 05017538; 40098001
Glass shrimp	Disulfoton	3.9	0.46 - 17	MRID 40094602
Stonefly	Disulfoton	5	0.36 - 13	MRID 229299, 40098001

These data suggest that multiple aquatic invertebrate species may be affected by disulfoton exposure at levels estimated in this assessment.

Disulfoton degrades to more stable degradates somewhat rapidly in the environment with a half-life of approximately 3 days. Therefore, aquatic prey of the CRLF may also be exposed to disulfoton degradates. EC50s for the sulfone and sulfoxy degradates were approximately 3 and 5 times, respectively, less toxic to daphnids than disulfoton. However, it is uncertain if other invertebrate species are more or less sensitive to degradates because studies evaluating the toxicity of disulfoton degradates in species other than daphnids are not available.

Because the acute LOC of 0.5 is approached or exceeded for all uses based on the most sensitive aquatic invertebrate tested, and the potential magnitude of effects to aquatic invertebrate species tested could result in indirect effects to the CRLF, the effect determination for potential indirect effects to the CRLF via reduction in available food supply is "likely to adversely affect."

5.2.2.3 Fish and Aquatic-Phase Frogs

Potential risk to freshwater fish were described in Section 5.2.1 (direct effects). It was concluded that labeled disulfoton uses are likely to adversely affect CRLFs. However, this does not necessarily correlate with potential indirect effects to CRLFs that consume fish because a LAA determination for a direct effect is made on the individual level (might a single individual be affected?). The potential for indirect effects is evaluated based on the potential magnitude of effects to the food item. RQs based on the most sensitive species tested are in **Table 5.12**.

Table 5.12. Summary of Direct Effect RQs for Aquatic-phase CRLFs Based on an LC50 of 37 ug/L and a NOAEC of 4 ug/L in Fish

Use	Peak EEC (µg/L)	Acute RQ (LC50: 37 ug/L)	Probability of Individual Effect (acute effects only)	60-Day EEC (µg/L)	RQ (NOAEC: 4 ug/L)	LOC Exceedance and Risk Interpretation
Beans, Broccoli, Cauliflower	1.8 – 3.6	0.05 – 0.097	1 in 4E8 to 1 in 4E5	1.3 – 2.8	0.33 – 0.70	The endangered species acute LOC is exceeded.
Residential, Cotton, Brussels sprouts, Christmas trees, lettuce	3.7 - 15	**0.10 – 0.41**	1 in 3E5 to 1 in 25	3.2 - 12	0.8 (residential), 0.88 (cotton), 1.1 – 3 (other uses)	Restricted use LOC is exceeded for these uses, and the chronic LOC is exceeded for Brussels sprouts, Christmas trees, and lettuce.
Asparagus, cabbage	23 - 24	**0.62 – 0.65**	1 in 6 to 1 in 5	18 - 22	3.5 – 8.8	Acute and chronic LOCs are exceeded for these uses.
Lettuce (drip irrigation)	67	**1.8**	1 in 1	54	14	Acute and chronic LOC exceeded

Freshwater fish RQs ranged from 0.05 (beans) to 1.8 (lettuce) for the most sensitive species tested (bluegill). Based on a default probit slope of 4.5, the estimated probability of an individual effect to the most sensitive fish species was approximately 1 in 3E5 or less for beans, broccoli, cauliflower, and residential However, this analysis was based on the default slope of 4.5. Based on a reasonable lower bound probit slope, the probability of an individual mortality would range between approximately 1 in 200 to 1 in 50. A probability of an individual mortality of this magnitude would result in an undetectable reduction in available prey of the CRLF and would, therefore, be an insignificant effect (an effect that may occur, but would not harm or harass the assessed species). Therefore, the effects determination for these uses is not likely to adversely affect.

However, for all other uses (broccoli, asparagus, Brussels sprouts, cauliflower, and cabbage), the magnitude of effect would not be considered discountable and was 1 in 25 or greater based on a default probit slope of 4.5. Therefore, the effects determination is likely to adversely affect the CRLF for these uses.

5.2.2.4 Terrestrial Invertebrates

When the terrestrial-phase CRLF reaches juvenile and adult stages, its diet is mainly composed of terrestrial invertebrates. The RQ used to estimate potential effects to terrestrial invertebrates was 7.7. Based on the default probit slope of 4.5, the probability of an individual mortality approaches 100%. Two degradates have been shown to be more toxic to bees than disulfoton. The LD50s for the sulfoxide and sulfone degradates are 1.1 and 0.96 ug/bee, respectively, compared with an LD50 of 4 ug/bee for disulfoton. These degradates have been shown to form up to 94% of parent in degradation studies. Therefore, exposure to these degradates could also impact terrestrial invertebrates and indirectly affect the CRLF. AgDrift analysis indicates that the non-endangered species LOC of 0.5 could be exceeded for up to approximately 100 feet from the treated site.

This analysis shows that terrestrial invertebrates could be impacted at a level that could indirectly affect the CRLF by reducing available food. Therefore, a finding of likely to adversely affect (LAA) was made for the potential for disulfoton and its degradates to potentially impact terrestrial invertebrate prey base as available food.

5.2.2.5 Mammals

Dietary information for terrestrial-phase CRLFs indicate that large adult frogs consume terrestrial vertebrates, including mice. Acute mammalian RQs were up to approximately 100 for disulfoton, and LOCs were also exceeded for the sulfone degradate (sulfoxide degradate was not assessed due to lack of toxicity data). At these RQs, the probability of an individual mortality would approach 100%. In addition, the reproduction RQs were as high as 4600 for disulfoton. The RQ analysis suggests that exposed mammals could be impacted to a level that could adversely affect individual CRLFs that depend on them for food. AgDisp analysis indicates that LOCs would be exceeded for >1000 feet from the application site (see **Table 5.14**). Therefore, it was concluded that disulfoton and its degradates are likely to adversely affect the CRLF.

5.2.2.6 Terrestrial-phase Amphibians

Terrestrial-phase adult CRLFs also consume other frogs. RQ values representing direct exposures of disulfoton to terrestrial-phase CRLFs are used to represent exposures of disulfoton to frogs in terrestrial habitats. As demonstrated in **Table 5.10**, acute RQs that incorporated herptile food intake levels exceeded the acute LOC of 0.5 for frogs that consume several potential prey items of CRLFs at an application rate of 1 lb a.i./acre (single application). Therefore, it was concluded that disulfoton and its degradates are likely to adversely affect the CRLF.

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 streambanks. 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 are typically assessed using RQs from freshwater aquatic vascular and non-vascular plant data. However, no aquatic plant studies were submitted. Therefore, an evaluation of potential effects to aquatic plants could not be quantified. Based on the low toxicity and risk to terrestrial plants, potential magnitude of impacts to aquatic plants are not expected to be such that the CRLF may be indirectly affected.

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. Concern levels for terrestrial plants were not exceeded. Therefore, it was concluded that use of disulfoton is expected to have "no effect" on the CRLF by affected terrestrial plants.

5.2.4 Modification to Designated Critical Habitat

Based on the lack of potential effects to the CRLF resulting from impacts to terrestrial or aquatic plants, labeled uses of disulfoton are not expected to impact critical habitat based on PCEs that are related to presence and maintenance of aquatic or terrestrial vegetation. However, two PCEs may be impacted by use of disulfoton:

1. Reduction and/or modification of food sources for terrestrial phase juveniles and adults

2. Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.

5.2.5 Distance From Treated Site Effects May Occur

This assessment concluded that labeled uses of disulfoton could adversely affect the CRLF by direct and/or indirect effects when used according to the label on asparagus, beans, broccoli, Brussels sprouts, cabbage, cauliflower, Christmas trees, cotton, lettuce, and residential areas (all uses). Therefore, the CRLF could be affected in its habitat that overlaps with areas that produce these commodities and residential areas. Potential effects are not limited to the treated field. The environmental fate properties indicate that runoff and spray drift represent significant potential transport mechanisms of disulfoton to the aquatic and terrestrial habitats of the CRLF. Therefore, there is potential for disulfoton to be transported outside of the area where it is directly applied. Two transport pathways were evaluated to determine the potential distance from treated sites that could be impacted. Spray drift deposition was evaluated to determine the distance from treated sites that spray drift deposition would no longer be expected to affect CRLFs. The only uses with spray application methods for which spray drift is a potential transport pathway are asparagus, cabbage, and cotton. Also, for aquatic phase frogs, the distance downstream from use sites needed to dilute disulfoton concentrations to an extent that would not longer result in direct or indirect effects to the CRLF was evaluated. Downstream dilution analysis is relevant for all crops. This analysis is described briefly below and in more detail in Appendix C.

Since this screening level risk assessment defines taxa that are predicted to be exposed through runoff and drift to disulfoton at concentrations above the Agency's Levels of Concern (LOC), analysis of the potential spatial extent of effects requires expansion of the area from the treated site to include all areas potentially impacted by this federal action. Two methods are used to define these areas: (1) the down stream dilution assessment for determining the extent of the affected lotic aquatic habitats (flowing water); and (2) the spray drift assessment for determining the extent of potentially affected terrestrial habitats.

5.2.5.1 Downstream Dilution

In order to determine the extent of potential effects to lotic (flowing) aquatic habitats, the agricultural uses resulting in the greatest ratios of the RQ to the LOC for any endpoint for aquatic organisms is used to determine the distance downstream for concentrations to be diluted below levels that would be of concern (*i.e*. result in RQs above the LOC). This analysis is in **Table 5.13** below. For this assessment, the greatest ratio was 5600 (the highest aquatic invertebrate RQ = 5600; LOC = 1; 5600 / 1= 5600; see Table 5.13) for indirect effects to the CRLF through reproductive effects to aquatic invertebrates exposed to disulfoton (lettuce chemigation use). Using methods described in Appendix C, downstream analysis using this RQ determined that 257 km is the maximum distance from the edge of any potential use area to a point where it falls below the LOC.

Table 5.13. RQ/LOC Ratio for Various Landcover Classes for Aquatic Organisms[a]

Direct/Indirect Effects to CRLF	Exposure	Cropland	
		Highest RQ[a]	RQ/LOC Ratio
Direct and Indirect - Fish and Aquatic Amphibians	**Acute**	1.8	36
	Chronic	54	54
Indirect-Aquatic Invertebrates	**Acute**	17	340
	Chronic	5600	5600

a RQ Calculations are presented in Section 5.1; LOC for acute and chronic effects is 0.05 and 1.0, respectively.

5.2.5.1 Spray Drift

Table 5.14 indicates that at distances greater than approximately 8336 feet from the treated site, RQs for terrestrial organisms will be below LOCs. This evaluation was based on potential indirect effects to the CRLF from potential reduction in prey. The endangered species LOC was used for this analysis for acute effects; however, the probability of an individual effect at the endangered species LOC is approximately 1 in 30,000 assuming a probit slope of 4.5. The resulting potential impact to animal abundance would not be detectable in the environment. Therefore, distances associated with alternative levels of risk were calculated and are presented in **Table 5.15**. The restricted use LOC (0.2) and acute LOC (0.5) was used for this analysis. Based on a probit slope of 4.5, the probability of an individual mortality at these LOCs are approximately 1 in 100 and 1 in 10, respectively.

Table 5.14. AgDISP predicted Buffer Distance resulting in no Endangered Species LOC Exceedance for Terrestrial Animals for Disulfoton

Effect/ Taxonomic Group	Acute or Chronic Effect	Highest RQ[a]	Spray drift Fraction Needed to Reduce RQs to Below LOCs[b]	Distance from Treated Site Fraction is Achieved	
				Aerial spray (Asparagus; 1 lb a.i./A)	Ground spray (Asparagus, Cabbage; 1 lb a.i./A)
Direct (avian RQs)	**Acute**	44	0.23 %	4091 ft	3123 ft
	Chronic	5.7	17.5 %	223 ft	256 ft [c]
Indirect-mammals	**Acute**	55	0.18 %	4452 ft	3218 ft
	Chronic	2600	0.04 %	8336 ft	4258 ft
Indirect-Terrestrial Invertebrates	Acute Contact Exposures (**small insect**)	4.4	1.14 %	2404 ft	2670 ft [c]

[a] RQ Calculations are presented in Section 5.

[b] Spray drift fraction = 1/(RQ/LOC); Acute LOC = 0.1 (end. species), Chronic LOC = 1, Terrestrial Invertebrate LOC = 0.05

[c] Drift levels from aerial applications are generally expected to be higher than similar ground boom applications. As a result, if the aerial model suggests a smaller action area, that would be protective of ground applications, even though in some cases, uncertainties in the AgDisp model can lead to results for ground applications which are greater than for aerial applications.

Table 5.15. Spraydrift Fraction Resulting in no Restricted Use or Acute LOC Exceedance for Terrestrial Animals for Disulfoton

Effect/Taxonomic Group	Acute or Chronic Effect	Highest RQ[a]	Spraydrift Fraction Needed to Reduce RQs to Below Restricted Use LOC [b]	Distance from Treated Site Fraction is Achieved (aerial)	Spraydrift Fraction Needed to Reduce RQs to Below Acute LOC[b]	Distance from Treated Site Fraction is Achieved (aerial)
Direct (avian RQs)	**Acute**	44	0.45 %	3228	1.14%	2404
Indirect-mammals	**Acute**	55	0.36 %	3474	0.91 %	2582

[a] RQ Calculations are presented in Section 5.

[b] Restricted Use LOC = 0.2, Acute LOC = 0.5.

Similar to the analysis described above, the buffer distance needed to get below the most sensitive aquatic LOC was determined. This distance identifies those locations where water bodies can be impacted by spray drift deposition alone (no runoff considered) resulting in concentrations above the LOC. As with the terrestrial assessment, for each aquatic taxa of concern, the fraction of the application rate needed to reduce exposures to levels below LOCs is calculated. Based on this fraction and estimation of spray drift patterns, AgDISP determines the buffer distance required between the treated field and the water body to result in exposure estimates below the level of concern. Distances were based on the highest RQs for aerial applications to asparagus at 1 lb a.i./A. Drift levels from aerial applications are generally expected to be higher than similar ground boom applications and so results of the aerial model would be protective of ground applications as well. The analysis yields much lower buffer distances than the terrestrial buffer, as presented in **Table 5.16**.

Table 5.16. Spraydrift Fraction Resulting in no LOC Exceedance for Aquatic Animals for Disulfoton

Effect/ Taxonomic Group	Acute or Chronic Effect	Highest RQ[a]	LOC Classification	Spray drift Fraction Needed to Reduce RQs to Below LOCs[b]	Distance from Treated Site Fraction is Achieved (aerial)
Direct & Indirect (Fish and Frogs)	**Acute**	1.8	End. Species	2.78 %	1151 ft
			Restricted	5.56 %	387 ft
			Acute	27.78 %	0 ft
	Chronic	54	NA	1.85 %	2057 ft
Indirect (Aquatic Invertebrates)	**Acute**	17	End. Species	0.29 %	3415 ft

[a] RQ Calculations are presented in Section 5.
[b] Spray drift fraction = 1/(RQ/LOC); Acute LOC = 0.05 (end. species), 0.1 (restricted), 0.5 (acute); Chronic LOC = 1

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. Additionally, for residential uses, labels do not express application rates on a per acre basis. Therefore, for purposes of aquatic modeling, conservative assumptions were made about the area that would be treated, leading to uncertainty in estimating exposure.

6.1.2 Aquatic Exposure Modeling of Disulfoton

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.

Surface water modeling using PRZM/EXAMS does not consider discharge of contaminated groundwater to stream baseflow as a potential route of aquatic exposure. Modeling results may therefore underestimate exposure in some surface waters relevant to CRLF habitat because discharging groundwater is likely to support low-order streams, wetlands, and intermittent ponds, environments that are favorable to CRLFs. Long-term chronic concentrations derived from the PRZM-EXAMS model are assumed to reflect background concentrations that might be found in discharged groundwater/stream baseflow. Groundwater monitoring data available from California sites have no detections of disulfoton or its transformation products. One study conducted in Wisconsin, though, found levels of up to 100 ug/L, indicating that disulfoton does have the potential to reach groundwater and suggesting that the assumption of lower disulfoton levels in discharging groundwater may not be conservative.

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. Except for the asparagus use, all aquatic modeling assumed that disulfoton was applied to bare soil which may overestimate exposure from foliar applications because foliar dissipation is not accounted for. Additionally, application dates were chosen to represent times within the potential application window when precipitation was highest, which leads to conservative exposure estimations. 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.

Aquatic modeling was conducted for total toxic residues, including disulfoton + d. sulfoxide + d. sulfone. This adds additional uncertainty to the modeling inputs because degradation half-lives were calculated from studies intended to investigate degradation patterns for the parent compound and may not include sufficient data to accurately represent transformation of the degradates as well. Koc inputs were based on the most mobile component of the total residues and so may overestimate mobility of the group.

Disulfoton labels require a 25 foot buffer around any surface water bodies. Tools are not currently available to evaluate the effectiveness of a vegetative setback on preventing 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.

Exposure estimates are based on modeling alone because limited monitoring data for total residues are available to compare to the modeled estimates. Available monitoring data are primarily for parent disulfoton and do not include the more persistent and more mobile degradates and so are not likely to capture all exposure. Both the NAWQA and CDPR include monitoring data in California for disulfoton in surface and ground water with no detections. The specific use patterns (e.g. application rates and timing, crops) associated with the agricultural areas are unknown, but they are assumed to be representative of potential disulfoton use areas. Both d. sulfoxide and d. sulfone have been detected in a subset of these data despite limited sampling. This demonstrates that total toxic residues are more likely to reach water bodies than parent alone and indicates that the lack of detections of parent disulfoton is insufficient to conclude that exposure is unlikely. The assessment discussion of disulfoton detections in surface and ground water monitoring data from sites outside of California. There is uncertainty in these data because the environmental conditions may not be representative of conditions in the area of concern.

6.1.3 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.

6.1.4 Terrestrial Exposure Modeling of disulfoton

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 reflect 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.

6.1.5 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 disulfoton from multiple applications, each application of disulfoton 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*.).

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 disulfoton are not available for frogs or any other aquatic-phase amphibian; therefore, freshwater fish are used as surrogate species for aquatic-phase amphibians. 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 Sublethal Effects

When assessing acute risk, the screening risk assessment relies on the acute mortality endpoint as well as a suite of sublethal responses to the pesticide, as determined by the testing of species response to chronic exposure conditions and subsequent chronic risk assessment. Consideration of additional sublethal data in the effects determination t is exercised on a case-by-case basis and only after careful consideration of the nature of the sublethal effect measured and the extent and quality of available data to support establishing a plausible relationship between the measure of effect (sublethal endpoint) and the assessment endpoints. However, the full suite of sublethal effects from valid open literature studies is considered for the purposes of defining the action area.

As discussed in Section 4, cytolologic effects were observed in the liver of fish exposed to disulfoton at levels as low as 0.1 ug/L (Arnold et al. (1996). These effects were not used in the current risk assessment for the purpose of this effects determination because the level of effects observed in the available studies have not been directly correlated with the assessment endpoints of survival or reproduction.

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 disulfoton 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 disulfoton. Additionally, the Agency has determined that there is the potential for modification of CRLF designated critical habitat from the use of the chemical. This determination applies to all currently labeled uses.

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 7.1a Effects Determination Summary for Direct and Indirect Effects of Disulfoton on the CRLF

Assessment Endpoint	Effects Determination[1]	Basis for Determination
	Aquatic-Phase CRLF (Eggs, Larvae, and Adults)	
<u>Direct Effects:</u> Survival, growth, and reproduction of CRLF individuals via direct effects on aquatic phases	**LAA**	Endangered species LOC was exceeded for all uses; chronic LOC was exceeded for all uses except cotton, beans, and residential uses. Potential effect was not considered discountable or insignificant.
<u>Indirect Effects:</u> Survival, growth, and reproduction of CRLF individuals via effects to food supply (*i.e.*, freshwater invertebrates, non-vascular plants, fish, and frogs)	<u>Freshwater invertebrates</u>: **LAA**	Acute and chronic RQs were exceeded for all uses. Acute RQs ranged from approximately 0.5 to 17 and chronic RQs ranged from 145 to 5600. The potential magnitude of effect could be sufficient to result in indirect effects to the CRLF.
	<u>Non-vascular aquatic plants</u>: **NE**	No aquatic plant toxicity data have been submitted or were located in the open literature. Disulfoton is an insecticide, and EC25s for terrestrial plants were greater than the maximum application rate.

Table 7.1a Effects Determination Summary for Direct and Indirect Effects of Disulfoton on the CRLF		
Assessment Endpoint	**Effects Determination[1]**	**Basis for Determination**
	Fish and frogs: **LAA** for some uses	Magnitude of potential impacts to fish and aquatic phase amphibians could be sufficient to indirectly affect the CRLF for some uses. The highest RQs occurred for the lettuce, cabbage, and asparagus uses.
Indirect Effects: 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: **NE** Vascular aquatic plants: **NE**	No aquatic plant toxicity data have been submitted or were located in the open literature. However, disulfoton is an insecticide, and EC25s for terrestrial plants were greater than the maximum application rate.
Indirect Effects: 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.	**NE**	The EC25 is greater than the highest labeled application rate for all uses except Christmas trees. The Christmas tree RQ would be <0.5.

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

Table 7.1b Effects Determination Summary for Direct and Indirect Effects of Disulfoton on the CRLF

Assessment Endpoint	Effects Determination[1]	Basis for Determination
Terrestrial-Phase CRLF (Juveniles and adults)		
Direct Effects: Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	**LAA**	Acute LOC (0.5) was exceeded for all uses for disulfoton and its degradates. Potential for reproductive effects also exists for all uses.
Indirect Effects: Survival, growth, and reproduction of CRLF individuals via effects on prey (*i.e.*, terrestrial invertebrates, small terrestrial vertebrates, including mammals and terrestrial phase amphibians)	Terrestrial invertebrates: **LAA**	The endangered species LOC of 0.05 was exceeded for all uses. Also, disulfoton is an insecticide, and the potential magnitude of effect could be sufficient to result in indirect effects to the CRLF.
	Mammals: **LAA**	Acute (0.5) and chronic (1.0) LOCs were exceeded for all uses. The potential magnitude of effect could be sufficient to result in indirect effects to the CRLF.
	Frogs: **LAA**	Acute (0.5) and chronic (1.0) LOCs were exceeded for all uses. The potential magnitude of effect could be sufficient to result in indirect effects to the CRLF.
Indirect Effects: Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.*, riparian vegetation)	**NE**	The EC25 is greater than the highest labeled application rate for all uses except Christmas trees. The Christmas tree RQ would be <0.5.

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

Table 7.2 Effects Determination Summary for the Critical Habitat Impact Analysis		
Assessment Endpoint	**Effects Determination[1]**	**Basis for Determination**
Aquatic-Phase CRLF 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.	**NE**	Effects determination for potential effects related to impacts on aquatic and terrestrial plants was No Effect.
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.[2]	**NE**	Effects determination for potential effects related to impacts on aquatic and terrestrial plants was No Effect.
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	**HM**	Effects determination for direct and indirect effects to the CRLF was LAA.
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.*, algae)	**NE**	Effects determination for potential effects related to impacts on aquatic plants was No Effect.
Terrestrial-Phase CRLF 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	**NE**	Effects determination for potential effects related to impacts on aquatic and terrestrial plants was No Effect.
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	**NE**	
Reduction and/or modification of food sources for terrestrial phase juveniles and adults	**HM**	Effects determination for indirect effects via reducing available food supply was LAA.
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	**HM**	Effects determination for direct and indirect effects was LAA.
[1] NE = No effect; HM = Habitat Modification [2] 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.		

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 adverse 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 (Citations with MRID Nos. in the assessment are in Appendix I)

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