

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

Pesticide Effects Determination

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

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Primary Authors:

Colleen Flaherty, Biologist

Keara Moore, Chemist

Pamela Hurley, Toxicologist

Secondary Review:

Brian Anderson, Biologist

James Hetrick, Senior Scientist

Branch Chief, Environmental Risk Assessment Branch 3:

Dana Spatz

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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 methidathion 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.

Methidathion is a non-systemic organophosphate insecticide/acaricide registered for use to control a wide range of sucking, leaf-eating, and scale insects. Currently labeled uses of methidathion considered as part of the federal action evaluated in this assessment include: Alfalfa, almond, apple, apricot, artichoke, calemondin, cherry, citron, citrus, clover, cotton, grapefruit, kiwi, kumquat, lemon, lime, mango, nectarine, olive, peach, pear, plum, prune, pummelo, safflower, sunflower, tangelo, tangerine, timothy, walnut, and nursery stock. Methidathion is formulated as an emulsifiable concentrate or a wettable powder. Applications can be applied by ground, airblast, or aerial spray with annual application rates that range from 1 lb active ingredient/A to 10 lb a.i./A. Label requirements include restrictions on spray heights and buffer widths in order to reduce risk from spray drift.

Based on laboratory studies, methidathion has generally low persistence in soil, with aerobic and anaerobic soil metabolism half-lives of less than two weeks. Dissipation rates appear to slow over time, however, so there is a possibility that in some conditions low levels may persist. Terrestrial field dissipation studies found half-lives of less than 30 days. In water, methidathion has limited hydrolysis except in alkaline conditions, but it is subject to aquatic photolysis with a half-life of 10 days. There are no aquatic metabolism data available. Soil adsorption studies indicate that methidathion is moderately mobile, but it has not been detected below 18 inches in any terrestrial field dissipation studies. There is some potential for long range atmospheric transport. Although the physical properties suggest that volatilization would be limited, monitoring studies have found methidathion residues at more than 20 km from any use site.

Soil metabolism of methidathion leads primarily to carbon dioxide and bound residues, but one unidentified degradate was formed at up to 13% and appeared to be persistent. Other major degradates include S-12956¹, formed through hydrolysis and through aquatic

¹ 5-methyl-1-3,4-thiadazol-2 (3H)-one

and soil photolysis, and des-methyl GS-13007² and PAMMTO³, formed through hydrolysis in some conditions. Methidathion oxon⁴ is formed only as a minor degradate, but it is included as a stressor of concern in this assessment because, based on data for other organophosphate pesticides, oxon degradates have the potential to be much more toxic than the parent. Laboratory studies demonstrate that at least 4.9% of applied methidathion can be transformed to methidathion oxon through soil photolysis, but no data are available regarding photo-oxidation in air, which could also be an important route of transformation.

Since CRLFs exist within aquatic and terrestrial habitats, exposure of the CRLF, its prey and its habitats to methidathion are assessed separately for the two habitats. Tier-II aquatic exposure models are used to estimate high-end exposures of methidathion in aquatic habitats resulting from runoff and spray drift from different uses. Peak model-estimated aquatic environmental concentrations resulting from different methidathion uses range from 7.3 to 114.7 µg/L. These estimates are supplemented with analysis of available California surface water monitoring data from U. S. Geological Survey's National Water Quality Assessment (NAWQA) program and the California Department of Pesticide Regulation. The maximum concentration of methidathion reported by NAWQA for California surface waters from 15 sites is 0.31 µg/L. The California Department of Pesticide Regulation surface water database included more than 4,000 samples from 135 locations in agricultural areas and reported a maximum measured concentration of 15.1 µg/L, which is in the range of peak modeled values for the different uses of methidathion. Ground water *per se* is not evaluated herein, but it is nonetheless significant 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 could reflect background concentrations that might be found in discharged groundwater/stream baseflow. NAWQA monitoring data did not detect methidathion in ground water sampling at more than 200 sites, most of which were collected in counties with high methidathion use in areas with land cover classified as agricultural or mixed use.

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

There are also atmospheric monitoring data which demonstrate long range transport of both methidathion and methidathion oxon. Both species have been detected in air, fog, and/or rain at local, intermediate, and long-range locations. In the monitoring study with

² des-methyl S-[(5-methoxy-2-oxo-1, 3, 4-thiadiazol-3 (2-1)-yl-methyl O,O-dimethylphosphorothioate]

³ phosphorothioic acid, 4-(mercaptomethyl)-2-methoxy-A2-,13,4-thiadiazolin-5-one

⁴ S-[(5-methione-2-oxo-1,3,4-thiadiazol-3(2H)-yl methyl o,o dimethyl phosphorothioate; (GS-13007)

data at the furthest range, methidathion was detected in air at 22 km from the nearest use site and methidathion oxon was detected at a site another 10 km away. Methidathion oxon was also detected on pine needles at the 22 km site.

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.

This assessment primarily considers effects of exposures to methidathion only but quantitative characterization is provided for methidathion oxon, the one known degradate of concern. Although no toxicity data are available for methidathion oxon, it is presumed to be of concern because toxicity data for oxon transformation products of similar pesticides in the organophosphate class suggest they will be of equal or greater toxicity than the parent. There are other degradates suggested by the degradation pathway but not tested for in laboratory studies that may also be of concern. No data are available regarding the formation and decline or the toxicity of methidathion oxon or of any of the other potential OP or oxon degradates. Given that acute and chronic risk quotients are exceeded for the parent compound alone, any contribution in toxicity from any of the major degradates or from methidathion oxon or other OP degradates would increase the risk estimates.

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 methidathion 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 each particular type of effect are below LOCs, the pesticide is determined to have "no effect" on the CRLF. 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 methidathion 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 methidathion. Additionally, the Agency has determined that there is the potential for modification of CRLF designated critical habitat from the use of the chemical. For direct effects, the acute listed species LOCs for freshwater fish and for birds (surrogate to the aquatic-and terrestrial-phase CRLF, respectively) are exceeded for all uses. The chronic LOC for both freshwater fish and birds is also exceeded for all uses. For indirect effects, with the exception of mangos on an acute dietary basis for non-listed species, the acute dose-based, acute dietary-based and chronic dietary-based RQs for birds (terrestrial-phase amphibians) exceed both the acute non-listed species LOC and/or the chronic LOC for all of the assessed uses. For aquatic and terrestrial invertebrates and mammals, the acute RQs exceed the acute non-listed LOC for aquatic and/or terrestrial animals for all uses. The chronic LOC for aquatic and/or terrestrial animals is also exceeded for aquatic invertebrates, mammals and fish/aquatic-phase amphibians (all uses). No studies are available for aquatic vascular and non-vascular and terrestrial plants; however, the weight of the evidence indicates an effect determination of may affect, not likely to adversely affect (NLAA).

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**. Use-specific determinations for direct and indirect effects to the CRLF are provided in **Tables 1.3 and 1.4**. Further information on the results of the effects determination is included as part of the Risk Description in Section 5.2. Given the LAA determination for the CRLF and potential modification of designated critical habitat, a description of the baseline status and cumulative effects for the CRLF is provided in **Attachment 2**.

Table 1.1 Effects Determination Summary for Methidathion Use and the CRLF		
Assessment Endpoint	Effects Determination ¹	Basis for Determination
Survival, growth, and/or reproduction of CRLF individuals	LAA¹	Potential for Direct Effects
		<p><i>Aquatic-phase (Eggs, Larvae, and Adults):</i> Highly toxic to freshwater fish. Acute listed species LOC exceeded for all uses for three tested surrogate species. Chronic effects based on survival and growth. Chronic LOC exceeded for all uses. Probability of an individual effect on an acute basis is high, both at the acute LOC and at the RQ levels. Incident data also indicate freshwater fish vulnerability, and monitoring has detected surface water concentrations above the acute endpoint. Significant overlap expected to exist between use sites and CRLF habitat, particularly when the spraydrift and downstream dilution buffers are added to the methidathion use area. Spraydrift distances range from 23 to over 1000 feet and the downstream dilution buffer is 285 km.</p>
		<p><i>Terrestrial-phase (Juveniles and Adults):</i> Moderately toxic to very highly toxic to avian species. Acute dose-based, acute dietary-based and chronic dietary-based RQs for the terrestrial-phase CRLF, using avian data as a surrogate, exceed acute listed species LOC and chronic LOC for all of the assessed uses. Chronic effects based on reduction in number of normal hatchlings/live 3-week embryos. Limited data indicate that on a dose basis, the acute LOC is exceeded for all uses, even with the least sensitive species at the lowest application rate (mangos). On a dietary basis, the same is true for all uses except mangos. Probability of an individual effect on an acute basis is high at the RQ levels. Incident data indicate that birds are vulnerable. Significant overlap exists between use sites and CRLF habitat, particularly when the spraydrift buffers are applied to the methidathion use area. Spraydrift distances range from 194 to over 1000 feet.</p>
		Potential for Indirect Effects
		<p><i>Aquatic prey items, aquatic habitat, cover and/or primary productivity</i> Very highly toxic to freshwater invertebrates. Acute and chronic RQs exceed acute LOC for non-listed species and chronic LOC, respectively for all assessed uses. The percentage effect to the aquatic invertebrate prey base for all uses is very high. For fish (aquatic-phase amphibians), acute non-listed species LOC exceeded for all uses. Chronic LOC exceeded for all uses. Percentage effect to the freshwater fish/aquatic-phase amphibian prey base is very high. No studies are available for aquatic vascular and non-vascular plants.</p>
		<p><i>Terrestrial prey items, riparian habitat</i> RQs for both small and large insects exceed acute LOC for non-listed species for all methidathion uses, even with only a single use. Insufficient studies to conduct a sensitivity analysis; however, lowest RQ, using highest terrestrial invertebrate endpoint still exceeds the acute list species LOC. Percentage effect to the terrestrial invertebrate prey base for all uses is very high. For terrestrial-phase amphibians using avian data as a surrogate, with the exception of mangos on an acute dietary basis, the acute dose-based, acute dietary-based and chronic dietary-based RQs exceed the acute non-listed species LOC and the chronic LOC for all of the assessed methidathion uses, respectively. Percentage effect to the avian/terrestrial-phase amphibian prey base is very high. For mammals, the acute and chronic RQs exceed the non-listed species acute LOC and the chronic LOC, respectively for all uses. At the lowest RQ, the percentage effect to the mammalian prey base is very high. No studies are available for terrestrial plants.</p>

¹ No effect (NE); May affect, but not likely to adversely affect (NLAA); May affect, likely to adversely affect (LAA)

Table 1.2 Effects Determination Summary for Methidathion Use and CRLF Critical Habitat Impact Analysis		
Assessment Endpoint	Effects Determination ¹	Basis for Determination
Modification of aquatic-phase PCE	Habitat Modification¹	<p>No studies available for aquatic vascular and non-vascular plants and terrestrial plants.</p> <p>For the aquatic-phase CRLF, acute non-listed species LOC for freshwater fish (aquatic-phase amphibians) exceeded for all uses. Chronic LOC for freshwater fish exceeded for all uses. Probability of an individual effect and percentage effect to the freshwater fish/aquatic-phase amphibian prey base on an acute basis at the RQ are high. Incident data support freshwater fish vulnerability. For freshwater invertebrates, acute and chronic RQs exceed acute LOC (non-listed species) and chronic LOC, respectively for all assessed uses. Again, the percentage effect to the aquatic invertebrate prey base is very high. Significant overlap expected between use sites and CRLF habitat, particularly when the spraydrift and downstream dilution buffers are added to the use area. Spraydrift distances range from 23 (mangos) to over 1000 feet and the downstream dilution buffer is 285 km.</p>
Modification of terrestrial-phase PCE		<p>No studies are available for terrestrial plants.</p> <p>For the terrestrial-phase CRLF, using avian data as a surrogate, with the exception of mangos on a dietary basis for non-listed species, the acute dose-based, acute dietary-based and chronic dietary-based RQs for the terrestrial-phase CRLF, exceed both the acute listed and non-listed species LOC and chronic LOC for all of the assessed uses. Probability of an individual effect on an acute basis at the lowest RQ level and percentage effect to the avian/terrestrial-phase amphibian prey base are very high. Incident data indicate that birds are vulnerable. RQs for both small and large insects exceed acute LOC for non-listed species for all methidathion uses, even with only a single use. Percentage effect to terrestrial invertebrate prey base for all uses are very high. For mammals, the acute and chronic RQs exceed the non-listed species acute LOC and the chronic LOC, respectively for all uses. The percentage effect to the mammalian prey base is very high for all uses. Significant overlap exists between use sites and CRLF habitat, particularly when the spraydrift buffers are applied to the methidathion use area. Spraydrift distances range from 194 (mangos) to over 1000 feet.</p>

¹ Habitat Modification or No effect (NE)

Table 1.3 Methidathion Use-specific Direct Effects Determinations¹ for the CRLF				
Use(s)	Aquatic Habitat		Terrestrial Habitat	
	Acute	Chronic	Acute	Chronic
All uses	LAA	LAA	LAA	LAA

¹ NE = No effect; NLAA = May affect, but not likely to adversely affect; LAA = Likely to adversely affect

Table 1.4 Methidathion Use-specific Indirect Effects Determinations ¹ Based on Effects to Prey										
Use(s)	Algae	Aquatic Invertebrates		Terrestrial Invertebrates (Acute)	Aquatic-phase frogs and fish		Terrestrial-phase frogs		Small Mammals	
		Acute	Chronic		Acute	Chronic	Acute	Chronic	Acute	Chronic
All uses	NLAA	LAA	LAA	LAA	LAA	LAA	LAA	LAA	LAA	LAA

¹ NE = No effect; NLAA = May affect, not likely to adversely affect; LAA = Likely to adversely affect

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 methidathion on various agricultural and non-agricultural uses. In addition, this assessment evaluates whether use on these crops is expected to result in modification of the species' designated critical habitat. This ecological risk assessment has been prepared consistent with a settlement agreement in the case *Center for Biological Diversity (CBD) vs. EPA et al.* (Case No. 02-1580-JSW(JL)) 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. 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 methidathion is based on an action area. The action area is the area directly or indirectly affected by the federal action, as indicated by the exceedence of the Agency's Levels of Concern (LOCs). It is acknowledged that the action area for a national-level FIFRA regulatory decision associated with a use of methidathion 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 methidathion 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 methidathion 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 methidathion.

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

Methidathion is used as an insecticide on a variety of terrestrial food and feed crops and terrestrial non-food crops. Based on usage data provided by the Biological and Economic Analysis Division (BEAD), on average, roughly 110,000 pounds of methidathion are applied annually to agricultural crops. Methidathion usage is highest on almonds and oranges, with annual average applications of 20,000 lbs. a.i. applied. The crop with the highest average percent crop treated with methidathion is artichokes at 60% (10,000 lbs. a.i. applied).

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 methidathion in accordance with the approved product labels for California is “the action” relevant to this ecological risk assessment.

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

Methidathion degrades into one known degradate of concern, methidathion oxon. Although no toxicity data are available for methidathion oxon, it is presumed to be of concern because toxicity data for oxon transformation products of similar pesticides in the organophosphate class suggest they will be of equal or greater toxicity than the parent. Methidathion oxon is the only degradate considered in this analysis. There are no toxicity data for the four major degradates, but these compounds no longer contain the OP moiety and so are assumed to be of lower toxicity than the parent.

Methidathion has no registered products that contain multiple active ingredients.

2.3 Previous Assessments

2.3.1 Methidathion Interim Registration Eligibility Decision, 2002

The Agency completed a screening-level ecological risk assessment (USEPA, 1999) in support of the Interim Reregistration Eligibility Decision (IRED) for methidathion (USEPA 2002). Completion of the organophosphate (OP) cumulative assessment (USEPA 2006b) resulted in finalization of the IRED as a Reregistration Eligibility Decision (RED) (USEPA 2006a), which is summarized below.

The Agency’s ecological risk assessment was based principally on methidathion’s use on cotton, citrus, stone fruits, nut crops, and artichokes. The assessment was based on data collected in the laboratory and in the field to characterize the fate and

ecotoxicological effects of methidathion. Data sources used in this assessment included: 1) registrant submissions in support of reregistration, 2) publicly available literature on ecological effects, and 3) incident reports of adverse effects on aquatic and terrestrial organisms associated with the use of methidathion.

The RED concluded that methidathion poses a risk to ecosystems in use areas. The levels of concern were exceeded for both acute and chronic effects to mammals, birds, fish, and aquatic invertebrates. In addition, risk to terrestrial invertebrates was presumed. The presumption of risk to non-target aquatic and terrestrial animals was supported by field studies and adverse ecological incidents. The potential risk to terrestrial plants was not assessed due to a lack of toxicity data. Toxicity data available for degradates of methidathion, including methidathion oxon, were not available for consideration in this assessment.

To address the presumed risk to birds, the Agency required the additional dilution of methidathion products (a minimum of 500 gallons of water per acre) in an attempt to reduce the amount of methidathion on food items. In addition, precautionary statements regarding the risk to birds were required to be added to the label in order to protect avian species.

To address the presumed risk to fish and aquatic invertebrates, the following risk mitigation strategies were required:

For all applications applied at rates greater than 3.0s a.i./A:

- Do not apply within 50 feet of lakes, reservoirs, rivers, permanent streams, natural ponds, marshes or estuaries
- For all applications applied at rates of 3.0s a.i./A or less:
 - Do not apply within 25 feet of lakes, reservoirs, rivers, permanent streams, natural ponds, marshes or estuaries
- For ground applications:
 - Shut off sprayer when turning at end rows
 - Do not apply when gusts or sustained winds exceed 12 mph
- For air blast application:
 - Adjust deflectors and aiming devices so that spray is only directed into the canopy
 - Block off upward pointed nozzles when there is no overhanging canopy
 - Use only enough air volume to penetrate the canopy and provide good coverage
 - Do not allow spray to go beyond the edge of the cultivated area. Spray the outside row only from outside the planting
- For aerial application:
 - Do not apply within 150 feet of water
 - Do not apply when gusts or sustained winds exceed 8 mph

In addition, to mitigate the risk to aquatic animals, a label amendment was required to include a surface water advisory statement.

The Agency was also concerned about the risk to beneficial insects from the use of methidathion. To reduce the likelihood for significant mortality to bees, precautionary labeling was required.

2.3.2. Organophosphate Cumulative Assessment, and Methidathion Reregistration Eligibility Decision, 2006

Because the Agency determined that methidathion shares a common mechanism of toxicity with the structurally-related organophosphate insecticides, a cumulative human health risk assessment for the organophosphate (OP) pesticides was necessary before the Agency could make a final determination of reregistration eligibility of methidathion. This cumulative assessment was finalized in 2006 (USEPA 2006b). The results of the Agency's ecological assessments for methidathion are discussed in the July 31, 2006, final Reregistration Eligibility Decision (RED) (USEPA 2006a).

The OP cumulative relied on a combined assessment methodology of modeling and monitoring data for human health exposure via drinking water. Unlike other assessments, the cumulative approach focused on regions of high OP use. No ecological risks were evaluated in the OP cumulative process, but its analysis of methidathion fate properties and its modeling approach remain relevant to this assessment. Additionally, the Agency requested toxicity data on methidathion oxon to further refine estimates.

2.3.3 Assessment of Potential Effects to 26 Evolutionarily Significant Units of Pacific Salmon and Steelhead

In April 2004, an assessment of methidathion uses in the Pacific northwest and California was conducted to determine potential effects to 26 evolutionarily significant units (ESUs) of Pacific salmon and steelhead. Based on toxicity and fate data, the usage information, habitat information and other factors, that assessment determined that uses of methidathion would have no effect on 7 ESUs, was not likely to adversely affect 9 ESUs and may affect the remaining 10 ESUs.

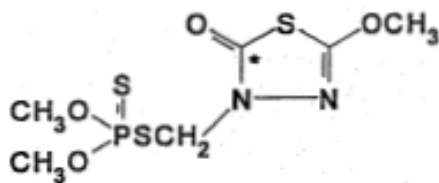
2.4 Stressor Source and Distribution

2.4.1 Environmental Fate and Transport

2.4.1.1. Physical and Chemical Properties

Chemical name: 3-Dimethoxyphosphinothioylthiomethyl-5-methoxy-1,3,4-thiadiazol-2(3H)-one.

Chemical structure:



Molecular formula:	C ₆ H ₁₁ N ₂ O ₄ PS ₃ .
Molecular weight:	302.3 g/mol
Physical state:	Colorless crystals.
Melting point:	39-40°C.
Solubility (20°C):	250 mg/L water;
Vapor pressure (20°C):	2.48 x 10 ⁻⁶ mm Hg
Henry's Law Constant:	3.97 x 10 ⁻⁹ atm m ³ /mol
Octanol/Water Partition Coefficient:	295 at pH 6.1

2.4.1.2. Environmental Fate Properties

Methidathion is classified as moderately mobile, and through most routes of transformation for which data are available, initial degradation is rapid and occurs with half-lives of less than two weeks, although in some cases, transformation slows and lower levels can persist for a longer time. **Table 2.1** summarizes the environmental fate properties of methidathion, along with the major and minor degradates detected in the submitted environmental fate and transport studies. See **Appendix A** for a bibliography of these submitted studies.

Table 2.1. Summary of Methidathion Environmental Fate Properties				
Study	Value (units)	Major Degradates^a Minor Degradates	MRID #	Study Status
Hydrolysis half life	pH 5: 37, 27d pH 7: 48, 25d pH 9: 13, 8d	GS-12956 Des-methyl GS-13007	42037701 44554501	Acceptable Acceptable
Direct Aqueous Photolysis	10.0 d ^b	GS-12956	42081709	Supplemental
Soil Photolysis	40 d ^c	GS-12956 <i>Methidathion oxon</i> (4.9%) GS-28307	42081710	Supplemental
Air Photolysis	No data	--	42215101	Unacceptable
Aerobic Soil Metabolism	11.3 d (sandy loam) 8.5 d (sandy loam) ^d	Unidentified ^e <i>Methidathion oxon</i> (0.2%) GS-12956, GS-28369 GS-28370, GS-20865	44545101 42262501	Acceptable Supplemental
Anaerobic Soil Metabolism	DT90 < 30 d (System)	GS-28369, GS-12956, <i>Two unidentified</i>	42262501	Supplemental
Anaerobic Aquatic Metabolism	No data	--		No study submitted
Aerobic Aquatic Metabolism	No data	--		No study submitted
Mobility	K _d : 2.5 to 14.8 mL/g Avg = 6.5 mL/g K _{oc} : 194 to 589 mL/g _{oc} Avg = 364 mL/g _{oc}	n/a	00158529	Acceptable
Laboratory Volatility	No data	--	42098801	Unacceptable
Terrestrial Field Dissipation	CA: 5-30 d (4 plots) NE: <15 d (1 plot)	GS-12956 <i>methidathion oxon</i>	40094103 41924401 41924402	Supplemental Supplemental Supplemental

^a Major degradates are those formed at >10% of the applied. Although a minor degradate, methidathion oxon is of toxicological concern and so the maximum percentage detected is reported here. Other oxon degradates that may be of toxicological concern may not have been detected because available studies did not include radiolabeling on the phosphate ester side chain of the parent compound.

^b The RED reported half-life of 11.6 days was not corrected for sunlight or for results from the dark control.

^c Problems with the study suggest that the measured half-life of 40 days is not representative of actual behavior. An assessment by the CDPR reports a soil photolysis half-life of 1.5 days.

^d Previous assessments reported this value as 3 days. That is in fact the DT50 and so this was updated to represent the linear first order half-life.

^e The degradate was unidentified but was tentatively characterized as a cyclic structure resulting from the reaction of carbazic acid and cysteine.

Both aerobic and anaerobic soil metabolism show initially rapid degradation. In two aerobic soil metabolism studies, DT50s were 3 to 7 days and linear first order half-lives were less than 2 weeks, although one study showed some slowing over time, indicating that methidathion may persist at lower levels for several months. One anaerobic study

was conducted but sampling was insufficient to determine a half-life. At the time of flooding, methidathion represented 41% of the initially applied radioactivity and by the first sampling point at 30 days post-flooding, it had been reduced to 6%. However, by 30 days later metabolism had not progressed, with 5% of the initially applied radioactivity remaining as methidathion. On the soil surface, photodegradation of methidathion may occur, but there is some uncertainty regarding the rate. In the submitted laboratory study, photolysis was slow with a half-life of 40.6 days, but problems in the study suggest this may underestimate the actual rate. An assessment by the California Department of Pesticide Regulation (CDPR) reports a much more rapid soil photolysis half-life of 1.5 days (Washburne, 2003).

Methidathion may reach water bodies through spray drift or runoff, in the dissolved phase or adsorbed to eroding soil. In water, methidathion is only moderately susceptible to abiotic hydrolysis in acidic and neutral conditions with half-lives in two studies ranging from 25 to 48 days. Dissipation rates increase in alkaline conditions with half-lives of 8 to 13 days at pH 9. In clear, shallow water, transformation may be more rapid due to photodegradation. A laboratory aquatic photolysis study measured a half life of 10.0 d, corrected for the dark control and natural sunlight. There are no data available for metabolism rates in aquatic environments.

In supplemental terrestrial field dissipation studies on four plots in California, methidathion had reported DT50s in the top 6 inches of 5 to 30 days, and in one plot in Nebraska, the DT50 was <15 days. Methidathion was not detected at depths below 18 inches in the field studies. There were several detections of methidathion in samples collected from the 12-18 inch cores, but these detections occurred in samples collected before any irrigation or rainfall event occurred and so appear to have been due to contamination during sample collection.

Dissipation of dislodgeable residues on foliar surfaces was found to occur with half-lives of ≤ 3 days based on the results of six studies, and two studies found half-lives for dissipation of total residues in foliage to be 3.5 and 5 days. Additionally, methidathion is not expected to bioaccumulate.

2.4.1.3. Transformation Products

Soil metabolism of methidathion leads primarily to carbon dioxide and bound residues. Other major degradates include GS-12956, formed through hydrolysis and through aquatic and soil photolysis (maxima from 18% to 66% of the applied radioactivity); des-methyl GS-13007 and PAMMTO, formed through hydrolysis at up to 21% and 18%, respectively; and an unidentified compound formed through aerobic soil metabolism and tentatively characterized as a cyclic structure resulting from the reaction of carbazic acid and cysteine. This compound appears to be persistent, as it reaches its maximum level of 13% of the applied radioactivity on day 11 but remained at 10% by the end of the study (day 263). Multiple minor degradates were formed, not all of them identified. See **Appendix B** for chemical names and structures of all identified degradates. None of the

major degradates contain the OP moiety and so they are likely to be less toxic than the parent.

The only detected degradate of toxicological concern is minor degradate methidathion oxon. Many of the OP pesticides can undergo oxidative desulfonation (cleavage of P=S bond to form P=O bond) to form oxons, either through photo-oxidation or chemical oxidation in the presence of oxidizing agents, and oxons have the potential to be considerably more toxic than the parent compounds. In submitted laboratory studies, methidathion oxon has been documented to form via chlorination, soil photolysis (up to 4.9% of the applied), and aerobic soil metabolism (0.2%), and the oxon was also detected in studies of terrestrial field dissipation and of dislodgeable foliar residues. The results of the soil photolysis study may underestimate oxon formation, because the maximum level was measured in the final sampling period and so oxon levels may continue to increase, and also because the study review concluded that due to a study deficiency, the rate of transformation observed may have been too low, supported by the more rapid transformation cited by CDPR (Washburn, 2003). In the dislodgeable residue study, methidathion was detected in citrus at up to 15 ng/cm² on leaves, with detectable residues through 7 days after application (MRID 00131096). No data are available regarding photo-oxidation in air of volatilized methidathion to its oxon forms, which could be an important route of transformation, as demonstrated by air monitoring data that find both methidathion and methidathion oxon at more than 20 km away from any use location. There are no data to characterize the fate of methidathion oxon.

It should be noted that due to an issue in the laboratory study design, there could be additional degradates of toxicological concern that were not observed. In laboratory transformation studies, only the thiadiazole ring of the compound had radioactive labeling and so degradates containing the phosphate ester portion of the parent compound may not have been observed. In particular, major degradate GS-12956 is formed through cleavage of the C-N bond. Therefore, GS-12956 does not contain the OP moiety but the cleavage would result in a corresponding phosphate ester (phosphorodithioic acid, o-o-s-trimethyl ester) which retains the OP moiety and so can be assumed to be toxic. Given that GS-12956 is formed in amounts up to 66% of the applied radioactivity, it is likely that the corresponding phosphate ester is formed as well. Additionally, this phosphate ester degradate could also undergo oxidative desulfonation to lead to additional oxon transformation products that would not have been detected in laboratory studies and may be of toxicological concern as well.

2.3.1.4. Environmental Transport Properties

The K_{oc} model appears to be appropriate to describe methidathion adsorption. Based on measured K_{oc} values of 113 to 342 mL/g_{oc}, methidathion is classified as moderately mobile according to the FAO classification scheme.

A number of studies have documented atmospheric transport and re-deposition of pesticides from the Central Valley to the Sierra Nevada Mountains (Fellers et al., 2004, Sparling et al., 2001, LeNoir et al., 1999, and McConnell et al., 1998). Prevailing winds

blow across the Central Valley eastward to the Sierra Nevada Mountains, transporting airborne industrial and agricultural pollutants into the Sierra Nevada ecosystems (Fellers *et al.*, 2004, LeNoir *et al.*, 1999, and McConnell *et al.*, 1998). Several sections of critical habitat for the CLRF are located east of the Central Valley. The magnitude of transport via secondary drift depends on methidathion's ability to be mobilized into air and its eventual removal through wet and dry deposition of gases/particles and photochemical reactions in the atmosphere. The vapor pressure of methidathion (2.48×10^{-6} mm Hg) and its Henry's Law constant (3.97×10^{-9} atm m³/mol) would suggest that it has a low potential to volatilize from dry or moist soils or from water, but monitoring studies have detected methidathion and its oxon in air at distances intermediate and up to 20 km away from any use sites (Royce *et al.*, 1993; Aston and Seiber, 1997; Majewski, 2006). Given the potential toxicity of these residues, this route of transport could be significant for assessing the extent of ecological effects.

2.4.2 Mechanism of Action

Methidathion is an insecticide belonging to the organophosphate class of pesticides. The pesticide acts through inhibition of acetylcholinesterase and is used to kill a broad range of insects and mites. Organophosphate toxicity is based on the inhibition of the enzyme acetylcholinesterase which cleaves the neurotransmitter acetylcholine. Inhibition of acetylcholinesterase by organophosphate insecticides, such as methidathion, interferes with proper neurotransmission in cholinergic synapses and neuromuscular junctions (USEPA 2000).

Target pests include the peach twig borer, scale insects, artichoke plume moth, leafminers, spider mites, boll weevils, bollworms, lygus bug, whitefly, aphid, pear psylla, mealybugs, thrips, sunflower stem weevil, sunflower moth, sunflower seed weevil, sunflower midge, Banks grass mite, flea beetle, hornworm, tobacco budworm, codling moth and hickory shuckworm.

2.4.3 Use Characterization

2.4.3.1 Use Statistics

Methidathion is used as an insecticide on a variety of terrestrial food and feed crops and terrestrial non-food crops. As shown in **Figure 2.1**, U.S. Geological Survey (USGS) National Water Quality Assessment Program (NAWQA) data indicate that in 2002, methidathion was used on agricultural crops predominantly in California, Washington, southern Texas, Pennsylvania, and New Jersey. At that time, the use of methidathion on citrus fruit represented about 30% of the national use. Based on national usage data compiled by the Biological and Economic Analysis Division (BEAD) primarily from 2001 to 2006, on average, roughly 110,000 pounds of methidathion are applied annually to agricultural crops, 95% of it in California. These data show that usage is highest on almonds and oranges, with annual average applications to each of 20,000 lbs. a.i. The crop with the highest average percent crop treated with methidathion is artichokes (60%).

METHIDATHION - insecticide 2002 estimated annual agricultural use

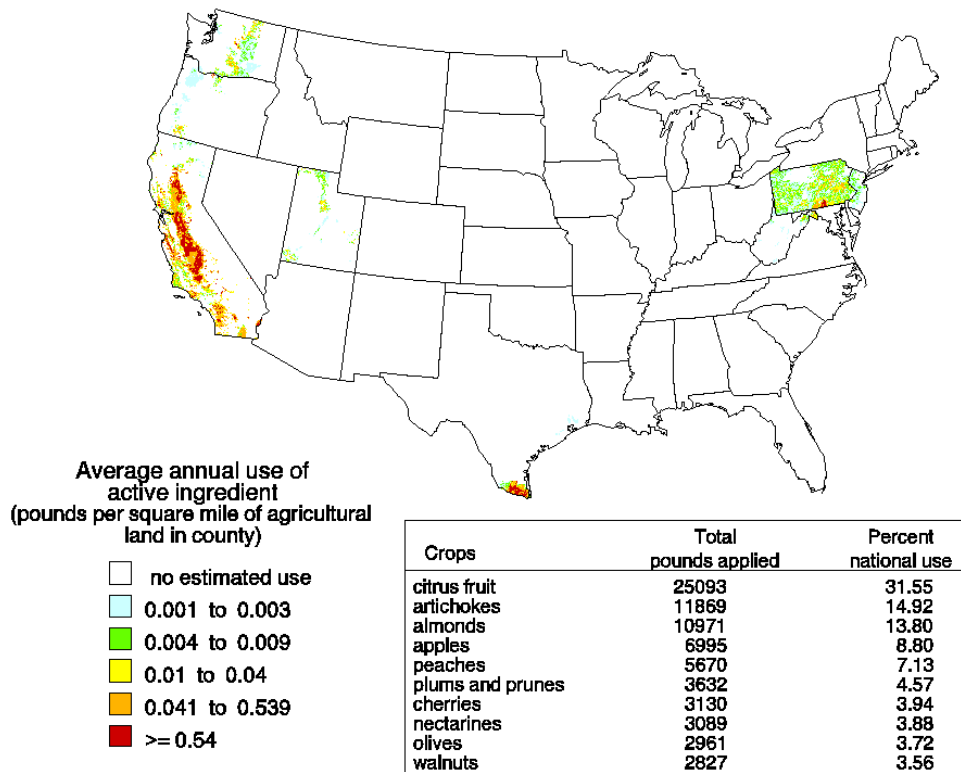


Figure 2.1. Map of Estimated Annual Agricultural Use of Methidathion in 2002.

Source: http://water.usgs.gov/nawqa/pnsp/usage/maps/show_map.php?year=02&map=m6037

Use data specific to California are available from the California Department of Pesticide Regulation's (CDPR) Pesticide Use Reporting (PUR) database, which includes every pesticide application made by professional applicators. The Biological and Economic Analysis Division (BEAD) summarized these data, from 1999 to 2006, to the county level by site, pesticide, and unit treated. Based on this analysis, an average of 82,031 lbs of methidathion was applied in California to an average of 52,823 acres per year. Use was at a maximum of 177,105 lbs in 1999 and then dropped by nearly half the following year, to 98,129 lbs, and remained relatively stable between 2003 and 2006 with average applications ranging from around 50,000 lb/year to 60,000 lb/year. From 1999-2006, methidathion was used in a total of 34 counties involving 41 different uses. Four counties accounted for 70% of the total lbs applied on average per county [Kern (25%), Tulare (20%), Monterey (14%), Fresno (11%)] (see **Figure 2.2**). Fruit orchards, including apple, apricot, cherry, nectarine, peach, pear, plum, and prune, accounted for approximately 30% of the total lbs applied per year in CA on average. Other major crops include almonds (23%), oranges (17%), and artichokes (14%). This analysis may not be entirely representative of current use patterns because labeled uses may have changed since these data were collected, and because it may also include misreporting. Complete data from the BEAD analysis of the CDPR PUR database are presented in **Appendix C**.

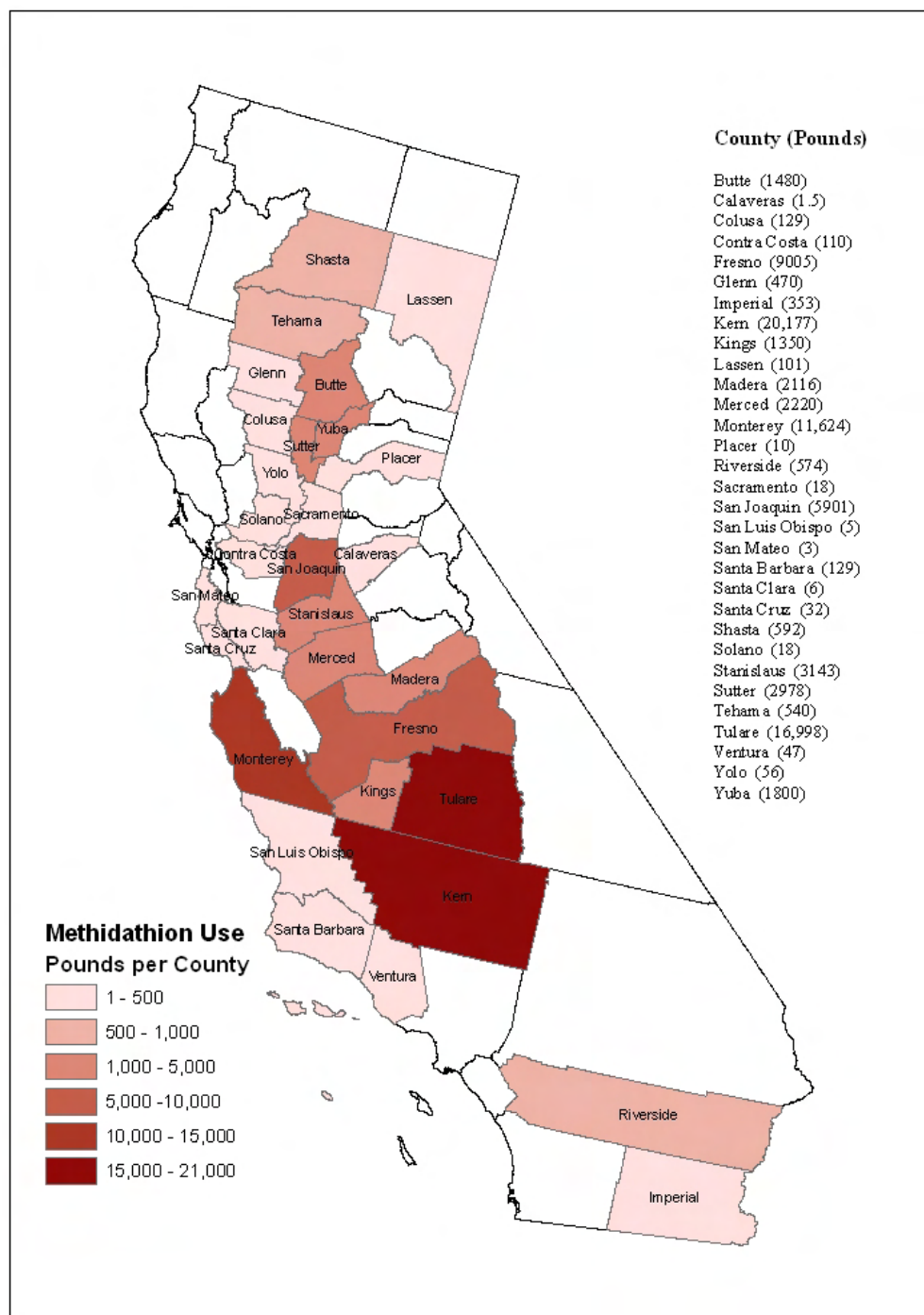


Figure 2.2 Average Pounds of Methidathion Applied Per County from 1996 - 2005

2.4.3.2 Application Rates and Methods

Analysis of labeled use information is the critical first step in evaluating the federal action. The current label for methidathion represents the FIFRA regulatory action; therefore, labeled use and application rates specified on the label form the basis of this assessment. The assessment of use information is critical to the development of the action area and selection of appropriate modeling scenarios and inputs. **Table 2.2** presents the uses and corresponding application rates considered in this assessment, based on a verification memorandum from SRRD (Dirk Helder, September 2, 2008).

Methidathion is formulated as a wettable powder in water-soluble bags (25% active ingredient) or as an emulsifiable concentrate (22% - 24% active ingredient). Application rates range from 0.25 to 5.0 lbs active ingredient/acre. Application is via foliar treatment and application equipment includes fixed wing aircraft, ground boom, air blast, low-pressure handwand or backpack sprayer, although based on application instructions for citrus, olives, and nursery stock, these uses are assumed to be applied through ground methods only. Labels have additional requirements in order to reduce spray drift. These include the buffers and other mitigation actions listed in Section 2.3.1 as well as a requirement for a spray with medium to coarse droplet size applied at a maximum boom height of 4 ft about the ground or canopy for ground spray and of 10 ft for aerial spray.

Table 2.2 Methidathion Uses Assessed for the CRLF				
Use	Max. Single Appl. Rate (lb ai/A)	Max. Number of Applications	Number of Crop Cycles Per Year	Minimum Application Interval (Days)
NON-FOOD/NON-FEED USES				
alfalfa	1	NS	2-9 ^a	NS
nursery stock ^b (includes ornamental herbaceous plants and ornamental woody shrubs and vines)	10 ^c	1/cc ^d	1 ^e	NA
FOOD/FEED USES				
alfalfa	1	1/cc	2-9 ^a	NA
almond	3	1/cc	1 ^e	EC: 14 WP: NS
apple	3	1/cc	1 ^a	NA
apricot	3	1/cc	1 ^e	NA
artichoke	1	8/cc	1 ^e	14
calamondin ^b	4	1/cc	1 ^e	NA
cherry	3	1/cc	1 ^e	NA
citron (citrus) ^b	4	1/cc	1 ^e	NA
citrus ^b	5	2/cc	1 ^e	45
citrus hybrids other than tangelo	4	1/cc	1 ^e	NA
clover	1	2/cc		NS
cotton (unspecified)	1	4/cc	1 ^a	5

Table 2.2 Methidathion Uses Assessed for the CRLF

Use	Max. Single Appl. Rate (lb ai/A)	Max. Number of Applications	Number of Crop Cycles Per Year	Minimum Application Interval (Days)
grapefruit	5	2/cc	1 ^a	45
kiwi fruit	2	1/1 yr	NA	NA
kumquat ^b	4	1/cc	1 ^c	NA
lemon ^b	5	2/cc	1 ^a	45
lime ^b	4	1/cc	1 ^c	NA
mango ^b	0.25	5/cc	1 ^c	21
nectarine	3	1/cc	1 ^a	NA
olive ^b	3	1/cc	1 ^c	NA
orange	5	2/cc	1 ^a	45
peach	3	1/cc	1 ^a	NA
pear	3	1/cc	1 ^a	NA
plum	3	1/cc	1 ^c	NA
prune	3	1/cc	1 ^c	NA
pummelo (shaddock)	4	1/cc	1 ^c	NA
safflower (unspecified)	0.5	3/1 yr	NA	7
sunflower	0.5	3/cc	1 ^c	7
tangelo ^b	4	1/cc	1 ^a	NA
tangerines ^b	4	1/cc	1 ^a	NA
timothy	1	NS	1 ^c	NA
walnut (english/black)	EC: 2 WP: 2	EC: 2/cc WP: 3/cc	1 ^c	EC: 2 WP: 2

NA = Not Applicable

NS = Not Specified on the label

^a U.S. EPA. 2007. Memo from Monisha Kaul (BEAD) to Melissa Panger (EFED). Subject: Maximum Number of Crop Cycles Per Year in California for Methomyl Use Sites. Dated February 28.^b Ground spray only^c The nursery stock application rate is reported on labels as 0.5 lb a.i./100 gal spray with no instructions for application on areal basis. Therefore, this rate is based on the general labels statement that methidathion should not be applied at greater than 10 lb a.i./A.^d cc = crop cycle^e Number of crop cycles per year as assumed by EFED.

2.5 Assessed Species

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

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

2.5.1 Distribution

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

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

The distribution of CRLFs within California is addressed in this assessment using four categories of location including recovery units, core areas, designated critical habitat, and known occurrences of the CRLF reported in the California Natural Diversity Database (CNDDDB) that are not included within core areas and/or designated critical habitat (see **Figure 2.2**). Recovery units, core areas, and other known occurrences of the CRLF from the CNDDDB 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 CNDDDB 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.

Other Known Occurrences from the CNDDDB

The CNDDDB provides location and natural history information on species found in California. The CNDDDB 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 CNDDDB.

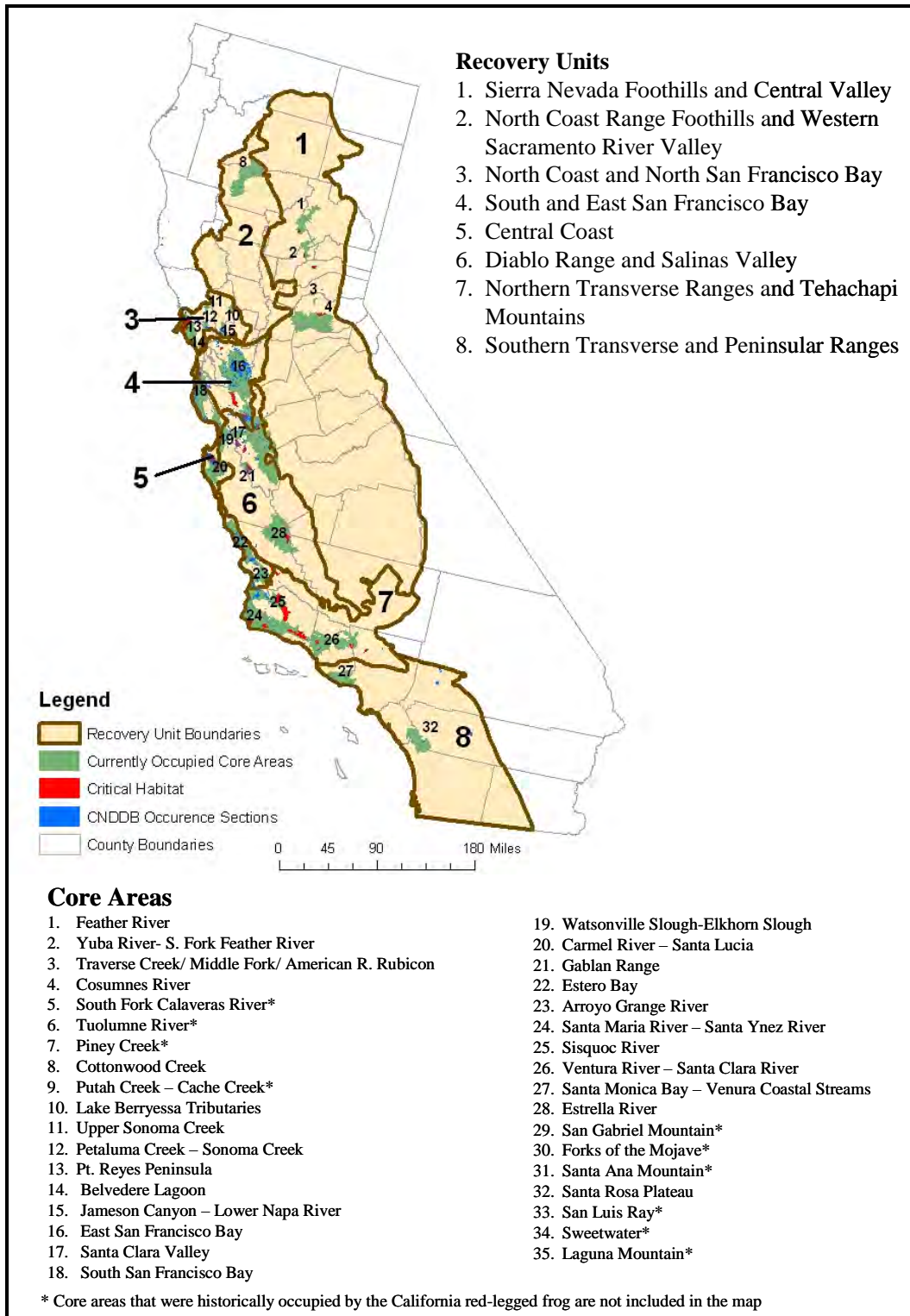


Figure 2.2 Recovery Unit, Core Area, Critical Habitat, and Occurrence Designations for CRLF

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 2.3** depicts CRLF annual reproductive timing.

Figure 2.3 – CRLF Reproductive Events by Month

J	F	M	A	M	J	J	A	S	O	N	D

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 (*Armadillidium 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 consist 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 **Attachment 1**.

‘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 modification with regard to actions carried out, funded, or authorized by a federal Agency. Section 7 requires consultation on federal actions that are likely to result in the destruction or modification of critical habitat.

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

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

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 modification standards for designated critical habitat (USFWS 2006). Activities that may destroy or modify critical habitat are those that alter the PCEs and jeopardize the continued existence of the species. Evaluation of actions related to use of methidathion 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) Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs.
- (3) Significant increase in sediment deposition within the stream channel or pond or disturbance of upland foraging and dispersal habitat 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.
- (4) 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.
- (5) Elimination of upland foraging and/or aestivating habitat or dispersal habitat.
- (6) Introduction, spread, or augmentation of non-native aquatic species in stream segments or ponds used by the CRLF.
- (7) Alteration or elimination of the CRLF's food sources or prey base (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 methidathion is expected to directly impact living organisms within the action area, critical habitat analysis for methidathion 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 methidathion 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 methidathion may be expected to have on the environment, the exposure levels to methidathion that are associated with those effects, and the best available information concerning the use of methidathion 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 methidathion. 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 methidathion, the following agricultural uses are considered as part of the federal action evaluated in this assessment:

- Alfalfa, almond, apple, apricot, artichoke, calemondin, cherry, citron, citrus, clover, cotton, grapefruit, kiwi, kumquat, lemon, lime, mango, nectarine, olive, oranges, peach, pear, plum, prune, pummelo, safflower, sunflower, tangelo, tangerine, timothy, and walnut.

In addition, the following non-food and non-agricultural uses are considered:

- Nursery stock, ornamental and herbaceous plants, woody shrubs and vines

Following a determination of the assessed uses, an evaluation of the potential “footprint” of methidathion 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 methidathion is presented in **Figure 2.4**.

The following land cover types were used for methidathion: cultivated crops, developed (low, medium and high intensity and open space), forest, open water, orchards and vineyards, pasture/hay, wetlands, turf and rights-of-way. More information regarding which specific uses are represented for each land cover types can be found in **Appendix D**.

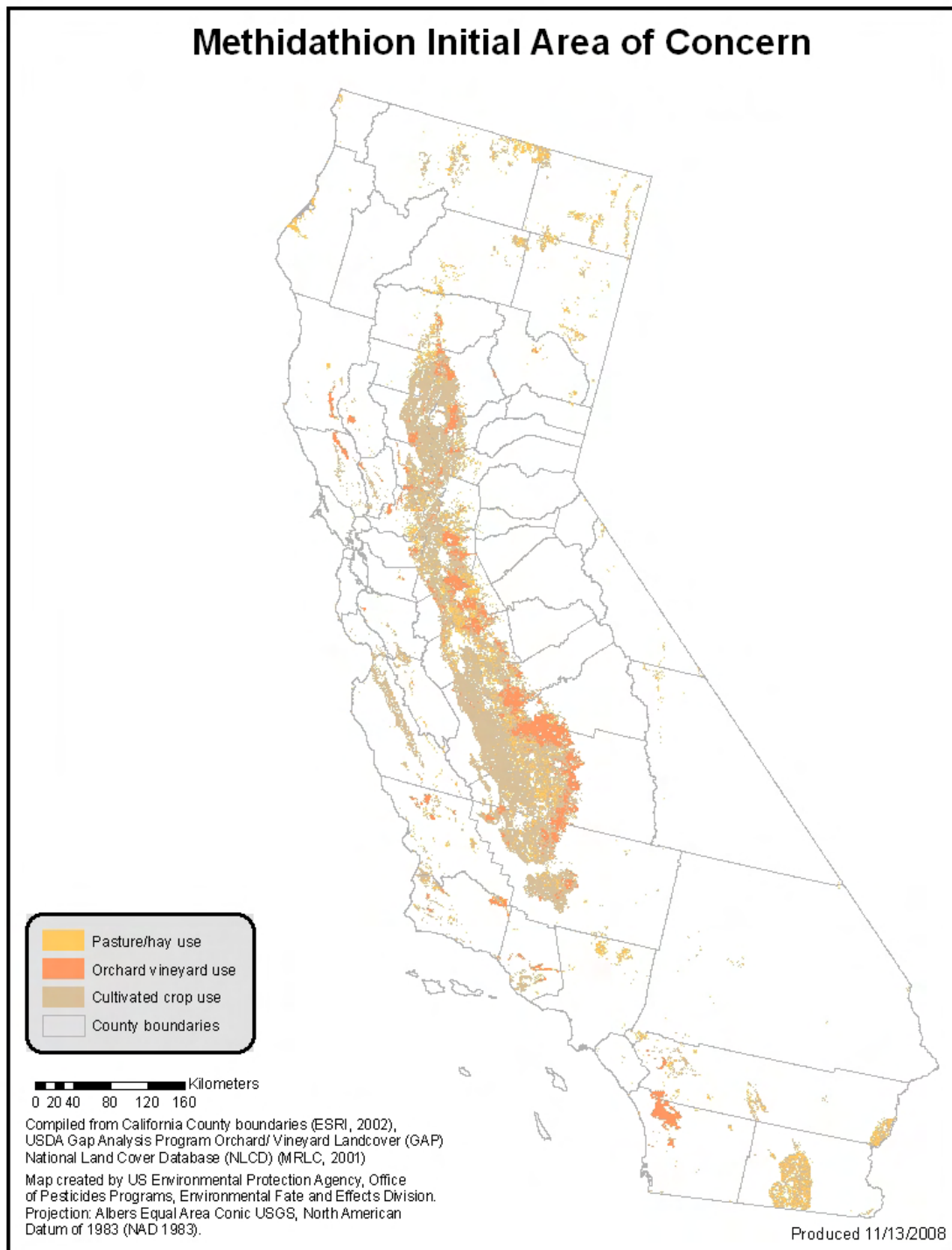


Figure 2.4 Initial area of concern, or “footprint” of potential use, for methidathion

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 toxic 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 effect concentration for a mammalian toxicity study (Yavuz et al., 2005; see Section 4.2.2 for study details), the spatial extent of the action area (i.e., the boundary where exposures and potential effects are less than the Agency's LOC) for methidathion 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).

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.”⁵ 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 methidathion (*e.g.*, runoff, spray drift, etc.), and the routes by which ecological receptors are exposed to methidathion (*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 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.

⁵ From U.S. EPA (1992). *Framework for Ecological Risk Assessment*. EPA/630/R-92/001.

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 methidathion is provided in **Table 2.3**.

Table 2.3 Assessment Endpoints and Measures of Ecological Effects	
Assessment Endpoint	Measures of Ecological Effects⁶
<i>Aquatic-Phase CRLF</i> (Eggs, larvae, juveniles, and adults) ^a	
<i>Direct Effects</i>	
1. Survival, growth, and reproduction of CRLF	1a. Freshwater fish 96-hour acute LC ₅₀ 1b. Freshwater fish chronic NOAEC
<i>Indirect Effects and Critical Habitat Effects</i>	
2. Survival, growth, and reproduction of CRLF individuals via indirect effects on aquatic prey food supply (i.e., fish, freshwater invertebrates, non-vascular plants)	1a. Freshwater fish 96-hour acute LC ₅₀ ; freshwater invertebrate 48-hr EC ₅₀ ; aquatic non-vascular plant acute EC ₅₀ 2b. Freshwater fish chronic NOAEC; freshwater invertebrate chronic NOAEC
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 3b. Non-vascular plant acute EC ₅₀
4. Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation	4a. Distribution of EC ₂₅ 's for monocots 4b. Distribution of EC ₂₅ 's for dicots
<i>Terrestrial-Phase CRLF</i> (Juveniles and adults)	
<i>Direct Effects</i>	
5. Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	5a. Avian acute LD ₅₀ 5b. Avian chronic NOAEC
<i>Indirect Effects and Critical Habitat Effects</i>	
6. Survival, growth, and reproduction of CRLF individuals via effects on terrestrial prey (i.e., terrestrial invertebrates, small mammals, and frogs)	6a. Terrestrial invertebrate acute LD ₅₀ ; mammalian acute LD ₅₀ 6b. Mammalian chronic NOAEC
7. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (i.e., riparian and upland vegetation)	7a. Distribution of EC ₂₅ 's for monocots 7b. Distribution of EC ₂₅ 's for dicots

^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 than exposure pathways on land.

^b According to Mayer, D. & C. Johansen. 1990. *Pollinator Protection: A Bee & Pesticide Handbook*. Wicwas Press. Cheshire, Conn. p. 161

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 methidathion 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.

⁶ All registrant-submitted and open literature toxicity data reviewed for this assessment are included in Appendix A.

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 methidathion effects data are available.

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

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

Measures of such possible effects by labeled use of methidathion on critical habitat of the CRLF are described in **Table 2.4**. Some components of these PCEs are associated with physical abiotic features (e.g., presence and/or depth of a water body, or distance between two sites), which are not expected to be measurably altered by use of pesticides. Assessment endpoints used for the analysis of designated critical habitat are based on the modification standard established by USFWS (2006).

Table 2.4 Summary of Assessment Endpoints and Measures of Ecological Effect for Primary Constituent Elements of Designated Critical Habitat^a	
Assessment Endpoint	Measures of Ecological Effect
<i>Aquatic-Phase CRLF PCEs</i> <i>(Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)</i>	
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 b. Distribution of EC ₂₅ values for terrestrial monocots c. Distribution of EC ₂₅ values for terrestrial dicots
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 b. Distribution of EC ₂₅ values for terrestrial monocots c. Distribution of EC ₂₅ values for terrestrial dicots
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	a. Freshwater fish 96-hour acute LC ₅₀ ; freshwater invertebrate 48-hr EC ₅₀ ; aquatic plant acute EC ₅₀ b. Freshwater fish chronic NOAEC; freshwater invertebrate chronic NOAEC
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (e.g., algae)	a. Most sensitive aquatic plant EC ₅₀
<i>Terrestrial-Phase CRLF PCEs</i> <i>(Upland Habitat and Dispersal Habitat)</i>	
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 b. Distribution of EC ₂₅ values for dicots c. Mammalian acute LD ₅₀ ; mammalian chronic NOAEC d. Freshwater fish 96-hour acute LC ₅₀ ; freshwater fish chronic NOAEC e. Avian acute LD ₅₀ ; avian acute LC ₅₀ ; avian chronic NOAEC f. Honey Bee acute LD ₅₀
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.

^b According to Mayer, D. & C. Johansen. 1990. *Pollinator Protection: A Bee & Pesticide Handbook*. Wicwas Press. Cheshire, Conn. p. 161

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

The labeled use of methidathion 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 methidathion and methidathion oxon release mechanisms, biological receptor types, and effects endpoints of potential concern. The conceptual models for terrestrial and aquatic exposures are shown in **Figures 2.5 and 2.6**, respectively, which

include the conceptual models for the aquatic and terrestrial PCE components of critical habitat. 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.

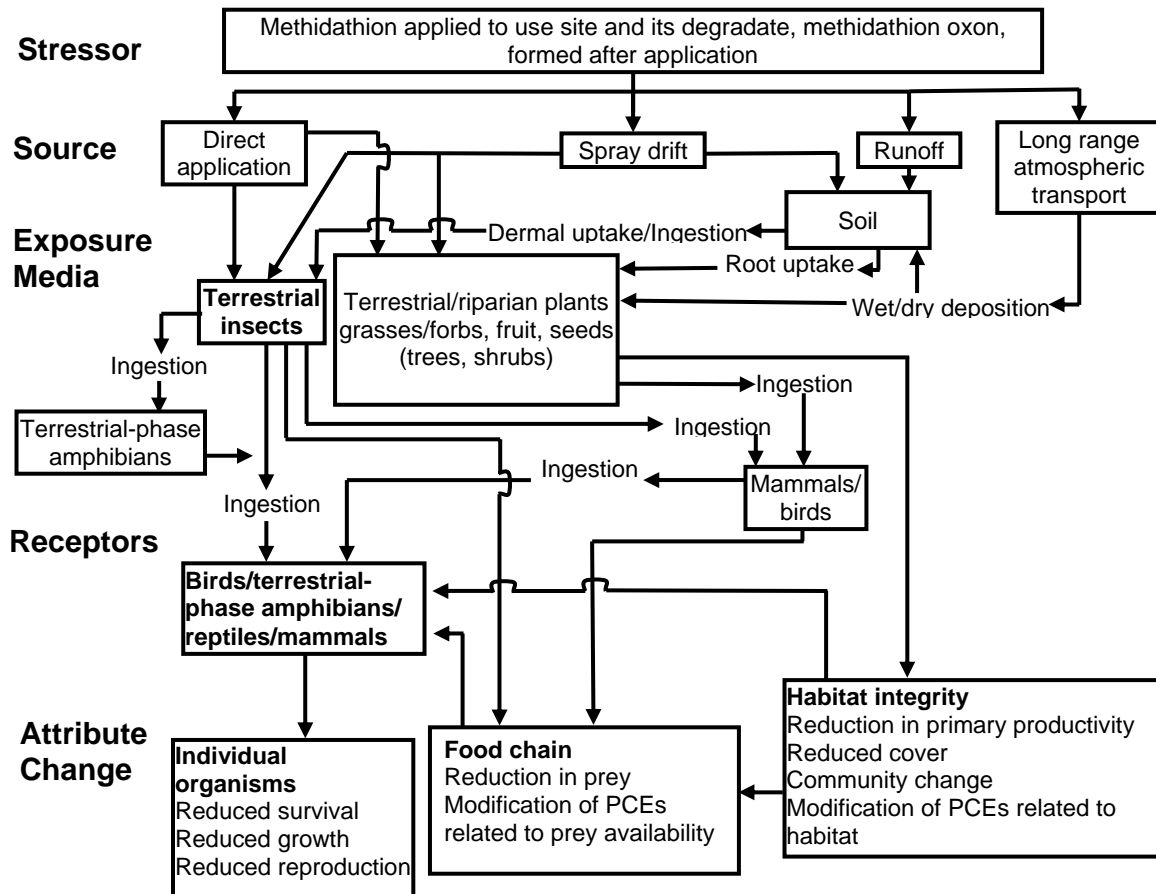


Figure 2.5 Conceptual Model for Methidathion Effects on Terrestrial Phase of the CRLF

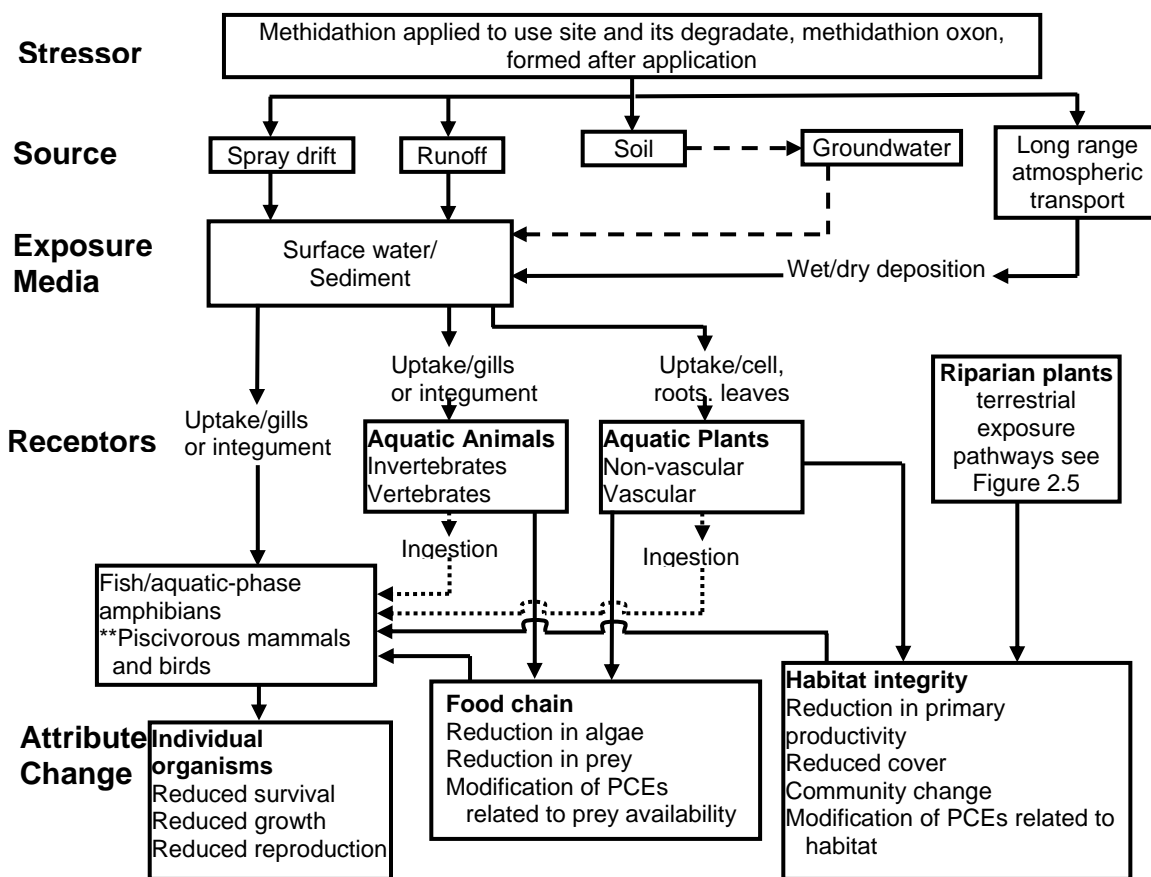


Figure 2.6 Conceptual Model for Methidathion Effects on Aquatic Phase of the CRLF

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 methidathion are characterized and integrated to assess the risks. This is accomplished using a risk quotient (ratio of exposure concentration to effects concentration) approach. 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 methidathion 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 methidathion along with available monitoring data indicate that runoff and spray drift are the principle potential transport mechanisms of methidathion to the aquatic and terrestrial habitats of the CRLF, and monitoring data indicate that long range atmospheric transport is a concern as well. In this assessment, transport of methidathion through runoff and spray drift is considered in deriving quantitative estimates of methidathion exposure to CRLF, its prey and its habitats. Long range atmospheric transport is considered qualitatively.

Measures of exposure are based on aquatic and terrestrial models that predict estimated environmental concentrations (EECs) of methidathion 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.

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 methidathion that may occur in surface water bodies adjacent to application sites receiving methidathion 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 m³ volume) with no outlet. PRZM/EXAMS was used to estimate screening-level exposure of aquatic organisms to methidathion. 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.

Exposure estimates for the terrestrial-phase CRLF and terrestrial invertebrates and mammals (serving as potential prey) assumed to be in the target area or in an area exposed to spray drift are derived using the T-REX model (version 1.3.1, 12/07/2006). This model incorporates the Kenega nomograph, as modified by Fletcher *et al.* (1994), which is based on a large set of actual field residue data. The upper limit values from the nomograph represented the 95th percentile of residue values from actual field

measurements (Hoerger and Kenega, 1972). For modeling purposes, direct exposures of the CRLF to methidathion 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 methidathion 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 are derived using TerrPlant (version 1.2.2, 12/26/2006). This model uses estimates of pesticides in runoff and in spray drift to calculate EECs. EECs are based upon solubility, application rate and minimum incorporation depth.

The spray drift model AgDrift (v. 2.01) was used to assess exposures of terrestrial phase CRLF and its prey to methidathion deposited on terrestrial habitats by spray drift from ground and aerial applications. In addition to the buffered area from the spray drift analysis, the downstream extent of methidathion 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 toxicity of methidathion to birds is similar to or less than the toxicity to the terrestrial-phase CRLF. The same assumption is made for fish and aquatic-phase CRLF. Algae, aquatic invertebrates, fish, and amphibians represent potential prey of the CRLF in the aquatic habitat. Terrestrial invertebrates, small mammals, and terrestrial-phase amphibians represent potential prey of the CRLF in the terrestrial habitat. Aquatic, semi-aquatic, and terrestrial plants represent habitat of CRLF.

The acute measures of effect used for animals in this screening level assessment are the LD₅₀, LC₅₀ and EC₅₀. LD stands for "Lethal Dose", and LD₅₀ 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 LC₅₀ is the concentration of a chemical that is estimated to kill 50% of the test organisms. EC stands for "Effective Concentration" and the EC₅₀ 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. For non-listed plants, only acute exposures are assessed (*i.e.*, EC₂₅ for terrestrial plants and EC₅₀ for aquatic plants).

It is important to note that 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 methidathion, 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 methidathion risks, the risk quotient (RQ) method is used to compare exposure and measured toxicity values. EECs are divided by acute and chronic toxicity values. The resulting RQs are then compared to the Agency's levels of concern (LOCs) (USEPA, 2004) (see **Appendix E**).

For this endangered species assessment, listed species LOCs are used for comparing RQ values for acute and chronic exposures of methidathion directly to the CRLF. If

estimated exposures directly to the CRLF of methidathion 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 methidathion 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”. Further information on LOCs is provided in **Appendix E**.

2.10.2 Data Gaps

For environmental fate, there are no data gaps which will affect conclusions regarding potential environmental exposure to the parent compound, methidathion. Guideline aerobic and anaerobic aquatic metabolism studies have not been submitted, and the anaerobic soil metabolism study is not sufficient to determine a degradation half-life. These data would be useful to refine characterization of the environmental fate of methidathion, but they would not change quantitative exposure estimates. Submitted laboratory volatility and photodegradation in air were determined to be unacceptable. These studies would be necessary to quantitatively assess the potential for atmospheric transport of methidathion and methidathion oxon.

Several fate studies do not provide adequate identification of potential degradates of methidathion. None of the available laboratory transformation studies include labeling of the phosphate ester portion of the methidathion compound and so potentially toxic degradates would not have been observed. This may lead to increased risk but cannot be characterized based on existing data and so is addressed as an uncertainty.

For environmental effects, there are no aquatic or terrestrial plant data. Limited data from ECOTOX are discussed in the risk description section along with the risk conclusions for aquatic and terrestrial plants from assessments with other organophosphate pesticides.

There are no toxicity data for methidathion oxon. Toxicity information for other organophosphate insecticides (*e.g.*, diazinon, chlorpyrifos) indicate that oxon degradates may be considerably more toxic than the parent OP. Methidathion oxon has been detected in monitoring data and has been observed to form in field and laboratory studies.

Without toxicity data, risk from this degradate of toxicological concern is calculated based on assumptions about formation and toxicity.

Although not specifically a data gap, it is noted that the chronic NOAEC of 6.3 µg/L for the fathead minnow is similar to the acute 96-hour LC₅₀ of several other freshwater fish species: rainbow trout (6.6 to 14 µg/L), bluegill sunfish (2.2 to 9 µg/L), and goldfish (6.8 µg/L). A chronic NOAEC for any of these other species would likely be lower than the available fathead minnow NOAEC. Therefore, the estimation of risk to freshwater fish and aquatic-phase amphibians is based on the most conservative acute to chronic ratio from other organophosphates that have both an acute and chronic study for rainbow trout.

3. Exposure Assessment

Methidathion is formulated as a wettable powder in water-soluble bags (25% active ingredient) and emulsifiable concentrate (22% - 24% active ingredient). Application equipment includes fixed wing aircraft, ground boom, air blast, low-pressure handwand or backpack sprayer. Application is via foliar treatment, and annual application rates range from 1.0 to 10.0 lbs a.i./A. Risks from ground boom and aerial applications are expected to result in the highest off-target levels of methidathion due to generally higher spray drift levels. Ground boom and aerial modes of application tend to use lower volumes of application applied in finer sprays than applications coincident with sprayers and spreaders and thus have a higher potential for off-target movement via spray drift.

3.1 Label Application Rates and Intervals

Methidathion labels may be categorized into two types: labels for manufacturing uses (including technical grade methidathion and its formulated products) and end-use products. While technical products, which contain methidathion 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 insects. The formulated product labels legally limit methidathion's potential use to only those sites that are specified on the labels.

Currently registered agricultural and non-agricultural uses of methidathion within California are alfalfa, almond, apple, apricot, artichoke, cherry, citrus (calemondin, citron, grapefruit, kumquat, lemon, lime, orange, pummelo, tangelo, and tangerine), clover, cotton, kiwi, mango, nectarine, nursery stock (including ornamental herbaceous plants and ornamental woody shrubs and vines), olive, peach, pear, plum, prune, safflower, sunflower, timothy, and walnut. Application rates and information for these uses were reported in **Table 2.2**.

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 methidathion use and represent a 10 hectare field that drains into a 1-hectare pond that is 2 meters deep and has no outlet. PRZM/EXAMS modeling does not account for transport to groundwater followed by discharge to surface water as a possible route of aquatic exposure. In this assessment, 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, but this factor remains an uncertainty, and will be discussed as such in Section 6.1.3.

3.2.2 Model Inputs

3.2.2.1 Physical Properties and Environmental Fate Inputs

Methidathion environmental fate data were discussed previously and are listed in **Table 2.1**. Chemical-specific model input parameters for PRZM and EXAMS are based on these data and are listed in **Table 3.1**.

Table 3.1. PRZM/EXAMS Environmental Fate Inputs for Aquatic Exposure to Methidathion			
Fate Property	Value	Comment ¹	Source
Molecular Weight	302.3		
Henry's constant	3.97×10^{-9} atm m ³ /mol		USEPA, 1999
Vapor Pressure	2.5×10^{-6} mm Hg		USEPA, 1999
Solubility in Water	2500 mg/L	Measured value x 10	USEPA, 1999
Photolysis in Water	10 days		MRID 42081709
Aerobic Soil Metabolism Half-lives	17.5 days	Upper 90% confidence bound on the mean of two values	MRID 44545101, MRID 42262501
Hydrolysis at pH 7	48 days	Maximum of two values	MRID 42037701
Aerobic Aquatic Metabolism (water column)	35 days	Twice the aerobic soil metabolism half-life.	
Anaerobic Aquatic Metabolism (benthic)	Stable	No data available	

Table 3.1. PRZM/EXAMS Environmental Fate Inputs for Aquatic Exposure to Methidathion			
Fate Property	Value	Comment ¹	Source
K _{oc}	364 mL/g _{oc}	Mean of four values	MRID 00158529

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

Crop-specific management practices for all of the assessed uses of methidathion were accounted for in aquatic exposure modeling. Application rates and intervals, application dates, and crop scenarios used as modeling inputs are listed in **Table 3.2** and the justification for these inputs are described below.

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. The scenarios used in modeling were selected from the group of standard and CRLF-specific scenarios representing crops grown in California. For most methidathion 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. For sunflower, the California Corn scenario was selected because it represents a non-irrigated row crop. For safflower, the California Wheat scenario was used because it is an annual, broadleaf oilseed crop that can be grown in the small-grain production areas of California. For kiwi, the California Grape scenario was used because the kiwi is a woody vine grown primarily in the same counties as grapes. Some scenarios represent multiple uses, as listed in **Table 3.2**. For these uses, the application rate modeled is the highest from all uses within the group. Unless otherwise specified, the application method is aerial spray.

Application dates are not specified on product labels. For modeling, an estimated application window was developed based on label instructions, crop profiles maintained by the USDA (<http://www.ipmcenters.org/cropprofiles/GetCropProfiles.cfm>; accessed 12/08), historical use data from the California PUR dataset, and planting dates and precipitation data specific to each scenario. From within this window, multiple application dates were modeled using a multi-run tool in pe5 and the date leading to the most conservative EEC was used. For example, the distribution of applications seen in the CDPR PUR data indicates that methidathion is typically applied to olives between mid-October and mid-December, while the scenario precipitation data show that within this time period, late November has the highest precipitation and so the greatest likelihood of runoff. Using the pe5 multi-run tool for this window, an application date of December 1 was shown to lead to the most conservative aquatic exposure estimates.

Table 3.2. PRZM/EXAMS Crop-Specific Inputs for Aquatic Exposure to Methidathion					
Scenario	Uses Represented by Scenario	Date of first application	Application Rate (lbs a.i./A)	Number of Applications Per Crop Cycle	Application Interval (Days)
CA Alfalfa	Alfalfa, Clover Timothy	8/15	1	2	5 ^a
CA Almond	Almond, Walnut	1/7	2	3	2
CA Citrus ^b	Calemondin, Citron, Citrus, Grapefruit, Kumquat, Lemon, Lime, Orange, Pummelo, Tangelo, Tangerine	8/15	5	2	45
CA Citrus ^c	Mango	8/15	0.25	1	NA
CA Corn	Sunflower	1/7	0.5	3	7
CA Cotton	Cotton	7/10	1	4	5
CA Grapes	Kiwi	2/15	2	1	NA
CA Tree Fruit	Apple, Apricot, Cherry, Nectarine, Peach, Pear, Plum, Prune	12/18	3	1	NA
CA Olive ^b	Olive	12/1	3	1	NA
CA Row Crop	Artichoke	7/25	1	8	14
CA Wheat	Safflower	5/25	0.5	3	7
CA Nursery ^d	Nursery	1/22	10 ^c	1	NA

^a The application interval is not specified on the label. A 5 day interval was assumed to be conservative because the target pests are similar to those for cotton.

^b Application through ground spray only.

^c Mango was modeled separately from the other citrus uses because it is the lowest application rate of all labeled uses and so EECs were needed to provide characterization of the full range of exposures.

^d The nursery stock application rate is reported on labels as 0.5 lb a.i./100 gal spray with no instructions for application on areal basis. Therefore, this rate is based on the general labels statement that methidathion should not be applied at greater than 10 lb a.i./A.

For all uses, application was modeled as foliar applied (CAM = 2) with no removal of foliar pesticide after harvest (IPSCND = 3). Application efficiency inputs were set to the default values corresponding to each application method: 99% efficiency for ground applications (citrus, olives, and nursery stock) and 95% for all other aerial spray applications. Spray drift inputs were estimated using the AgDRIFT model in order to account for the effect of the labels' mitigation requirements, which include buffer zones of 25 to 150 ft between cropped areas and water bodies (see Section 2.3.1) and a medium to coarse droplet size. Using these inputs, AgDRIFT estimated spray drift of 0.6% of the applied active ingredient for ground applications and of 2% for aerial uses.

3.2.3 Results

PRZM/EXAMS EECs representing 1-in-10 year peak, 21-day, and 60-day concentrations of total toxic residues of methidathion in the aquatic environment are located in **Table 3.5**. All model outputs are included in **Appendix F**. Estimated aquatic exposures from terrestrial food and feed uses are highest for the CA Almond scenario, representing methidathion use on almonds and walnuts, with a peak EEC of 49.3 ug/L. Peak EECs for other agricultural crops ranged from 4.6 ug/L for alfalfa to 20.7 ug/L for sunflowers.

The non-food use in ornamental nurseries resulted in a peak EEC of 115.6 ug/L. This result is based on an application rate of 10 lb a.i./A, the maximum application rate reported in the general instructions, because labeled application rates specific to the nursery use are reported in terms of product per foot of plant, rather than per unit area, as modeling requires. Aquatic EECs for the non-food uses on alfalfa, clover, and timothy have a peak EEC of 4.6 ug/L, but these results may underestimate exposure because the modeled use only includes a single crop cycle, while these crops can be grown at up to nine crop cycles per year.

Table 3.3 Aquatic EECs (µg/L) for Methidathion Uses in California				
Scenario	Crops Represented	Peak EEC	21-day average EEC	60-day average EEC
CA Alfalfa	Alfalfa, Clover Timothy	4.6	3.4	2.5
CA Almond	Almond, Walnut	49.3	40.2	26.8
CA Citrus	Calemondin, Citron, Citrus, Grapefruit, Kumquat, Lemon, Lime, Orange, Pummelo, Tangelo, Tangerine	18.3	13.6	9.0
CA Citrus	Mango	0.45	0.34	0.22
CA Corn	Sunflower	20.7	16.2	10.9
CA Cotton	Cotton	13.0	9.3	6.4
CA Grapes	Kiwi	6.6	5.0	3.2
CA Tree Fruit	Apple, Apricot, Cherry, Nectarine, Peach, Pear, Plum, Prune	15.6	9.2	5.0
CA Olive	Olive	15.2	11.7	7.6
CA Row Crop	Artichoke	16.0	12.1	8.7
CA Wheat	Safflower	13.6	10.4	6.6
CA Nursery	Nursery	115.6	84.8	54.3

3.2.4 Existing Monitoring Data

3.2.4.1 USGS NAWQA Surface Water Data

The USGS NAWQA surface water dataset in California includes 322 samples collected between 2001 and 2007 at 15 sites, all but one in the San Joaquin and Sacramento study areas. Six of the sites are located on Mustang Creek in Merced County in an area with landcover classified as “other, ” and the remaining sites represent a mix of landcover classes. Methidathion was detected 13 times at 7 sites, but more than half of the detections (8 detections, 0.01 to 0.04 ppb) were measured in one 24 hour period at two sites on Mustang Creek. At these and other Mustang Creek sites, sampling was conducted in winter months with multiple sampling per day, with the detections occurring in February, 2004. The other nine sites were generally sampled between 30 and 60 times, biweekly or bimonthly for one to several years. Methidathion was detected at four of these sites at levels of 0.1 to 0.31 ppb. Limits of quantitation (LOQ) were 0.006 ug/L to 0.009 ug/L. The sampling did not include analysis for any of the degradates of methidathion.

3.2.4.2 USGS NAWQA Groundwater Data

The NAWQA groundwater California dataset included 271 samples from 223 wells analyzed for methidathion between 2001 and 2006, with no detections. Detection limits were 0.0058 to 0.0087 ug/L. Most of the samples (77%) were in the San Joaquin study area, primarily in Fresno, Merced, and Stanislaus Counties. The remaining sites were in the Sacramento study area with a few in the Santa Ana study area as well. 30 sites in Sacramento county were classified as urban landcover, while the remaining samples were about half classified as agricultural landcover and half classified as mixed or other. No samples were analyzed for methidathion degradates.

3.2.4.3 California Department of Pesticide Regulation (CDPR) Surface Water Data

CDPR’s monitoring dataset for methidathion includes 4,044 samples from 135 sites, most targeted to agricultural use areas, although specific land cover information is not available. The sampling was conducted for 19 different studies, primarily in the Sacramento and San Joaquin River drainages, although there was no sampling in the southern San Joaquin Valley (Fresno, Kern, and Tulare Counties), which is the area with the greatest methidathion use (see Figure 2.2). More than half of the samples (58%) were collected at five sites. They are primarily from two studies which were targeted to OP use areas and that had daily to weekly sampling over several years. These studies have implications more specific to methidathion and so the results will be discussed separately in more detail below.

The remainder of the sampling included 1,694 samples from 130 sites, with approximately one-third of the samples (28%) from the Sacramento Valley and another 44% in the northern San Joaquin Valley. LOQs were between 0.0028 ug/L to 0.10 ug/L. In the Sacramento Valley, methidathion was detected in 46 out of 472 samples, with a maximum detection of 15.1 ug/L in Sutter County, and an average across all detections of 1.2 ug/L. In the San Joaquin Valley, there were 30 detections out of 738 samples, with the the maximum detection of 2.4 ug/L found in San Joaquin County and the average of 0.3 ug/L. Additionally, there were 5 detections at two sites in Orange County at concentrations of 0.05 ug/L to 0.17 ug/L. Acute toxicity endpoints for fish and aquatic invertebrates are 2.2 ug/L and 3.0 ug/L, respectively, so monitoring data demonstrate the potential for surface water concentrations in excess of these endpoints. Although Monterey is one of the highest use counties, methidathion was not detected in any of the 144 samples taken there, which were mostly from sites on waterways dominated by summer agricultural runoff. Overall, although only 35% of the total samples were collected in January or February, 90% of the detections (72 samples) were measured in those months. This reflects the fact that the typical use period of methidathion on fruit and nut orchards is in the winter dormant season.

Two studies monitored for methidathion oxon in surface water as well as for the parent. There were no detections of the oxon in either study, even though in some cases methidathion had been detected in samples collected at the same time. In most cases, the limit of quantitation was 0.05 ug/L. One study included 450 samples, collected weekly to monthly at 7 sites, with no detections (Ganapathy, 1997). At one Merced County site in this study, methidathion had been detected in each of 4 samples collected weekly in January and February. The second methidathion monitoring study included 290 samples at 26 sites, primarily in Stanislaus and Merced County (Ross et al, 2000). Methidathion had been detected at 7 sites in this study area, at concentrations up to 12.4 ug/L. There were no detections of methidathion oxon sampled at the same times.

Studies targeted to organophosphate pesticide use included extensive monitoring at several sites on Orestimba Creek, a tributary of the San Joaquin River located in Stanislaus County in an area expected to be vulnerable to runoff due to local soil and hydrology conditions. Agriculture in the area includes almond and walnut orchards as well as alfalfa. The sampling sites were selected primarily due to expected use of diazinon and chlorpyrifos, but methidathion is used in this area as well. In one study (Poletika & Robb, 1998), daily samples were taken at three sites on Orestimba Creek between June, 1996 to April, 1997. Methidathion was detected in 58 of 967 samples at levels from 0.001 ug/L to 0.331 ug/L, with an average across detected samples of 0.035, and based on an LOQ of 0.002 ug/L. There were 9 detections at the upstream site, with a maximum of 0.023 ug/L and mostly occurring in the first week of November, 1996. The midstream location had 22 detections, up to 0.078 ug/L, and the downstream site near the confluence with the San Joaquin River had 27 detections of up to 0.331 ug/L. Both sites had multiple detections in late July and early August. The highest peak occurred at the downstream site on March 20 and had decreased to undetectable levels by April 12. The study report indicates that, according to data from the county agricultural commissioner, five applications of methidathion were made in fields in the drainage during the sampling

period. However, the timing of the detections did not correspond with any of the reported pesticide applications, so there were either unreported applications or alternative sources of runoff. Additional monitoring at the downstream site on Orestimba Creek was conducted by CDPR, with 136 samples collected, primarily during winter months in 1997 to 2002. There were four detections of 0.06 ug/L to 2.14 ug/L (CDPR, metadata reference 10).

Another study conducted by the USGS included long term monitoring at two sites draining to the San Francisco Bay (Maccoy, et al, 1995). Unlike the previously discussed study, which was in a small water body local to use areas, the sites sampled here are near outlets of large agricultural watersheds, one site on the Sacramento River and downstream of most other riverine inputs in the Sacramento Valley, and one on the San Joaquin River and downstream of most other riverine inputs in the San Joaquin Valley. The San Joaquin Valley is the area of highest methidathion use and the Sacramento Valley has substantial use as well, so these locations represent high potential for methidathion exposure. On the San Joaquin River near Vernalis, sampling went from January, 1991 to April, 1994, and methidathion was detected 49 times out of 515 samples. All detections occurred in January or February at concentrations of 0.028 ug/L to 0.802 ug/L. On the Sacramento River, sampling began in May, 1991 and methidathion was detected in 23 of the 562 samples taken, all in February. In 1993, methidathion was detected in 16 of 23 samples collected in February at levels from 0.03 ug/L to 0.21 ug/L, and in 1994, 7 of the 17 samples collected in February had methidathion, concentrations of 0.04 ug/L to 0.7 ug/L.

3.2.4.4 Atmospheric Monitoring Data

There are several studies available which monitor for methidathion and methidathion oxon in air, fog, and/or rain onsite and at intermediate and long range distances from treated fields. A literature review by Majewski (1996) found three studies which tested air and fog and two studies which tested fog only for a total of 11 sites tested in California, both in the San Joaquin Valley and on the coast near Monterey. At eight of the sites, monitoring was also conducted for methidathion oxon. Methidathion was detected in both matrices at all 11 locations in all samples tested, while methidathion oxon was detected at 7 locations (all but one of those monitored) and in most of the samples tested. Detected concentrations of methidathion in air ranged from 0.01 ng/m³ to 23.8 ng/m³ and in fog from 4 x 10⁻⁵ ug/L (0.04 ng/L) to 15.5 ug/L. This maximum concentration in fog exceeds the effects levels for fish and aquatic invertebrates. Concentrations for methidathion oxon were not reported. Majewski attributes the high detection frequency to the fact that most sampling was done in or near orchards, and the fact that most samples were collected in January, during the typical use period. It is likely, then, that at least some of these detections were the result of spray drift rather than atmospheric transport. The study near Monterey, however, was conducted in September and states that at least one of the three sites was in a non-agricultural area, so there is the possibility that longer range transport occurred. The frequency of detection of the oxon indicates that oxidation was occurring, either in the air directly or in soil or on plant surfaces followed by volatilization.

Another air monitoring study was conducted for the California DPR in support of their Toxic Air Contaminant Program (Ross, 1991). In Tulare County, an area with high methidathion use on oranges, air sampling was conducted at five sites, four within a quarter mile of orange groves. The fifth site was intended to be used as background and was located in a downtown location with no orange groves in the immediate area. At each site, 17 samples per site were taken from June 27, 1991 to July 25, 1991, the time of peak use locally. Samplers were placed on the roof of a building or in an open area and collection occurred over a 23 to 25 hour period. All sites, including the intended background site, had samples with measured concentrations above the level of detection (LOD) for both methidathion (16 samples; LOD = 10 ng/m³) and methidathion oxon (27 samples; LOD = 30 ng/m³). Methidathion was detected above the LOQ in 7 samples (32 to 78 ng/m³) at two sites and methidathion oxon was detected in 7 samples (11 to 120 ng/m³) at 4 sites, with the lowest detection found at the background site. Methidathion oxon was detected more frequently and at higher concentrations than the parent compound, including detections at a site with no use in the immediate area, indicating that transformation occurs and that there is a potential for off-site atmospheric transport.

Vogel et al. (2008) monitored for methidathion in rain at a local and intermediate scale at a site in the Merced River basin where a variety of crops, including grapes, almonds, and corn, were grown. Between February and April of 2003 and December 2003 and April 2004, methidathion was measured for in 23 single event rain samples at lower, middle, and upper positions in the watershed. Detections above the limit of quantitation of 0.006 ug/L were found in 11 samples, with the highest detection of 0.69 ug/L found on December 16, 2003. Seven samples had detections between 0.01 ug/L and 0.1 ug/L and the other three were <0.01 ug/L. The detections were presumed to be the result of atmospheric transport because while the highest detection occurred before spraying began in local orchards, during that time methidathion was used on crops at the intermediate and broad scales. No sampling for methidathion oxon was conducted.

Another study by Aston and Seiber (1997) showed the most clear evidence of long range transport of both methidathion and its oxon degradate with detections in air at up to 30 km from any agricultural site and on pine needles 22 km away from use areas. In May through October of 1994, Aston and Seiber (1997) monitored for methidathion and methidathion oxon at three sites near Sequoia National Park. Biweekly samples of air and of pine needles were taken at three sites; the first was located in Tulare County next to a citrus grove treated with methidathion and within an area of high citrus production, the second was at a site within Sequoia National Park that was 22 km east of the nearest agricultural spraying, and the third site at an elevation on 1920 m was another 10 km away to the northeast. Each air sample represented air collected over a 24 h period, and pine needles were collected from potted pines placed at each site. The flow of air from the agricultural areas of the Central Valley to the two long range sites was reported to be unobstructed.

Methidathion was detected in air in every sample at the citrus grove location and concentrations and detection frequency decreased with distance. Methidathion oxon was also detected in all samples at the citrus grove and it was more persistent with distance

than the parent. One week following spraying in the onsite grove, methidathion was detected in air at the highest concentration of 17.0 ng/m³. Concentrations at the four sampling events prior to spraying ranged from 1.1 ng/m³ to 15.0 ng/m³ and in the four following sampling events air concentrations ranged from 0.4 ng/m³ to 2.7 ng/m³. Over the sampling period, methidathion oxon was detected onsite at levels from 0.28 ng/m³ to 10.0 ng/m³, with the highest concentration at the initial sampling period, followed by steady decline interrupted by a peak after spraying. At the intermediate site, methidathion was detected only in the initial sampling, June 7, at 0.23 ng/m³. The oxon degradate was detected on the first date as well, at 0.66 ng/ng/m³, and declined over the next month when it was detected a third time, at 0.21 ng/m³. At all but one of the 5 final sampling events, methidathion oxon was detected but it was either below the limit of quantitation (0.17 ng/m³) or it was not quantifiable due to inconsistencies in duplicate samples. At the far site, methidathion was reported as being unquantifiable in all samples and methidathion oxon was detected at 0.21 ng/m³ on June 20 and was undetected or unquantifiable in all other samples.

In pine needles, both methidathion and methidathion oxon were detected in all samples at the citrus grove site but only the oxon degradate was detected at the first site in Sequoia National Park and neither was detected at the further site. Next to the citrus grove, methidathion concentrations ranged from approximately 50 ng/g to 65 ng/g and methidathion oxon from approximately 15 ng/g to 150 ng/g (values estimated based on charts presented in the study report). The highest methidathion concentration was measured a week after spraying. Local spray drift is unlikely to be the cause because the pots had been removed before spraying and were not returned for 48 hours, although spray drift from other fields may be a possibility if they were treated at the same time. At this site, methidathion oxon was highest on September 18. At the intermediate site, there were no detections of methidathion, but methidathion oxon was detected at the three sampling events between July 11 and August 8 at levels from approximately 8 ng/g to 50 ng/g. There were no detections of either compound on pine needles at the furthest site.

3.3 Terrestrial Animal Exposure Assessment

3.3.1 Modeling Approach

T-REX (Version 1.3.1) is used to calculate dietary and dose-based EECs of methidathion 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 this assessment, spray applications of methidathion are considered, as discussed in below.

Terrestrial EECs for foliar formulations of methidathion were derived for the uses summarized in **Table 3.4**. In an effort to be consistent with the aquatic exposure assessment, the same uses and rates were modeled for terrestrial exposure estimation (see **Table 3.2**). For terrestrial exposure estimation purposes, a half-life of 6.6 days was used as an input parameter based the 90% upper bound of the total foliar dissipation half-life

values, 3.5 and 5.0 days, on alfalfa (Willis and McDowell, 1987). Use specific input values, including number of applications, application rate and application interval are provided in **Table 3.4**. An example output from T-REX is available in **Appendix G**.

Table 3.4 Input Parameters for Foliar Applications Used to Derive Terrestrial EECs for Methidathion with T-REX			
Uses	Application Rate (lbs a.i./A)	Number of Applications Per Crop Cycle	Application Interval (Days)
Artichoke	1	8	14
Citrus	5	2	45
Clover	1	2	7 (assumed)
Cotton	1	4	5
Kiwi	2	1	NA
Nursery	10	1	NA
Safflower, Sunflower	0.5	3	7
Tree Fruit, Olives, Almonds	3	1	NA
Walnut	2	3	2

T-REX is also used to calculate EECs for terrestrial insects exposed to methidathion. 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 terrestrial invertebrates. Available acute contact toxicity data for bees exposed to methidathion (in units of $\mu\text{g a.i./bee}$), are converted to $\mu\text{g a.i./g}$ (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 methidathion 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 mammalian 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 3.5**). Dietary-based EECs for small and large insects reported by T-REX as well as the resulting adjusted EECs are available in **Table 3.6**. An example output from T-REX v. 1.3.1 is available in **Appendix G**.

Table 3.5 Upper-bound Kenega Nomogram EECs for Dietary- and Dose-based Exposures of the CRLF and its Prey to Methidathion				
Use	EECs for CRLF		EECs for Prey (small mammals)	
	Dietary-based EEC (ppm)	Dose-based EEC (mg/kg-bw)	Dietary-based EEC (ppm)	Dose-based EEC (mg/kg-bw)
Artichoke	175	200	312	297
Citrus	681	776	1211	1154
Clover	200	227	355	339
Cotton	290	330	516	492
Kiwi	270	308	480	458
Nursery	68	77	120	114
Safflower, Sunflower	115	131	205	196
Tree Fruit, Olive, Almonds	405	461	720	686
Walnut	666	759	1184	1129

Table 3.6 EECs (ppm) for Indirect Effects to the Terrestrial-Phase CRLF via Effects to Terrestrial Invertebrate Prey Items		
Use	Small Insect	Large Insect
Artichoke	175	19
Citrus	681	76
Clover	200	22
Cotton	290	32
Kiwi	270	30
Nursery	68	8
Safflower, Sunflower	115	13
Tree Fruit, Olive, Almonds	405	45
Walnut	666	74

3.3.2 Field Studies

In a field study on California citrus (Brewer et al., 1998; MRID 44806601), methidathion was applied to foliage by air blast spray equipment for one application at 10.0 lb ai/A. Immediately after application the residues on hulled millet seed ranged from 248 to 305 (mean 276) ppm, and residues on crickets ranged from 22 to 59 (mean 40) ppm. T-REX model estimates – 150 ppm for seeds, 1350 ppm for small insects, and 150 ppm for large insects – are similar to actual field residues, and in the case of residues on seeds, the model may underestimate exposure. It is noted, however, that although it may underestimate potential risks to seed eaters, this dietary food item is not used in the assessment, and the foliage data did not suggest an underestimation of exposures.

3.4 Terrestrial Plant Exposure Assessment

Since there are no terrestrial plant toxicity data available, exposures were not quantitatively estimated. See Section 5.2 for a qualitative discussion regarding the potential effects of methidathion on CRLF via effects to terrestrial plants.

4. Effects Assessment

This assessment evaluates the potential for methidathion 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 methidathion.

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 information obtained on 24 August 2008. 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 methidathion.

Citations of all open literature not considered as part of this assessment because they were either rejected by the ECOTOX screen or accepted by ECOTOX but not used (e.g., the endpoint is less sensitive) 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 I**. **Appendix I** also includes a summary of the human health effects data for methidathion.

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 (EIIIS), are conducted to further refine the characterization of potential ecological effects associated with exposure to methidathion. A summary of the available aquatic and terrestrial ecotoxicity information, use of the probit dose response relationship, and the incident information for methidathion are provided in Sections 4.1 through 4.4, respectively.

There are no toxicity data available for methidathion oxon, a degradate of the parent methidathion; however, based on toxicity information for other organophosphate insecticides (*e.g.*, diazinon, chlorpyrifos), methidathion oxon may be considerably more toxic than methidathion. The potential for additional risk to the CRLF from exposure to methidathion oxon as a transformation product of applied methidathion will be characterized in the risk description (**Section 5.2**).

A detailed summary of the available ecotoxicity information for all methidathion degradates and formulated products is presented in **Appendix A**.

4.1 Toxicity of Methidathion to Aquatic Organisms

Table 4.1 summarizes the most sensitive aquatic toxicity endpoints for the CRLF, based on an evaluation of both the submitted studies and the open literature, as previously

discussed. A brief summary of submitted and open literature data considered relevant to this ecological risk assessment for the CRLF is presented below. Additional information is provided in **Appendix J**.

Table 4.1 Freshwater Aquatic Toxicity Profile for Methidathion				
Assessment Endpoint	Species	Toxicity Value Used in Risk Assessment	MRID	Study Classification
Acute Direct Toxicity to Aquatic-Phase CRLF	Bluegill Sunfish <i>Lepomis macrochirus</i>	LC ₅₀ = 2.2 µg/L	00011841	Supplemental
Chronic Direct Toxicity to Aquatic-Phase CRLF	Fathead minnow <i>Pimephales promelas</i>	NOAEC = 6.3 µg/L LOAEC = 12.0 µg/L post-hatch survival; growth	00015735	Acceptable
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Freshwater Invertebrates (i.e. prey items)	Water Flea <i>Daphnia magna</i>	LC ₅₀ = 3.0 µg/L	42081704	Acceptable
Indirect Toxicity to Aquatic-Phase CRLF via Chronic Toxicity to Freshwater Invertebrates (i.e. prey items)	Water Flea <i>Daphnia magna</i>	NOAEC = 0.66 µg/L LOAEC = 1.13 µg/L survival; number of young per female per reproductive day	42081707	Acceptable
Indirect Toxicity to Aquatic-Phase CRLF via Toxicity to Non-vascular Aquatic Plants	No data available			
Indirect Toxicity to Aquatic-Phase CRLF via Toxicity to Vascular Aquatic Plants	No data available			

Toxicity to aquatic fish and 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	
LC₅₀ (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 methidathion 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 methidathion to the CRLF. Effects to freshwater fish resulting from exposure to methidathion could 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

Available data indicate that methidathion is very highly toxic on an acute basis to three surrogate freshwater fish species (**Appendix J**). The acute 96-hour LC₅₀s for bluegill sunfish, rainbow trout, and goldfish range from 2.2 to 14 µg/L. The most sensitive endpoint, the bluegill sunfish 96-hour LC₅₀ of 2.2 (0.9 – 5.1) µg a.i./L (MRID 00011841), will be used to calculate RQs for direct effects to the aquatic-phase CRLF. The probit dose-response slope is 2.9 (1.9 – 4.0) for this study.

Two acute toxicity studies with the methidathion formulation 2E (25.2% a.i.) are also available for consideration in this risk assessment. These studies suggest that the tested formulation and technical grade methidathion are similarly toxic to bluegill sunfish and rainbow trout on an acute basis.

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

Chronic fish toxicity data for methidathion are limited to one early life stage study with fathead minnow as the test organism (MRID 00015730 or 45822701) (**Appendix J**). The 35-day flow-through study investigated the chronic effects following mean-measured concentration levels up to 12 µg ai/L. For survival, total length, and wet weight, the NOAEC, LOAEC, and EC₅₀ were 6.3, 12, and >12 µg ai/L respectively. It is important to note that the chronic NOAEC of 6.3 µg/L for the fathead minnow is similar to the acute 96-hour LC₅₀ of several other test species: rainbow trout (6.6 to 14 µg/L), bluegill sunfish (2.2 to 9 µg/L), and goldfish (6.8 µg/L). A chronic NOAEC for any of these other species would likely be lower than the available fathead minnow NOAEC. Therefore, instead of using the chronic NOAEC for fathead minnows, this assessment estimates a chronic NOAEC for rainbow trout using an acute to chronic ratio (ACR) calculation. Methidathion is an organophosphate insecticide. The EFED toxicity database, which contains the results from submitted studies that have been previously reviewed for scientific soundness, was accessed to derive an acute to chronic ratio of all organophosphate insecticides that have an acute LC₅₀ and an early life stage fish study for rainbow trout (the species which would most likely have chronic data). Nineteen organophosphates were found that have both an acute and chronic study for rainbow trout. The ACRs ranged from 5.4 for Terbufos to 140 for Dichlorvos. Using this range of ACRs, the estimated chronic NOAECs for rainbow trout tested with methidathion would range from 0.07 to 1.9 ppb. In order to provide the most conservative estimate for the chronic freshwater fish NOAEC for methidathion, the ACR of 140 is utilized. Using that value and an LC₅₀ of 10 ppb for rainbow trout tested with methidathion (MRID 00011841), the estimated chronic NOAEC for rainbow trout is **0.07 ppb**.

The following section presents a modification (for methidathion) of the methodology used in deriving a freshwater fish ACR for the acephate CRLF assessment (http://www.epa.gov/espp/litstatus/effects/redleg-frog/acephate/analysis_acephate.pdf). Of the 19 organophosphates found to have both acute and chronic rainbow trout data, 12 were evaluated for the ACR extrapolation Table 4.3.

The estimated fish (aquatic phase amphibians) chronic NOAEC for methidathion is derived as follows. The (methidathion) rainbow trout LC₅₀ used for the ACR calculation is 10 ppb ai. The largest acute-to-chronic ratio from the organophosphates is 140 for Dichlorvos. This ratio is used to calculate the final NOAEC for methidathion.

ACR ratio for Dichlorvos: 0.75 ppm (acute LC₅₀)/0.0052 ppm (chronic NOAEC) = 140

Estimated NOAEC for methidathion = $\frac{\text{LC}_{50}}{\text{NOAEC}} = \frac{10 \text{ ppb}}{\text{est. NOAEC}} = 140$

Estimated Trout NOAEC for methidathion = 10/140 = **0.07 ppb ai**

The table below (4.3) shows the inputs for the organophosphates that were considered for the methidathion ACR.

Acute to Chronic Table for Organophosphates

Table 4.3. Methidathion Acute to Chronic Ratio for Rainbow Trout NOAEC						
Chemical	96-hr LC₅₀ (ppm ai)^a	MRIDs	NOAEC (ppm ai)^a	MRIDs	ACR^a	Methidathion NOAEC (ppb ai)^a
Azinphos methyl	0.0088	03125193	0.00029	00145592	30	0.33
Coumaphos	0.89	40098001	0.012	43066301	76	0.13
Dichlorvos	0.75	43284702	0.0052	43788001	140	0.07
Dimethoate	7.5	TN 1069 ^b	0.43	43106303	17	0.59
Disulfoton	1.9	40098001	0.22	41935801	8.4	1.2
Fenamiphos	0.07	40799701	0.0038	41064301	18	0.56
Fenitrothion	2.0	40098001	0.046	40891201	43	0.23
Fenthion	0.83	40214201	0.0075	40564102	110	0.09
Fonofos	0.05	00090820	0.0047	40375001	11	0.91
Isofenphos	1.8	00096659	0.15	00126777	12	0.83
Phosmet	0.11	40098001	0.0032	40938701	33	0.30
Terbufos	0.0076	40098001	0.0014	41475801	5.4	1.9

^a Rounded to two significant figures

^b TN 1069 is test number for EPA's Animal Biology Lab, McCann, 1977

4.1.1.3 Freshwater Fish: Sublethal Effects and Additional Open Literature Information

In the bluegill sunfish acute toxicity study (MRID 00011841) used to calculate RQs, it was reported that test organisms in the highest exposure group (1000 µg/L) exhibited slight spastic motions and swam on their sides.

In the chronic fish early life stage study for methidathion (MRID 00015730 or 45822701), clinical signs of toxicity during the definitive toxicity test were not provided in the raw data. Total length and wet weight were the most sensitive endpoints. There were statistically significant reductions detected for both endpoints at the highest measured treatment level (12 µg ai/L) relative to the pooled control. The NOAEC and LOAEC for both endpoints were 6.3 and 12 µg ai/L, respectively.

There is one additional fish toxicity study available in the open literature for methidathion. The test organism, the common eel (*Anguilla anguilla*), appears to be about 3 orders of magnitude *less* sensitive than the bluegill sunfish, the surrogate organism used here to assess direct effects to the aquatic-phase CRLF. See **Appendix I** for more details.

4.1.2 Toxicity to Freshwater Invertebrates

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

Available data indicate that methidathion is very highly toxic on an acute basis to a surrogate freshwater invertebrate species, *Daphnia magna* (**Appendix J**). A daphnid acute toxicity study is available for methidathion technical grade active ingredient (TGAI) and for the methidathion formulation 2E (25.2% a.i.). The 48-hour LC₅₀ for methidathion (TGAI) is 7.2 µg a.i./L, and the LC₅₀ for methidathion formulation 2E (25.2% a.i.) is 11.9 µg product/L or 3.0 µg a.i./L. The most sensitive endpoint, 48-hour LC₅₀ of 3.0 µg a.i./L (MRID 42081704), will be used to calculate RQs for indirect effects to the aquatic-phase CRLF. The probit dose-response slope is the default of 4.5 (2–9) for this study because there are less than two concentrations at which the percent dead is between 0 and 100.

4.1.2.2 Freshwater Invertebrates: Chronic Exposure Studies

Chronic toxicity data are available for a common freshwater zooplankton, *Daphnia magna* (**Appendix J**). In a 21-day lifecycle study (MRID 42081707), survivorship was reduced by 97% at 1.1 µg a.i./L; the NOAEC (mortality) was 0.66 µg a.i./L. This endpoint will be used to calculate RQs in this assessment. There were no significant effects of methidathion on reproduction or growth in the study.

4.1.2.3 Freshwater Invertebrates: Open Literature Data

There are no freshwater invertebrate toxicity studies identified in the open literature for methidathion (**Appendix H**).

4.1.3 Toxicity to Aquatic Plants

There are no aquatic plant toxicity data for methidathion with which to assess the potential for indirect effects to the CRLF via effects on habitat, cover, and/or primary productivity or effects to the primary constituent elements (PCEs) relevant to the aquatic-phase CRLF.

4.2 Toxicity of Methidathion to Terrestrial Organisms

Table 4.4 summarizes the most sensitive terrestrial toxicity endpoints for 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.

Endpoint	Species	Toxicity Value Used in Risk Assessment	MRID	Study Classification
Acute Direct Toxicity to Terrestrial-Phase CRLF (LD ₅₀)	Mallard duck <i>Anas platyrhynchos</i>	LD ₅₀ = 6.7 mg/kg	00159201	Supplemental
Acute Direct Toxicity to Terrestrial-Phase CRLF (LC ₅₀)	Bobwhite quail <i>Colinus virginianus</i>	LC ₅₀ = 224 ppm	42081701	Acceptable
Chronic Direct Toxicity to Terrestrial-Phase CRLF	Mallard Duck <i>Anas platyrhynchos</i>	NOAEC = 10 ppm LOAEC = 30 ppm Number of normal hatchlings/live 3-week embryos	44381602	Acceptable
Indirect Toxicity to Terrestrial-Phase CRLF (via acute toxicity to mammalian prey items)	Rat <i>Rattus norvegicus</i>	LD ₅₀ = 12 mg/kg	00012714	Acceptable
Indirect Toxicity to Terrestrial-Phase CRLF (via chronic toxicity to mammalian prey items)	Rat <i>Rattus norvegicus</i>	NOAEL = 5 ppm LOAEL = 25 ppm Decreased mating index, decreased pup weight, tremors, decreased food consumption during lactation, transient decrease in body weight	40079812 40079813	Acceptable

Table 4.4 Terrestrial Toxicity Profile for Methidathion				
Endpoint	Species	Toxicity Value Used in Risk Assessment	MRID	Study Classification
		(males and females)		
Indirect Toxicity to Terrestrial-Phase CRLF (via acute toxicity to terrestrial invertebrate prey items)	Honey Bee <i>Apis mellifera</i>	LD ₅₀ = 0.236 µg a.i./bee	0036935	Acceptable
Indirect Toxicity to Terrestrial- and Aquatic-Phase CRLF (via toxicity to terrestrial plants)	<u>Seedling Emergence</u> Monocots	No data available		
	<u>Seedling Emergence</u> Dicots			
	<u>Vegetative Vigor</u> Monocots			
	<u>Vegetative Vigor</u> Dicots			

Acute toxicity to terrestrial animals is categorized using the classification system shown in **Table 4.5** (U.S. EPA, 2004). Toxicity categories for terrestrial plants have not been defined.

Table 4.5 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 methidathion; therefore, acute and chronic avian toxicity data are used to assess the potential direct effects of methidathion to terrestrial-phase CRLFs.

4.2.1.1 Birds: Acute Exposure (Mortality) Studies

Based on acute oral toxicity studies for several bird species, methidathion is categorized as moderately to very highly toxic to birds (**Appendix J**). The mallard duck was the most sensitive of the species tested, with an 8-day LD₅₀ of 6.7 (5.4 – 8.4) mg/kg methidathion (MRID 00159201). This endpoint will be used for risk estimation in this assessment. These data do not fit the probit dose-response model; thus, a default slope of 4.5 (2-9)

will be used to calculate the probability of individual effects to the terrestrial-phase CRLF.

Avian subacute dietary studies are available for methidathion (TGAI) and a formulation (40% a.i.) (**Appendix J**). Based on the available information, methidathion and the tested formulation appear to be similarly toxic to birds on a subacute dietary basis. The bobwhite quail (*Colinus virginianus*) was the most sensitive of the species tested, with an 8-day LC₅₀ of 224 (177 – 281) ppm a.i. (MRID 42081701). This endpoint will be used for risk estimation in this assessment. The probit dose-response slope was 8.7 (3.5 – 13.8).

4.2.1.2 Birds: Chronic Exposure (Growth, Reproduction) Studies

Several avian chronic toxicity studies are available for methidathion. The mallard duck study (MRID 44381602) reported the most sensitive chronic toxicity endpoint, which will be used for risk estimation. In this study, the one-generation reproductive toxicity of methidathion TGAI to 6-month-old mallard ducks was assessed over 140 days. Methidathion TGAI was administered to the birds in the diet at 0, 1, 10, and 30 ppm diet. The number of normal hatchlings/live 3-week embryos was significantly reduced (a ratio of 0.5 versus 0.61 or 18% less than the control ratio) at the highest treatment, 30 ppm (LOAEC); the NOAEC for this effect is 10 ppm. A significant increase in the number of eggs cracked was observed when compared to the control group at the 10 ppm dietary level. In addition, a decrease in the number of eggs not cracked/eggs laid was observed at the same dietary level (i.e., NOAEC = 1 ppm); however, this is not a relevant assessment endpoint for the CRLF because endpoints having to do with egg cracking are not relevant to frogs, whose eggs do not have shells. There were no apparent behavioral abnormalities or other treatment-related signs of toxicity on the parental generation.

4.2.1.3 Birds: Sublethal Effects and Additional Open Literature Information

In the mallard duck acute oral toxicity study that was used to calculate RQs (MRID 00159201), toxic symptoms included depression, reduced reaction to external stimuli, wing droop, convulsions, and salivation. In the bobwhite quail subacute dietary study used for risk estimation, toxic symptoms including depression, reduced reaction to external stimuli, wing droop, loss of coordination, lower limb weakness, ruffled appearance, prostrate posture, and loss of righting reflex were observed at and above 178 ppm a.i., and body weight gain and food consumption were reduced at levels above 316 ppm a.i.

In the mallard duck chronic toxicity study (MRID 44381602) used for risk estimation, methidathion technical was administered to the birds in the diet at 0 (control), 1, 10, and 30 ppm diet. None of the ducks showed symptoms of toxicity or behavioral abnormalities during the experiment. The number of eggs cracked was significantly increased in the 10 ppm (150%) and 30 ppm (86%) exposure concentrations. The number of eggs not

cracked/eggs laid was also significantly reduced in the 10 ppm and 30 ppm treatment groups. The number of normal hatchlings/live 3-week embryos was significantly reduced (5%) at the highest treatment, 30 ppm. Body weight of 14-day-old survivors was reduced (6%) at 10 ppm, but not at the higher treatment levels. Brain cholinesterase activity was significantly reduced in drakes at the 10 ppm treatment level during weeks 2, 10, and 20, and at the 30 ppm level during weeks 10 and 20. During week 20, hens showed a trend toward reduced brain cholinesterase activity at the 10 ppm and 30 ppm treatment levels.

Additional avian acute toxicity studies for methidathion were identified in the open literature (**Appendices H and I**). However, none of the open literature studies resulted in a more sensitive acute toxicity threshold than the 8-day mallard duck LD₅₀ of 6.7 mg/kg (MRID 00159201).

4.2.2 Toxicity to Mammals

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

The study used to expand the initial area of concern to define the action area examined the effects of subchronic methidathion administration on vascular wall damage (Yavuz et al., 2005; ECOTOX ref. 80451). Methidathion was administered by gavage for 5 days a week for 4 weeks at a dose level of 5 mg/kg/day. The levels of malondialdehyde (MDA), a biomarker for oxidative stress, were determined in the vascular tissue.

Histopathological examination was performed in the thoracic aortic tissue. The level of MDA was significantly higher, and cholinesterase activity was significantly lower in the methidathion group ($p < 0.01$). Subchronic methidathion administration led to irregular, prominent breaks and fragmentation of the elastic fibers in the aortic wall. Aortic lesions such as these are similar to the precursor lesion of a human aortic aneurysm. Since only one treatment level was tested, the LOAEC is 5 mg/kg/day, and a NOAEC was not defined. Therefore, a threshold for this effect could not be defined, and thus, this study was used to define the action area as the entire state of California for this assessment.

4.2.2.1 Mammals: Acute Exposure (Mortality) Studies

Available acute toxicity information suggests that methidathion is very highly toxic (Category I) to small mammals on an acute oral basis (**Appendices J and K**). The most sensitive endpoint, the acute rat (weanling) LD₅₀ of 12 mg/kg, will be used to estimate risk to the CRLF via indirect effects to mammals. The probit dose-response slope is assumed to be 4.5 (2–9) for this study because individual animal data are not readily available for this study.

4.2.2.2 Mammals: Chronic Exposure (Growth, Reproduction) Studies

Chronic mammalian reproduction toxicity studies are available for methidathion (**Table 9**). In a 2-generation reproduction study in rats (MRID 40079812, -13), rats were treated with 0 (control), 5, 25, or 50 ppm methidathion in the diet. The parental systemic NOAEC was 5 ppm and the LOAEC was 25 ppm, based on tremors and decreased food consumption during lactation, and decreased ovarian weight. In addition, there was also a slight decrease in body weight early in the F1 growth phase at 50 ppm. The reproductive NOAEC was 5 ppm and the LOAEC was 25 ppm based on a decreased mating index and a generalized indication of pup unthriftiness while nursing (e.g., decreased pup weight and an increased incidence of hypothermia with an appearance of starvation). In addition, there was an increase in stillbirths and decreased pup survival at birth and during lactation at the 50 ppm treatment level. An NOAEL of 5 ppm will be used for risk estimation in this assessment.

4.2.3 Toxicity to Terrestrial Invertebrates

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

4.2.3.1 Terrestrial Invertebrates: Acute Exposure (Mortality) Studies

Methidathion is classified as very highly toxic to bees, with an acute contact LD₅₀ of 0.236 µg/bee or 1.84 ppm (MRID 00036935). This endpoint will be used to quantitatively assess the risk to the CRLF via indirect effects to terrestrial invertebrates. The probit dose-response slope from this study was 9.06.

In addition, a residual toxicity study indicates that the RT₂₅ (i.e., the residual time to kill 25% of the tested population) is greater than 3 days when methidathion (Supracide 2E, 25.2% a.i.) is applied at a rate of 5 lbs a.i./A (MRID 42081708). That is, in addition to very high acute contact toxicity, field weathered spray residues of methidathion can result in significant honey bee mortality for more than several days.

4.2.3.2 Terrestrial Invertebrates: Open Literature Studies

There are several terrestrial invertebrate toxicity studies available in the open literature (**Appendices H and I**). However, none of these studies identified a more sensitive endpoint than the submitted honey bee toxicity study that determined an acute contact LD₅₀ of 0.236 µg/bee or 1.84 ppm (MRID 00036935).

4.2.4 Toxicity to Terrestrial Plants

Terrestrial plant toxicity data are used to evaluate the potential for methidathion to affect riparian zone and upland vegetation within the action area for the CRLF. Impacts to riparian and upland (i.e., grassland, woodland) vegetation could result in indirect effects

to both aquatic- and terrestrial-phase CRLFs, as well as modification to designated critical habitat PCEs via increased sedimentation, alteration in water quality, and reduction in of upland and riparian habitat that provides shelter, foraging, predator avoidance and dispersal for juvenile and adult CRLFs.

There are no registrant-submitted terrestrial plant toxicity data for methidathion with which to assess the potential for indirect effects to the aquatic- and terrestrial-phase CRLF via effects to riparian vegetation or effects to the primary constituent elements (PCEs) relevant to the aquatic- and terrestrial-phase CRLF. However, there is limited evidence in the open literature that methidathion has the potential to elicit phototoxic effects: Godfrey and Holtzer (1992; Ecotox ref. 64451) reported that methidathion significantly affected corn photosynthesis when applied at 0.5 lbs a.i./A, a rate that is lower than many of the currently registered rates. In addition, potential phytotoxic effects have been highlighted on methidathion labels; e.g., EPA SLN No. TX-050003 warns that methidathion application may result in phytotoxic effects, such as spotting, reddening, or chlorosis of the leaves, in certain sorghum varieties. In the absence of vegetative vigor and seedling emergence toxicity data, the potential risk to the CRLF via indirect effects to terrestrial plants is described in a qualitative manner (Section 5.2).

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 methidathion 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.

4.4 Incident Database Review

A review of the EIIS database for ecological incidents involving methidathion was completed on 08 September 2008. A total of two aquatic and six terrestrial incidents are reported, all of which occurred in California. The reported aquatic incidents occurred in 1976 and 2002; all of the reported terrestrial incidents occurred in 1994 and 1995. A brief description of each of the reported incidents is provided below. A complete list of the incidents involving methidathion is included as **Appendix L**.

4.4.1 Terrestrial Incidents

On January 18, 1994, one red-tailed hawk was found dead in Colusa County, California following application of methidathion to an unknown treatment site. No residue analysis was completed. The certainty index for this incident (I003351-003) is probable.

Following application of methidathion to an unknown treatment site, a red-tailed hawk was found dead on January 27, 1994, in Stanislaus County, California. Blood and footwash sample were collected and sent to the PIU (Department of Fish and Game Pesticides Investigation Unit (California)) for examination. Blood plasma cholinesterase and acetylcholinesterase levels were within the normal range. The results of the footwash showed detections of methidathion at 2.7 ppb. In addition, organophosphates including chlorpyrifos and diazinon were also detected in the footwash sample at respective concentrations of 0.2 ppb and 4.1 ppb. It is likely that the hawk was exposed by perching on substrates which had recently been coated with the detected pesticides. The certainty index for this incident (I005042-012) is probable.

On February 3, 1994, a red-tailed hawk was found entangled in a fence near an almond orchard in Merced County. The hawk was handed over to the Stanislaus Wildlife Care Center. Upon admission to the care center, blood and footwash samples were collected. The hawk was released following recovery from the injuries. Samples were sent to the PIU for examination. Plasma cholinesterase and acetylcholinesterase levels were found significantly below the normal range. Footwash analysis results show that methidathion was detected at a concentration of 2.7 ppb. Chlorpyrifos and diazinon were also detected in the footwash sample at respective concentrations of 0.7 ppb and 0.4 ppb. It appears that the hawk was affected due to exposure via perching on substrates, such as tree branches, which had recently been treated with these organophosphate pesticides. The certainty index for this incident (I05042-010) is highly probable.

A red-tailed hawk was recovered from an orchard on December 23, 1994 and taken to the Stanislaus Wildlife Care Center. The bird died one day later. A blood sample and the carcass were sent to the PIU for examination. The plasma cholinesterase level was low and the brain cholinesterase level was severely depressed. Feathers and footskin samples were analyzed. Methidathion was detected at a concentration of 0.7 ppm in the footskin. In addition, chlorpyrifos and diazinon were detected in both the feathers and footskin at concentrations ranging from 0.02 to 0.08 ppb. It is likely that all three organophosphates contributed to the death of this bird. The certainty index for this incident (I005042-015) is possible.

Following spraying of plum trees in Madera County, California with methidathion on Feb. 4, 1995 and spraying of a nearby location on the 5th and 6th, pollination hives were placed in the orchard on those same dates. Heavy fog was present the 4th and 5th, which limited foraging activity of the bees. Dead bees (number not specified) were found beginning Feb. 8 and continuing until Feb. 13. Residue analysis of the dead bees was completed, showing the presence of 2.6 ppm methidathion and 0.02 ppm diazinon. The Madera County authorities did not indicate any fault or liability, and attributed the bee kill to poor weather and too early an introduction of the hives. The certainty index for this incident (I001920-001) is possible.

The most recent terrestrial incident involving methidathion was reported on March 15, 1995. Following application of methidathion to an agricultural area, an unknown number of bees were killed in an unknown county in California. The incident was reported as accidental under the misuse category. No further information was provided in the incident report. No residue analysis was completed, and the certainty index for this incident (I005895-512) is probable.

4.4.2 Aquatic Incidents

On May 25, 1976, approximately 3,000 fish of unknown species were killed following an aerial application of methidathion to an agricultural area in Sacramento County, California. Methidathion had been aerially sprayed on a nearby seed clover field, and irrigation water from the field had entered Laguna Creek in the area directly upstream of the fish kill. The incident was attributed to accidental misuse of methidathion. Samples of the water collected from the field drain six days after the material was applied to the field showed methidathion concentrations of 60 ppb. It is presumed that the concentration of methidathion would have been much higher if the samples had been collected immediately following aerial application of the field. No fish tissue analysis was completed. The certainty index for this incident (B0000-218-10) is highly probable.

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 methidathion in CA. The risk characterization provides an estimation (Section 5.1) and a description (Section 5.2) of the likelihood of adverse effects; articulates risk assessment assumptions, limitations, and uncertainties; and synthesizes an overall conclusion regarding the likelihood of adverse effects to the CRLF or its designated critical habitat (i.e., “no effect,” “likely to adversely affect,” or “may affect, but not likely to adversely affect”).

5.1 Risk Estimation

Risk is estimated by calculating the ratio of exposure to toxicity. This ratio is the risk quotient (RQ), which is then compared to pre-established acute and chronic levels of

concern (LOCs) for each category evaluated (**Appendix E**). 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 methidathion usage scenarios summarized in **Table 3.2** and the appropriate aquatic toxicity endpoint from **Table 4.1**. Risks to the terrestrial-phase CRLF and its prey (*e.g.* terrestrial insects, small mammals and terrestrial-phase frogs) are estimated based on exposures resulting from applications of methidathion (**Tables 3.5 through 3.6**) and the appropriate toxicity endpoint from **Table 4.3**. Due to lack of toxicity data, exposures are not derived for terrestrial plants.

The minor degradate methidathion oxon is of toxicological concern, but because there are no toxicity data, this degradate cannot be included directly in calculations of RQs. Potential risk from methidathion oxon is characterized in the risk description (**Section 5.2**).

5.1.1 Exposures in the Aquatic Habitat

5.1.1.1 Direct Effects to Aquatic-Phase CRLF

Direct effects acute RQs for the aquatic-phase CRLF are presented in **Table 5.1**. Based on 1-in-10 year peak aquatic EECs from the PRZM/EXAMS model and the lowest acute 96-hour LC₅₀ for freshwater fish (surrogate for the aquatic-phase CRLF), acute RQs for the aquatic-phase CRLF exceed the listed species LOC (0.05) for all of the assessed methidathion uses. Acute RQs for methidathion range from 2.1 for the alfalfa scenario to 52.5 for the nursery scenario. As a note, for the spatial analyses sections, the lowest and highest EECs were estimated following a single application. The lowest acute RQ for a single application is calculated to be 0.20 for the lowest use scenario, on mangoes. Therefore, even after a single application, the acute listed species LOC would be exceeded for all of the methidathion use scenarios.

Table 5.1 Summary of Acute Direct Effect RQs for the Aquatic-phase CRLF^a					
Scenario	Crops Represented	Peak 1-in-10 Year EEC (µg/L)^b	Acute RQ^c	Probability of Individual Effect at LOC^d	Probability of Individual Effect at RQ^d
Alfalfa	Alfalfa, Clover, Timothy	4.6	2.1	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1.2
Almond	Almond, Walnut	49.3	22.4	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
Citrus	Calemondin, Citron, Citrus, Grapefruit, Kumquat, Lemon, Lime, Mango, Pummelo, Tangelo, Tangerine	18.3	8.3	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
Corn	Sunflower	20.7	9.4	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
Cotton	Cotton	13.0	5.9	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
Grapes	Kiwi	6.6	3.0	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1.1
Tree Fruit	Apple, Apricot, Cherry, Nectarine, Peach, Pear, Plum, Prune	15.6	7.1	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
Olive	Olive	15.2	6.9	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
Row Crop	Artichoke	16.0	7.3	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
Wheat	Safflower	13.6	6.2	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
Nursery	Nursery	115.6	52.5	~1 in 12,400 (~1 in 10,300,000 to ~1 in 149)	~1 in 1
^a Based on bluegill sunfish (<i>Lepomis macrochirus</i>) acute 96-hour LC ₅₀ = 2.2 µg/L ^b See Table 3.3 . ^c Acute RQs that exceed the acute endangered species LOC of 0.05 are in bold . ^d The effect probability was calculated based on a probit dose-response slope of 2.9 (1.9 – 4.0) for the bluegill sunfish acute toxicity study.					

Direct chronic risk estimates for the aquatic-phase CRLF are based on the 60-day average EECs from the PRZM/EXAMS model and the lowest chronic NOAEC for freshwater fish (surrogate for the aquatic-phase CRLF). Using the estimated chronic NOAEC of 0.07 ppb based on an ACR of 140 and the acute LC₅₀ value of 10 ppb, the chronic RQs exceed the LOC (1.0) for all of the assessed methidathion uses (**Table 5.2**). Based on

acute and chronic LOC exceedances, methidathion may affect the aquatic-phase of the CRLF.

Table 5.2 Summary of Chronic Direct Effect RQs for the Aquatic-phase CRLF^a			
Scenario	Crops Represented	60-day EEC (µg/L)^b	Chronic RQ^c
Alfalfa	Alfalfa, Clover, Timothy	2.5	35.7
Almond	Almond, Walnut	26.8	383
Citrus	Calemondin, Citron, Citrus, Grapefruit, Kumquat, Lemon, Lime, Mango, Pummelo, Tangelo, Tangerine	9.0	129
Corn	Sunflower	10.9	156
Cotton	Cotton	6.4	91.4
Grapes	Kiwi	3.2	45.7
Tree Fruit	Apple, Apricot, Cherry, Nectarine, Peach, Pear, Plum, Prune	5.0	71.4
Olive	Olive	7.6	109
Row Crop	Artichoke	8.7	124
Wheat	Safflower	6.6	94.3
Nursery	Nursery	54.3	776
^a Based on estimated chronic NOAEC of 0.07 µg/L from ACR ratio (see Section 4.1.1.2).			
^b See Table 3.3 .			
^c Chronic RQs that exceed the chronic LOC of 1.0 are in bold .			

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

Non-vascular Aquatic Plants

Indirect effects of methidathion to the aquatic-phase CRLF (tadpoles) via reduction in non-vascular aquatic plants in its diet cannot be quantitatively estimated because there are no aquatic plant toxicity data available for methidathion. For a qualitative risk description, see Section 5.2.2.1.

Aquatic Invertebrates

Indirect acute effects to the aquatic-phase CRLF via effects to prey (invertebrates) in aquatic habitats are based on peak 1-in-10 year EECs from the PRZM/EXAMS model and the lowest 48-hour LC₅₀ for freshwater invertebrates. These acute RQs for aquatic invertebrates range from 1.5 to 38.5, and thus exceed the acute risk LOC for non-listed species (0.5) (**Table 5.3**).

Table 5.3 Summary of Acute RQs Used to Estimate Indirect Effects to the CRLF via Direct Effects on Aquatic Invertebrates as Dietary Food Items

Scenario	Crops Represented	Peak 1-in-10 Year EEC (µg/L) ^b	Indirect Effects Acute RQ ^c	% Expected Effect on Prey Population at RQ ^d
Alfalfa	Alfalfa, Clover, Timothy	4.6	1.5	79
Almond	Almond, Walnut	49.3	16.4	100
Citrus	Calemondin, Citron, Citrus, Grapefruit, Kumquat, Lemon, Lime, Mango, Pummelo, Tangelo, Tangerine	18.3	6.1	100
Corn	Sunflower	20.7	6.9	100
Cotton	Cotton	13.0	4.3	100
Grapes	Kiwi	6.6	2.2	94
Tree Fruit	Apple, Apricot, Cherry, Nectarine, Peach, Pear, Plum, Prune	15.6	5.2	100
Olive	Olive	15.2	5.1	100
Row Crop	Artichoke	16.0	5.3	100
Wheat	Safflower	13.6	4.5	100
Nursery	Nursery	115.6	38.5	100

^a Based on water flea (*Daphnia magna*) acute 48-hour LC₅₀ = 3.0 µg/L

^b See **Table 3.3**.

^c Acute RQs that exceed the acute LOC for nonlisted species of 0.5 are in **bold**.

^d The acute daphnid toxicity study data do not fit the probit model; therefore, the effect probability was calculated based on a default slope assumption of 4.5 with upper and lower 95% confidence intervals of 2 and 9 (Urban and Cook, 1986).

Risk estimates for indirect effects to the CRLF via chronic effects to aquatic invertebrates are based on 21-day average EECs and the lowest chronic NOAEC for freshwater invertebrates. Chronic RQs for freshwater invertebrates range from 3.4 to 128.4, and thus exceed the LOC (1.0) for all of the assessed methidathion uses (**Table 5.4**). Based on acute and chronic LOC exceedences for all of the assessed uses, methidathion may indirectly affect the CRLF via reduction in freshwater invertebrates prey items.

Table 5.4 Summary of Chronic RQs Used to Estimate Indirect Effects to the CRLF via Direct Effects on Aquatic Invertebrates as Dietary Food Items			
Scenario	Crops Represented	21-day EEC (µg/L)^b	Indirect Effects Chronic RQ^c
Alfalfa	Alfalfa, Clover, Timothy	3.4	5.2
Almond	Almond, Walnut	40.2	60.9
Citrus	Calemondin, Citron, Citrus, Grapefruit, Kumquat, Lemon, Lime, Mango, Pummelo, Tangelo, Tangerine	13.6	20.6
Corn	Sunflower	16.2	24.5
Cotton	Cotton	9.3	14.1
Grapes	Kiwi	5.0	7.6
Tree Fruit	Apple, Apricot, Cherry, Nectarine, Peach, Pear, Plum, Prune	9.2	13.9
Olive	Olive	11.7	17.7
Row Crop	Artichoke	12.1	18.3
Wheat	Safflower	10.4	15.8
Nursery	Nursery	84.8	128.4

^a Based on water flea (*Daphnia magna*) chronic NOAEC = 0.66 µg/L
^b See **Table 3.3**.
^c Chronic RQs that exceed the chronic LOC of 1.0 are in **bold**.

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 (**Tables 5.1 - 5.2**) are used to assess potential indirect effects to the CRLF based on a reduction in freshwater fish and frogs as food items. Since the acute and chronic RQs exceed the LOC, methidathion may indirectly affect the CRLF via reduction in freshwater fish and frogs as food items.

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

Indirect effects to the CRLF via direct toxicity to aquatic plants cannot be quantitatively estimated because there are no aquatic plant toxicity data available for methidathion. For a qualitative risk description, see Section 5.2.3.1.

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 foliar applications of methidathion. Potential direct acute effects to the terrestrial-phase CRLF are derived by considering dose- and dietary-based EECs modeled in T-REX for a small bird (20 g) consuming small invertebrates (**Table 3.5**) and acute oral and subacute dietary toxicity endpoints for avian species. Potential direct chronic effects of methidathion to the terrestrial-phase CRLF are derived by considering

dietary-based exposures modeled in T-REX for a small bird (20g) consuming small invertebrates. Chronic effects are estimated using the lowest available toxicity data for birds. EECs are divided by toxicity values to estimate chronic dietary-based RQs. Acute dose-based, acute dietary-based, and chronic RQs exceed the LOCs for all of the assessed methidathion uses (**Table 5.5-5.7**). As a result, methidathion may affect the terrestrial-phase of the CRLF.

Table 5.5. Summary of Direct Effect Acute Dose-Based RQs for the Terrestrial-phase CRLF for Spray Applications of Methidathion				
Use	App. Rate (lb ai/A) Number of Apps. App. Interval (days)	Broadleaf Plants/ Small Insects		
		Acute Dose-Based RQ ^a	Probability of Individual Effect at LOC ^b	Probability of Individual Effect at RQ ^b
Artichoke	1 lb a.i./acre 8 apps 14 days	57.39	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Citrus	5 lb a.i./acre 2 apps 45 days	222.94	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Clover	1 lb a.i./acre 2 apps. 7 days	65.39	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Cotton	1 lb a.i./acre 4 apps 5 days	94.95	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Kiwi	2 lb a.i./acre 1 app NA	88.39	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Mango	0.25 lb a.i./acre 5 apps. 21 days	12.42	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Nursery	10 lb a.i./A 1 app NA	441.97	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Safflower, Sunflower	0.5 lb a.i./acre 3 apps 7 days	37.77	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Tree Fruit, Olives	3 lb a.i./acre 1 app NA	132.59	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
Walnut	2 lb a.i./acre 3 apps. 2 days	218.11	~1 in 294,000 (~1 in 8.86E+18 to ~1 in 44)	~1 in 1
^a Based on mallard duck acute oral LD ₅₀ = 6.7 mg/kg ^b The acute data do not fit the probit model; therefore, the effect probability was calculated based on a default slope assumption of 4.5 with upper and lower 95% confidence intervals of 2 and 9 (Urban and Cook, 1986). ^c Acute RQs that exceed the acute endangered species LOC of 0.1 are in bold .				

Table 5.6. Summary of Direct Effect Acute Dietary-Based RQs for the Terrestrial-phase CRLF for Spray Applications of Methidathion				
Use	App. Rate (lb ai/A) Number of Apps. App. Interval (days)	Broadleaf Plants/ Small Insects		
		Acute Dietary-Based RQ ^a	Probability of Individual Effect at LOC ^b	Probability of Individual Effect at RQ ^b
Artichoke	1 lb a.i./acre 8 apps 14 days	0.78	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 5 (~1 in 14 to ~1 in 2)
Citrus	5 lb a.i./acre 2 apps 45 days	3.04	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 1
Clover	1 lb a.i./acre 2 apps. 7 days	0.89	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 3 (~1 in 4 to ~1 in 2)
Cotton	1 lb a.i./acre 4 apps 5 days	1.29	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 1
Kiwi	2 lb a.i./acre 1 app NA	1.21	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 1
Mango	0.25 lb a.i./acre 5 apps. 21 days	0.17	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 9.28E+10 (~1 in 8.3E+25 to ~1 in 2.83E+2)
Nursery	10 lb a.i./A 1 app NA	6.03	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 1
Safflower, Sunflower	0.5 lb a.i./acre 3 apps 7 days	0.52	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 148 (~1 in 22,500 to ~1 in 6)
Tree Fruit, Olives	3 lb a.i./acre 1 app NA	1.81	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 1
Walnut	2 lb a.i./acre 3 apps. 2 days	2.97	~1 in 6.03E+17 (~1 in 7.85E+42 to ~1 in 4,300)	~1 in 1
^a Based on bobwhite quail acute dietary LC ₅₀ = 224 ppm				
^b The effect probability was calculated using a probit dose-response slope of 8.7 (3.5 – 13.8) from the bobwhite quail study.				
^c Acute RQs that exceed the acute endangered species LOC of 0.1 are in bold .				

Table 5.7. Summary of Direct Effect Chronic RQs for the Terrestrial-phase CRLF for Spray Applications of Methidathion		
Use	App. Rate (lb ai/A) Number of Apps. App. Interval (days)	Broadleaf Plants/ Small Insects
		Chronic RQ^a
Artichoke	1 lb a.i./acre 8 apps 14 days	17.53^b
Citrus	5 lb a.i./acre 2 apps 45 days	68.10
Clover	1 lb a.i./acre 2 apps. 7 days	19.97
Cotton	1 lb a.i./acre 4 apps 5 days	29.00
Kiwi	2 lb a.i./acre 1 app NA	27.00
Nursery	10 lb a.i./A 1 app NA	135.00
Mango	0.25 lb a.i./acre 5 apps. 21 days	3.79
Safflower, Sunflower	0.5 lb a.i./acre 3 apps 7 days	11.54
Tree Fruit, Olives	3 lb a.i./acre 1 app NA	40.50
Walnut	2 lb a.i./acre 3 apps. 2 days	66.62
^a Based on mallard duck chronic NOAEC = 10 ppm		
^b Chronic RQs that exceed the chronic LOC of 1.0 are in bold .		

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 risks of methidathion 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 LD₅₀ of 0.236 µg/bee by 1 bee/0.128g, which is based on the weight of an adult honey bee. EECs (ppm) calculated by T-REX for small and large insects are divided by the calculated toxicity value for terrestrial invertebrates, which is 1.84 ppm (**Table 5.8**). Based on acute LOC exceedances for all of

the assessed uses, methidathion may affect the CRLF via reduction in terrestrial invertebrate prey items.

Table 5.8 Summary of RQs For Indirect Effects to the Terrestrial-phase CRLF via Direct Effects on Terrestrial Invertebrates as Dietary Food Items					
Use	App. Rate (lb ai/A) Number of Apps. App. Interval (days)	Small Insect RQ	Large Insect RQ	Small Insect % Expected Effect on Prey Population at RQ^b	Large Insect % Expected Effect on Prey Population at RQ^b
Artichoke	1 lb a.i./acre 8 apps 14 days	95	10	100	100
Citrus	5 lb a.i./acre 2 apps 45 days	370	41	100	100
Clover	1 lb a.i./acre 2 apps. 7 days	109	12	100	100
Cotton	1 lb a.i./acre 4 apps 5 days	158	17	100	100
Kiwi	2 lb a.i./acre 1 app NA	147	16	100	100
Mangos	0.25 lb a.i./acre 5 apps 21 days	21	2	100	100
Nursery	10 lb a.i./A 1 app NA	734	82	100	100
Safflower, Sunflower	0.5 lb a.i./acre 3 apps 7 days	63	7	100	100
Tree Fruit, Olives	3 lb a.i./acre 1 app NA	220	24	100	100
Walnut	2 lb a.i./acre 3 apps. 2 days	362	40	100	100
^a Based on honey bee LD ₅₀ of 0.236 µg/bee or 1.84 ppm ^b The % population was calculated using a probit dose-response slope of 9.06 from the honey bee study. ^c Acute RQs that exceed the acute non-listed species LOC of 0.5 are in bold .					

5.1.2.2.2 Mammals

Risks associated with ingestion of contaminated small mammals by large terrestrial-phase CRLFs are derived for dietary-based and dose-based exposures modeled in T-REX for a small mammal (15g) consuming short grass. Acute and chronic effects are estimated using the most sensitive mammalian toxicity data. EECs are divided by the toxicity value to estimate acute dose-based RQs as well as chronic dietary- and dose-based RQs. As shown in **Tables 5.9-5.10**, the acute and chronic RQs exceed the LOC for all of the

assessed methidathion uses. As a result, methidathion may affect the CRLF via reduction in small mammal prey items.

Table 5.9. Summary of Acute RQs For Indirect Effects to the Terrestrial-phase CRLF via Direct Effects on Small Mammals as Dietary Food Items			
Use	App. Rate (lb ai/A) Number of Apps. App. Interval (days)	Acute Dose-Based RQ^a	% Expected Effect on Prey Population at RQ^b
Artichoke	1 lb a.i./acre 8 apps 14 days	11.27	100
Citrus	5 lb a.i./acre 2 apps 45 days	43.76	100
Clover	1 lb a.i./acre 2 apps. 7 days	12.84	100
Cotton	1 lb a.i./acre 4 apps 5 days	18.64	100
Kiwi	2 lb a.i./acre 1 app NA	17.35	100
Mango	0.25 lb a.i./A 5 apps 21 days	2.44	96
Nursery	10 lb a.i./A 1 app NA	86.76	100
Safflower, Sunflower	0.5 lb a.i./acre 3 apps 7 days	7.41	100
Tree Fruit, Olives	3 lb a.i./acre 1 app NA	26.03	100
Walnut	2 lb a.i./acre 3 apps. 2 days	42.82	100
^a Based on rat acute oral LD ₅₀ = 12 mg/kg-bw. ^b The effect probability was calculated based on a default slope assumption of 4.5 with upper and lower 95% confidence intervals of 2 and 9 (Urban and Cook, 1986). ^c Acute RQs that exceed the acute non-listed species LOC of 0.5 are in bold .			

Table 5.10. Summary of Chronic RQs For Indirect Effects to the Terrestrial-phase CRLF via Direct Effects on Small Mammals as Dietary Food Items			
Use	App. Rate (lb ai/A) Number of Apps. App. Interval (days)	Chronic Dose-Based RQ^a	Chronic Dietary-Based RQ^b
Artichoke	1 lb a.i./acre 8 apps 14 days	540.74	62.33
Citrus	5 lb a.i./acre 2 apps 45 days	2100.70	242.13
Clover	1 lb a.i./acre 2 apps. 7 days	616.11	71.01
Cotton	1 lb a.i./acre 4 apps 5 days	894.65	103.12
Kiwi	2 lb a.i./acre 1 app NA	832.90	96.00
Mango	0.25 lb a.i./A 5 apps 21 days	117.00	13.49
Nursery	10 lb a.i./A 1 app NA	4164.5	480.00
Safflower, Sunflower	0.5 lb a.i./acre 3 apps 7 days	355.92	41.02
Tree Fruit, Olives	3 lb a.i./acre 1 app NA	1249.35	144.00
Walnut	2 lb a.i./acre 3 apps. 2 days	2055.21	236.88
^a Based on rat NOAEL = 0.25 mg/kg-bw. ^b Based on rat NOAEC = 5 mg/kg-diet. ^c Chronic RQs that exceed the chronic LOC of 1 are in bold .			

5.1.2.2.3 Frogs

An additional prey item of the adult terrestrial-phase CRLF is other species of frogs. In order to assess risks to these organisms, dietary-based and dose-based exposures modeled in T-REX for a small bird (20g) consuming small invertebrates are used. See Section 5.1.2.1 and associated tables (**Tables 5.3 – 5.5**) for results. Acute and chronic RQs exceed the LOCs for all of the assessed methidathion uses. As a result, methidathion may affect the CRLF via reduction in frogs as prey items.

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

Indirect effects to the CRLF via reduction in terrestrial plant community cannot be quantitatively estimated because there are no vegetative vigor or seedling emergence plant toxicity data available for methidathion. For a qualitative risk description, see Section 5.2.3.2.

5.1.3 Primary Constituent Elements of Designated Critical Habitat

For methidathion 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.

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

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

- Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.
- Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.
- Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.*, algae).

Risk estimations for potential effects to aquatic and/or terrestrial plants were not conducted because no toxicity data are available for either aquatic or terrestrial plants. Therefore, it cannot be estimated whether or not methidathion is likely to affect aquatic-phase PCEs of designated habitat related to effects on aquatic and/or terrestrial plants. Risks to aquatic and terrestrial plants will be discussed qualitatively in sections 5.2.2.1, 5.2.3.2 and 5.2.3.2.

The remaining aquatic-phase PCE is “alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.” To assess the impact of methidathion on this PCE, acute and chronic freshwater fish and

invertebrate toxicity endpoints, as well endpoints for aquatic non-vascular plants, are used as measures of effects. RQs for these endpoints were calculated in Sections 5.1.1.1 and 5.1.1.2. Based on exceedances of the listed and non-listed species LOCs following both acute and chronic exposure for freshwater fish and invertebrates, methidathion is likely to affect aquatic-phase PCEs of designated habitat related to effects of alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source. As stated previously, there are no aquatic plant toxicity data available for methidathion. Therefore, effects to this aquatic-phase PCE based on effects to aquatic non-vascular plants will be discussed qualitatively in Section 5.2.2.1.

5.1.3.2 Terrestrial-Phase (Upland Habitat and Dispersal Habitat)

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

- Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance
- Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal

The risk estimation for terrestrial-phase PCEs of designated habitat related to potential effects on terrestrial plants cannot be quantitatively addressed because there are no vegetative vigor or seedling emergence plant toxicity data available for methidathion. The risk will be discussed qualitatively in Section 5.2.3.2.

The third terrestrial-phase PCE is “reduction and/or modification of food sources for terrestrial phase juveniles and adults.” To assess the impact of methidathion on this PCE, acute and chronic toxicity endpoints for birds, mammals, and terrestrial invertebrates are used as measures of effects. RQs for these endpoints were calculated in Sections 5.1.2.1 and 5.1.2.2. Acute and chronic LOCs for listed and non-listed species were exceeded for all uses for birds and mammals and acute LOCs for listed and non-listed terrestrial invertebrates were also exceeded for all uses. Therefore, methidathion is likely to affect the third terrestrial-phase PCE.

The fourth terrestrial-phase PC is based on alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source. Direct acute and chronic RQs for terrestrial-phase CRLFs are presented in Section 5.1.2.1. Acute and/or chronic LOCs for listed and non-listed species were exceeded for freshwater fish and invertebrates, birds, mammals and terrestrial invertebrates. No data are available for either aquatic or terrestrial plants. However, based on the results with aquatic and terrestrial animals, methidathion is likely to affect the fourth terrestrial - phase PCE.

5.1.4 Spatial Extent of Potential Effects

Since this screening level risk assessment defines taxa that are predicted to be exposed through runoff and drift to methidathion 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 spray drift assessment for determining the extent of terrestrial and aquatic habitats potentially affected through drift alone; and (2) the down stream dilution assessment for determining the extent of the affected lotic aquatic habitats (flowing water). This analysis is described briefly below and in more detail in **Appendix D**.

An LAA effects determination applies to those areas where it is expected that the pesticide's use will directly or indirectly affect the CRLF or its designated critical habitat. To determine this area, the footprint of methidathion's use pattern is identified, using corresponding land cover data. The spatial extent of the effects determination also includes areas beyond the initial area of concern that may be impacted by runoff and/or spray drift. The identified direct and indirect effects and modification to critical habitat are anticipated to occur only for those currently occupied core habitat areas, CNDDDB occurrence sections and designated critical habitat for the CRLF that overlap with the initial area of concern plus greater than 1000 feet from its boundary. It is assumed that non-flowing waterbodies (or potential CRLF habitat) are included within this area.

In addition to the spray drift buffer, the results of the downstream dilution extent analysis result in a distance of 285 kilometers which represents the maximum continuous distance of downstream dilution from the edge of the initial area of concern. If any of these streams reaches flow into CRLF habitat, there is potential to affect either the CRLF or modify its habitat. These lotic aquatic habitats within the CRLF core areas and critical habitats potentially contain concentrations of methidathion sufficient to result in LAA determination or modification of critical habitat.

The determination of the buffer distance and downstream dilution for spatial extent of the effects determination is described below.

5.1.4.1 Spray Drift

In order to determine terrestrial and aquatic habitats of concern due to methidathion exposures through spray drift, it is necessary to estimate the distance that spray applications can drift from the treated area and still be present at concentrations that exceed levels of concern. A quantitative analysis of spray drift distances was completed using AgDrift (v. 2.01).

Spatial analysis of spray drift effects is limited to consideration of a single application because, due to variable wind conditions, multiple applications are not likely to impact the same location each time. Spray drift distances depend on both application rate and

method, and so, in order to understand the range of possible impacts, modeling was done for the uses with the highest single application rates for each method, which include tree fruit and nuts treated by aerial spray, nursery stock treated by ground spray, and citrus treated by spray that is presumed to be airblast. Use on mangos was modeled as well to represent the lower boundary of potential effects.

Methidathion labels have specific application requirements in order to reduce potential spray drift, including restrictions on wind speed, release height and droplet size. AgDrift inputs are based on these requirements, as presented in **Table 5.11**. For inputs not specified on the labels, default values are used. When Tier I models were used, it is because no higher tier models are available.

Table 5.11. Input parameters for simulation of methidathion in spray drift using AgDrift (v. 2.01)

Parameter Description	Tree Fruit / Nuts	Nursery Stock	Citrus	Mangos
Single Application Rate	3 lb a.i./A	10 lb a.i./A	5 lb a.i./A	0.5 lb a.i./A
Application method	Aerial	Ground	Airblast	Airblast
Droplet Size Distribution (DSD)	Medium to Coarse	Fine to Medium ¹	NA	NA
Release height	NA	High Boom ²	NA	NA
Orchard type	NA	NA	Dense Orchard	Dense Orchard

¹ Labels require a medium to coarse DSD, but AgDrift does not include this as an option for Tier I Ground mode.

² Labels require a release height for ground spray applications of less than 4 feet above the crop canopy. AgDrift does not use specific release heights, so modeling for this use is based high boom spray, the option with the highest release height, 4 ft (1.27 m).

The terrestrial analysis is based on the honey bee acute contact LD₅₀ of 0.236 µg/bee, or 1.84 ppm, which is the most sensitive terrestrial endpoint and is used as a surrogate for terrestrial invertebrates. Based on this endpoint, the initial average deposition level that would lead to exceedance of the terrestrial invertebrate LOC (0.05) is 0.00068 lb a.i./A, where the deposition is calculated as the RQ divided by the LOC and multiplied by the application rate. The most sensitive endpoint upon which the aquatic analysis is based is the bluegill sunfish 96-hour acute LC₅₀ of 2.2 µg a.i./L. With this toxicity endpoint, the endangered species LOC of 0.05 will be exceeded at any concentration greater than 0.11 µg a.i./L.

Table 5.12 includes uses with the maximum single application rates for each application method and presents a summary of the buffer distances at which spray drift deposition from these uses drop below levels of concern (e.g., RQs will be below LOCs). These distances represent the maximum extent where effects are possible using the most sensitive data and the listed species LOC of 0.05 for either terrestrial invertebrates or freshwater fish. For each application method, lower application rates will yield a smaller buffer distance than reported here.

Table 5.12 Summary of maximum predicted distances for potential spray drift effects.

Application Method	Application Rate (lb a.i./A)	Uses Represented	Terrestrial LD ₅₀ Distance (ft)	Aquatic LC ₅₀ Distance (ft)
Aerial	3	Almond & Tree Fruit	> 1000	>1000
Ground	10	Nursery	> 1000	> 1000
Airblast	5	Citrus	> 1000	> 1000
Airblast	0.25	Mango	197	23

The use of methidathion on almonds and tree fruit is predicted to have effects for terrestrial invertebrates at distances greater than 1,000 ft (0.3 km), the limit of AgDrift modeling. Given the uncertainty in AgDrift and the assumptions used in the modeling, the potential for effects from drift cannot be quantified beyond 1000 ft and so this value is used to represent the potential for effects to terrestrial invertebrates and to define the spatial extent of the effects determination (i.e., this buffer distance is added to the initial area of concern). Given the toxicity of methidathion and the calculated RQs, the extent of spray drift effects may be at least an order of magnitude higher than this.

5.1.4.2 Downstream Dilution Analysis

The maximum downstream extent of methidathion exposure in streams and rivers where the EEC can potentially be above levels that would exceed the most sensitive LOC may be estimated by utilizing the greatest ratio of aquatic RQ to LOC. This is based on the assumption that there is uniform runoff across the landscape in treated areas with streams flowing through them (i.e. the initial area of concern), represented by the modeled EECs, and that as those waters move downstream, the influx of non-impacted water will dilute the concentrations of methidathion present.

Using an LC₅₀ value of 2.2 ug/L for freshwater fish (the most sensitive species) and a maximum peak EEC of 114.7 µg/L for applications to nursery stock yields an RQ/LOC ratio of 1042 (52.2/0.05). Using the downstream dilution approach (described in more detail in **Appendix D**) yields a target percent crop area (PCA) of 27.8%. This value has been input into the downstream dilution approach and results in a distance of 285 kilometers, which represents the maximum continuous distance of downstream dilution from the edge of the initial area of concern. Similar to the spray drift buffer described above, the LAA/NLAA determination is based on the area defined by the point where concentrations exceed the LC₅₀ value.

5.1.4.3 Overlap between CRLF habitat and Spatial Extent of Potential Effects

An LAA effects determination is made for those areas where it is expected that the pesticide's use will directly or indirectly affect the CRLF or its designated critical habitat and the area overlaps with the core areas, critical habitat and available occurrence data for CRLF.

Figure 5.1 shows that there is some overlap between the initial area of concern mapped according to methidathion's use pattern and the CRLF habitat, including currently occupied core areas, CNDDDB occurrence sections, and designated critical habitat. This map does not include areas beyond the initial area of concern that may be impacted by runoff and/or spray drift. It is expected that any additional areas of CRLF habitat that are located >1000 ft (to account for offsite migration via spray drift) and 285 kilometers of stream reach (to account for downstream dilution) outside the initial area of concern may also be impacted and are part of the full spatial extent of the LAA/modification of critical habitat effects determination.

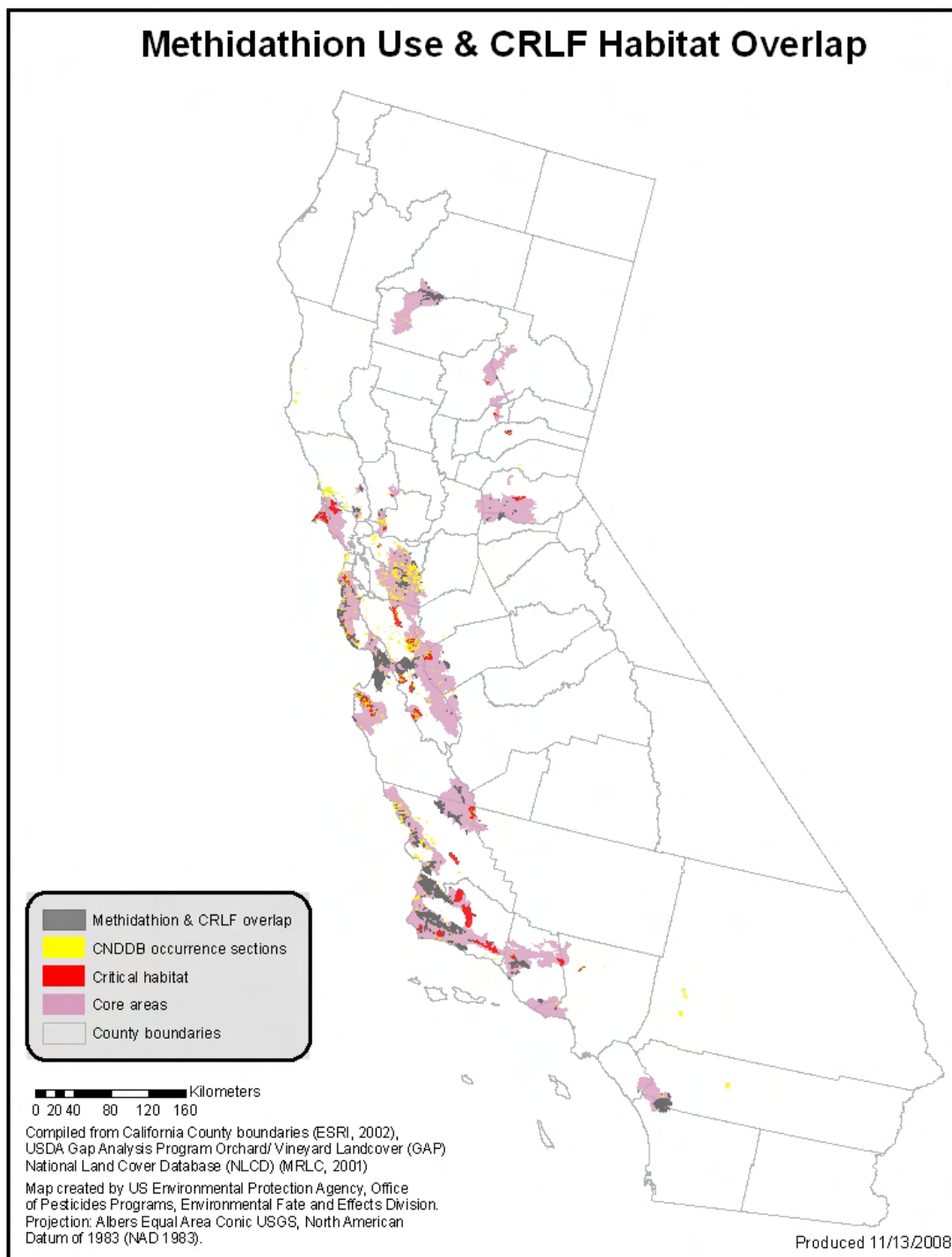


Figure 5.1. Overlap Map: CRLF Habitat and Methidathion Initial Area of Concern

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 methidathion’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 methidathion. A summary of the results of the risk estimation results are provided in **Table 5.13** for direct and indirect effects to the CRLF and in **Table 5.14** for the PCEs of designated critical habitat for the CRLF.

These results represent risk from exposure to applied methidathion and do not include the additional risk posed by any degradates of toxicological concern. Given that acute and chronic risk quotients are exceeded for the parent compound alone, any contribution in toxicity from any of the major degradates or from methidathion oxon or other OP degradates would increase the risk estimates. Potential risk from methidathion oxon is characterized in **Section 5.2.1.3**.

Table 5.13 Risk Estimation Summary for Methidathion - Direct and Indirect Effects to CRLF		
Assessment Endpoint	LOC Exceedances (Y/N)	Description of Results of Risk Estimation
<i>Aquatic Phase (eggs, larvae, tadpoles, juveniles, and adults)</i>		
Direct Effects Survival, growth, and reproduction of CRLF individuals via direct effects on aquatic phases	Y	There are LOC exceedances for listed species following both acute and chronic exposure using freshwater fish as the surrogate for aquatic-phase amphibians.
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects to food supply (<i>i.e.</i> , fish, freshwater invertebrates, non-vascular plants)	Y	The acute LOC for non-listed species (all uses) and the chronic LOC (all uses except alfalfa, clover, timothy and kiwi) are exceeded for freshwater fish. There are LOC exceedances for non-listed species following both acute and chronic exposure to freshwater invertebrates. No aquatic plant data are available. A qualitative discussion of risk will be provided.
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects on habitat, cover, and/or primary productivity (<i>i.e.</i> , aquatic plant community)	Unknown	No aquatic plant data are available. A qualitative discussion of risk to aquatic plants will be provided.

Table 5.13 Risk Estimation Summary for Methidathion - Direct and Indirect Effects to CRLF		
Assessment Endpoint	LOC Exceedances (Y/N)	Description of Results of Risk Estimation
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.	Unknown	No terrestrial plant data are available. A qualitative discussion of risk will be provided.
<i>Terrestrial Phase (Juveniles and adults)</i>		
Direct Effects Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	Y	There are LOC exceedances for listed species following both acute and chronic exposure using birds as the surrogate for terrestrial-phase amphibians.
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects on prey (<i>i.e.</i> , terrestrial invertebrates, small terrestrial mammals and terrestrial phase amphibians)	Y	There are LOC exceedances for non-listed species following both acute and/or chronic exposure to terrestrial invertebrates, mammals and birds (surrogate to terrestrial-phase amphibians).
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects on habitat (<i>i.e.</i> , riparian vegetation)	Unknown	No terrestrial plant data are available. A qualitative discussion of risk will be provided.

Table 5.14 Risk Estimation Summary for Methidathion – PCEs of Designated Critical Habitat for the CRLF		
Assessment Endpoint	Habitat Modification (Y/N)	Description of Results of Risk Estimation
<i>Aquatic Phase PCEs (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)</i>		
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.	Unknown	No aquatic and terrestrial plant data are available. A qualitative discussion of risk will be provided.
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.	Unknown	No aquatic and terrestrial plant data are available. A qualitative discussion of risk will be provided.

Table 5.14 Risk Estimation Summary for Methidathion – PCEs of Designated Critical Habitat for the CRLF		
Assessment Endpoint	Habitat Modification (Y/N)	Description of Results of Risk Estimation
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	Y	LOC exceedances for both listed and non-listed species following both acute and chronic exposure for freshwater fish and invertebrates. No aquatic plant toxicity data are available.
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (e.g., algae)	Unknown	No aquatic plant data are available. A qualitative discussion of risk will be provided.
<i>Terrestrial Phase PCEs (Upland Habitat and Dispersal Habitat)</i>		
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	Unknown	No terrestrial plant data are available. A qualitative discussion of risk will be provided.
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	Unknown	No terrestrial plant data are available. A qualitative discussion of risk will be provided.
Reduction and/or modification of food sources for terrestrial phase juveniles and adults	Y	Acute and chronic LOCs for listed and non-listed species were exceeded for all uses for birds and mammals and acute LOCs for listed and non-listed terrestrial invertebrates were also exceeded for all uses.
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	Y	Acute and/or chronic LOCs for listed and non-listed species were exceeded for freshwater fish and invertebrates, birds, mammals and terrestrial invertebrates. No data are available for either aquatic or terrestrial plants.

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

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

- Significance of Effect: Insignificant effects are those that cannot be meaningfully measured, detected, or evaluated in the context of a level of effect where “take” occurs for even a single individual. “Take” in this context means to harass or harm, defined as the following:
 - Harm includes significant habitat modification or degradation that results in death or injury to listed species by significantly impairing behavioral patterns such as breeding, feeding, or sheltering.
 - Harass is defined as actions that create the likelihood of injury to listed species to such an extent as to significantly disrupt normal behavior patterns which include, but are not limited to, breeding, feeding, or sheltering.
- Likelihood of the Effect Occurring: Discountable effects are those that are extremely unlikely to occur.
- Adverse Nature of Effect: Effects that are wholly beneficial without any adverse effects are not considered adverse.

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

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 methidathion.

As stated in the risk estimation section, acute RQs for the aquatic-phase CRLF exceed the listed species LOC (0.05) for all of the assessed methidathion uses. The acute RQs range from 2.1 for the alfalfa scenario to 52.5 for the nursery scenario. In addition, the lowest acute RQ following a single application is estimated to be 0.20 for the lowest mango scenario, which still exceeds the acute LOC for listed species.

Limited acute toxicity data are available to assess sensitivity across freshwater fish species. The lowest and highest acute 96-hour LC₅₀s values from the submitted studies on bluegill sunfish, rainbow trout and goldfish are 2.2 (bluegill sunfish) and 14 µg/L (rainbow trout). Estimated acute RQs from these studies indicate that they would exceed the listed species acute LOC for all of the assessed methidathion uses for any one of these three surrogate freshwater fish species. It is noted that one study on the common eel (*Anguilla anguilla*) was available in the open literature with a 96-hr acute LC₅₀ that is close to three orders of magnitude higher (1510 µg/L) than the most sensitive submitted freshwater fish species. If acute RQs were estimated based on that study, they would range from 0.003 for the alfalfa scenario to 0.08 for the nursery scenario, the only

scenario that exceeds the acute LOC for listed species. Therefore, there will be some fish species that may not be at risk following acute exposure to some of the methidathion use scenarios, and the relative sensitivity of aquatic amphibians to fish is unknown.

Based on a probit dose-response slope of 2.9 (1.9 – 4.0) from the bluegill sunfish acute toxicity study with an acute LC_{50} of 2.2 $\mu\text{g/L}$, the probability of an individual effect at the LOC is estimated to be 1 in 12,400 and the probability of an individual effect at the RQ is close to 1 in 1 for all scenarios. Sublethal effects were reported in the bluegill sunfish acute toxicity study at concentration levels higher than the endpoints utilized in this risk assessment (MRID 00011841). In this study, it was reported that test organisms in the highest exposure group (1000 $\mu\text{g/L}$) exhibited slight spastic motions and swam on their sides.

As stated previously, the chronic NOAEC of 6.3 $\mu\text{g/L}$ for the fathead minnow is similar to the acute 96-hour LC_{50} of several other test species, ranging from 2.2 to 14 $\mu\text{g/L}$. A chronic NOAEC for any of these other species would likely be lower than the available fathead minnow NOAEC. Therefore, this assessment estimated a chronic NOAEC for rainbow trout using an acute to chronic ratio (ACR) calculation from data from other organophosphates. The most conservative acute to chronic ratio from the other organophosphate data is 140. Using that ACR value with an acute LC_{50} value of 10 ppb for rainbow trout, the chronic NOAEC used in this assessment is estimated to be 0.07 ppb. With this value, the chronic RQs exceed the LOC (1.0) for all of the assessed methidathion uses (**Table 5.2**). If the chronic NOAEC of 6.3 ppb from the fathead minnow study is used, then the chronic RQs would exceed the LOC (1.0) for all of the assessed methidathion uses except for the alfalfa scenario (RQ = 0.40: alfalfa, clover and timothy); the grape scenario (RQ = 0.51: kiwi) and the tree fruit scenario (RQ = 0.79: apple, apricot, cherry, nectarine, peach, pear, plum and prune). Therefore, even with the endpoint from the chronic study in fathead minnows, the chronic LOC is still exceeded with many methidathion uses.

Sublethal effects in the chronic study include effects on total length and wet weight at the highest measured treatment level (12 $\mu\text{g ai/L}$) relative to the pooled control.

Surface water monitoring data support the conclusion of risk determined based on calculated RQs. Monitoring in the Sacramento Valley found 46 detections out of 472 samples with a maximum of 15.1 $\mu\text{g/L}$ and an average across the detections of 1.2 $\mu\text{g/L}$. In the San Joaquin Valley, the maximum of 30 detections out of 738 samples was 2.4 $\mu\text{g/L}$ and the average was 0.3 $\mu\text{g/L}$. Atmospheric monitoring detected methidathion concentrations in fog of 15.5 $\mu\text{g/L}$. Compared to 96-hr acute LC_{50} values of 2.2 $\mu\text{g/L}$ to 14 $\mu\text{g/L}$, these data demonstrate the potential for aquatic exposures that could lead to direct effects, particularly because monitoring is generally not expected to capture peak values.

In 1976, approximately 3,000 fish of unknown species in a nearby creek were killed following an aerial application of methidathion to an agricultural area in Sacramento County, California. The incident was attributed to accidental misuse of methidathion.

Samples of the water collected from the field drain six days after the material was applied to the field showed methidathion concentrations of 60 ppb. The certainty index for this incident is highly probable.

In summary, methidathion is very highly toxic to freshwater fish, the surrogate for the aquatic-phase CRLF. The acute LOC (listed species) and the chronic LOC are exceeded for all of the methidathion uses. There is one study in the open literature that indicates that there will be some fish species that may not be at risk following acute exposure from some of the methidathion use scenarios. This is an uncertainty. The probability of an individual effect on an acute basis approaches 100% at the RQ levels. Incident data indicate that freshwater fish are vulnerable, particularly if there are misuses. Spatial analyses indicate that a significant overlap will exist between the use sites for methidathion and the CRLF habitat, particularly when the spraydrift and downstream dilution buffers are applied to the “footprint” of the methidathion use area. Therefore, based on the weight-of-evidence, there is a potential for direct impact to the aquatic-phase CRLF based on the endpoints generated from the freshwater fish data and the effects determination is Likely to adversely affect (LAA).

As stated previously, the only detected degradate of toxicological concern is the minor degradate methidathion oxon, which may be more toxic than the parent. However, no data are available on the environmental fate, toxicity, and occurrence of methidathion oxon in the environment and so it is not considered directly in calculated RQs. Given that acute and chronic risk quotients are exceeded for the parent compound alone, any contribution in toxicity from any of the major degradates or from methidathion oxon or other OP degradates would increase the risk estimates. Further characterization of risk from methidathion oxon is provided in **Section 5.2.1.3**.

5.2.1.2 Terrestrial-Phase CRLF

As stated in the risk estimation section, acute dose-based and acute dietary-based RQs for the terrestrial-phase CRLF exceed the listed species LOC (0.1) for all of the assessed methidathion uses. The acute RQs range from 0.17 for mangos on a dietary basis to 442.0 for nursery uses on a dose basis. Limited acute toxicity data are available to assess sensitivity across avian species on a dose-basis. The acute oral LD₅₀'s range from 6.7 mg/kg for mallard ducks (*Anas platyrhynchos*) (MRID 000159201) to 225 mg/kg for Chukar (*Alectoris chukar*) (MRID 00060823). Other species fall within that range. Even with the highest LD₅₀ for the Chukar, the dose-based acute RQ for avian species is still over 150 times higher than the acute LOC of 0.1 for listed terrestrial animals with the nursery uses. With the lowest application rate of 0.25 lb a.i./A for mangos and the highest acute LD₅₀ value for the Chukar, the acute dose-based RQ for small birds eating small insects (0.29) is still greater than the acute LOC of 0.1 for endangered birds. On a dietary basis, there are only two studies with the technical material, one with bobwhite quail and one with mallard ducks. The acute dietary LC₅₀s are 224 and 543 ppm for bobwhite quail and mallard ducks, respectively. Again, with the lowest application rate of 0.25 lb a.i./A for mangos and using the bobwhite quail LC₅₀ value of 224 ppm, the acute RQ on a dietary basis for small birds eating small insects is 0.17, which exceeds the

LOC for listed species. Using the mallard duck LC₅₀ value of 543 ppm, the acute RQ on a dietary basis for small birds eating small insects is 0.07, which is just under the LOC for endangered species. However, with the next highest application rate of 0.5 lbs a.i./A for safflower, even when applied only once per year, the RQ using the highest LC₅₀ of 543 ppm for mallard ducks, the RQ would be 0.12, which is greater than the acute LOC for endangered birds. Therefore, even within the range of dose- and dietary-based acute toxicity values for avian species, there is potential acute risk to endangered birds for all uses with the possible exception of mangos with some of the less sensitive birds.

Chronic RQs also exceed the chronic LOC for all of the assessed methidathion uses (**Table 5.7**). The chronic endpoint used in this assessment (10 ppm) is based on a study with mallard ducks (MRID 44381602). In bobwhite quail, no effects were observed in two studies at the highest concentrations tested (30 and 35 ppm; MRIDs 44381601 and 44381602, respectively). Using the highest bobwhite quail NOAEC value of 35 ppm, the chronic RQ with the lowest application rate (mangos) is 1.08, which is still greater than the chronic LOC of 1. Therefore, within the available range of dietary-based chronic toxicity values for avian species (two), there is potential risk following chronic exposure for all of the assessed methidathion uses.

In an effort to refine the acute dose-based risk estimates, the T-REX model was modified to account for the lower metabolic rate and lower caloric requirement of amphibians (compared to birds). Acute dose-based RQs were recalculated using the T-HERPS (Version 1.0) model for small (1 g), medium (37 g), and large (238 g) frogs (**Table 5.15, Appendix M**). Using this refinement, the acute dose-based RQs still exceed the LOC (0.1) for nearly all of the modeled scenarios for methidathion. The T-HERPS model can only be used to refine dose-based risk estimates at this time; thus, further refinement of the acute or chronic dietary-based RQs is not possible.

Table 5.15. Upper Bound Kenaga, Acute Terrestrial Herpetofauna Dose-Based Risk Quotients											
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 ¹	EEC	RQ ¹	EEC	RQ ¹	EEC	RQ ¹	EEC	RQ ¹
ARTICHOKE											
1.4	6.70	6.81	1.02	0.76	0.11	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	6.69	1.00	0.74	0.11	194.24	28.99	12.14	1.81	0.23	0.03
238	6.70	4.39	0.65	0.49	0.07	30.20	4.51	1.89	0.28	0.15	0.02
CITRUS											
1.4	6.70	26.46	3.95	2.94	0.44	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	26.00	3.88	2.89	0.43	754.62	112.63	47.16	7.04	0.90	0.13
238	6.70	17.04	2.54	1.89	0.28	117.31	17.51	7.33	1.09	0.59	0.09
CLOVER											
1.4	6.70	7.76	1.16	0.86	0.13	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	7.63	1.14	0.85	0.13	221.32	33.03	13.83	2.06	0.26	0.04

Table 5.15. Upper Bound Kenaga, Acute Terrestrial Herpetofauna Dose-Based Risk Quotients											
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 ¹	EEC	RQ ¹	EEC	RQ ¹	EEC	RQ ¹	EEC	RQ ¹
238	6.70	5.00	0.75	0.56	0.08	34.41	5.14	2.15	0.32	0.17	0.03
COTTON											
1.4	6.70	11.27	1.68	1.25	0.19	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	11.07	1.65	1.23	0.18	321.38	47.97	20.09	3.00	0.38	0.06
238	6.70	7.26	1.08	0.81	0.12	49.96	7.46	3.12	0.47	0.25	0.04
KIWI											
1.4	6.70	10.49	1.57	1.17	0.17	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	10.31	1.54	1.15	0.17	299.20	44.66	18.70	2.79	0.36	0.05
238	6.70	6.76	1.01	0.75	0.11	46.51	6.94	2.91	0.43	0.23	0.04
MANGO											
1.4	6.70	1.47	0.22	0.16	0.02	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	1.45	0.22	0.16	0.02	42.03	6.27	2.63	0.39	0.05	0.01
238	6.70	0.95	0.14	0.11	0.02	6.53	0.98	0.41	0.06	0.03	0.00
NURSERY											
1.4	6.70	2.62	0.39	0.29	0.04	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	2.58	0.38	0.29	0.04	74.80	11.16	4.67	0.70	0.09	0.01
238	6.70	1.69	0.25	0.19	0.03	11.63	1.74	0.73	0.11	0.06	0.01
SAFFLOWER, SUNFLOWER											
1.4	6.70	4.48	0.67	0.50	0.07	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	4.41	0.66	0.49	0.07	127.85	19.08	7.99	1.19	0.15	0.02
238	6.70	2.89	0.43	0.32	0.05	19.88	2.97	1.24	0.19	0.10	0.01
TREE FRUIT, OLIVES											
1.4	6.70	15.73	2.35	1.75	0.26	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	15.46	2.31	1.72	0.26	448.79	66.98	28.05	4.19	0.54	0.08
238	6.70	10.14	1.51	1.13	0.17	69.77	10.41	4.36	0.65	0.35	0.05
WALNUT											
1.4	6.70	25.88	3.86	2.88	0.43	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	25.44	3.80	2.83	0.42	738.28	110.19	46.14	6.89	0.88	0.13
238	6.70	16.67	2.49	1.85	0.28	114.77	17.13	7.17	1.07	0.58	0.09
^a Acute RQs that exceed the acute endangered species LOC of 0.1 are in bold .											

On a dose basis, based on a default probit dose-response slope of 4.5 (2 – 9) from the mallard duck acute oral toxicity study with an acute LD₅₀ of 6.7 mg/kg bw, the probability of an individual effect at the RQ is approximately 1 in 1 for all scenarios. On a dietary basis, the probit dose-response slope was 8.7 (3.5-13.8) from the bobwhite quail dietary study with an LC₅₀ of 224 ppm. The probability of an individual effect at the RQ ranged from 1 in 1 for 5 use scenarios to 1 in 9.28E+10 for use on mangos (see **Tables 5.5 and 5.6**).

Sublethal effects were reported in the mallard acute oral toxicity study (MRID 00159201). Toxic symptoms included depression, reduced reaction to external stimuli, wing droop, convulsions, and salivation. In ducks, pheasants and/or partridges (MRID 00060823), toxic symptoms included goose-stepping, ataxia, dyspnea, lacrimation, salivation, ataxia, wing-spread seizures, and terminal opisthotonos. Symptoms appeared as soon as 20 minutes after dosage; recovery of survivors took 1-2 days. The NOAELs and LOAELs for these sublethal effects were not reported.

There are no other acute or chronic toxicity data with birds that have lower endpoints than the studies used in this assessment.

Four incidents were reported in which red-tailed hawks were found either dead or injured after potential exposure to methidathion. One incident was classified as possible, two as probable and the fourth as highly probable. The incidents were all reported in 1994 in several counties in California (Colusa, Stanislaus and Merced). In one case, blood plasma cholinesterase and acetylcholinesterase levels were within the normal range; however, the results of the footwash showed detections of methidathion at 2.7 ppb (other organophosphates, including chlorpyrifos and diazinon were also detected in the footwash sample at respective concentrations of 0.2 ppb and 4.1 ppb). In a second case, the hawk recovered. Plasma cholinesterase and acetylcholinesterase levels were found significantly below the normal range. Footwash analysis results show that methidathion was detected at a concentration of 2.7 ppb. Again, chlorpyrifos and diazinon were also detected in the footwash sample at respective concentrations of 0.7 ppb and 0.4 ppb. In the third case where the hawk died, the plasma cholinesterase level was low and the brain cholinesterase level was severely depressed. Methidathion was detected at a concentration of 0.7 ppm in the footskin. Chlorpyrifos and diazinon were detected in both the feathers and footskin at concentrations ranging from 0.02 to 0.08 ppb. It is likely that all three organophosphates contributed to the death of this bird.

In summary, the acute dose-based, acute dietary-based and/or chronic dietary-based RQs for the terrestrial-phase CRLF, using avian data as a surrogate, exceed the acute listed species LOC (0.1) and/or the chronic non-listed species LOC (1) for all of the assessed methidathion uses. In addition, the probability of an individual effect on an acute basis is high at the RQ levels. Incident data indicate that birds are vulnerable. Spatial analyses indicate that a significant overlap will exist between the use sites for methidathion and the CRLF habitat, particularly when the spraydrift buffers are applied to the “footprint” of the methidathion use area. Therefore, based on the weight-of-evidence, there is a potential for direct impact to the terrestrial-phase CRLF using the endpoints generated from the avian data and the effects determination is LAA.

As stated previously, the only detected degradate of toxicological concern is the minor degradate methidathion oxon, which may be more toxic than the parent. However, no data are available on the environmental fate, toxicity, and occurrence of methidathion oxon in the environment and so it is not considered directly in calculated RQs. Given that acute and chronic risk quotients are exceeded for the parent compound alone, any

contribution in toxicity from any of the major degradates or from methidathion oxon or other OP degradates would increase the risk estimates. Further characterization of risk from methidathion oxon is provided in **Section 5.2.1.3**.

5.2.1.3 Direct Effects from Methidathion Oxon to Aquatic-Phase and Terrestrial-Phase CRLF

As described in **Section 2.1.4**, methidathion oxon formation in soil was demonstrated in soil photolysis, aerobic soil metabolism, and terrestrial field dissipation studies, and formation on foliage was demonstrated in dislodgeable residue studies. Long range transport has also been shown to occur, with methidathion oxon detected in air and on pine needles at more than 20 km from the nearest use site. Although formed at less than 10%, methidathion oxon is a degradate of concern based on comparison with other OP pesticides (*e.g.*, diazinon, chlorpyrifos), for which oxon degradates have been shown in many cases to be of equal or greater toxicity than the parent. This analysis does not include consideration of other oxon degradates which, based on the degradation pathway for methidathion, may have been formed but were not detected in fate studies due to the lack of appropriate labeling.

No environmental fate or ecological effects data are available for methidathion oxon. Therefore, it is difficult to quantify the increased risk which may result from potential exposure to this degradate; however, based on conservative assumptions, some broad conclusions can be reached. In laboratory studies, transformation to the oxon occurred at maximum amounts under soil photolysis conditions, where 5% of the applied parent was present as methidathion oxon at study termination. For quantitative modeling purposes, then, the assumption can be made that methidathion oxon is initially present on soil and on foliage at 5% of the initially applied parent compound.

For both aquatic modeling using PRZM/EXAMS and terrestrial modeling using T-REX, EECs are linear with the application rate. Therefore, based on an application rate of 5% of the applied methidathion for the methidathion oxon, and assuming that its fate properties are similar to the parent, aquatic and terrestrial EECs for the oxon alone would be approximately 5% of those estimated for the parent⁷. If the oxon were of equal toxicity to the parent, then the acute aquatic and terrestrial RQs for the oxon alone would be 5% of those calculated for the parent. For acute aquatic risk, the acute RQs would range from 0.01 for the use on mangos to 1.12 for almonds and 2.62 for the nursery use, as shown in **Table 5.16**. For terrestrial risk, acute dose-based RQs would range from 0.62 for mangos to 11.1 for citrus and 22.1 for the nursery use, as shown in **Table 5.17**. Therefore, at equal toxicity to the parent, risk from the oxon degradate alone would exceed the endangered species LOCs for acute risk to both the aquatic phase CRLF (LOC = 0.05) and the terrestrial phase CRLF (LOC = 0.1) for all except the lowest application rates.

⁷ Modeled aquatic EECs for the oxon would not be exactly 5% because spray drift would not be a factor and so model inputs would change to 100% application efficiency and 0% drift, rather than 95% and 2%, respectively. This, along with a correction for molecular weight, would be likely to change the percentage slightly but not enough to affect conclusions.

However, oxons can be considerably more toxic than the parent and risk from methidathion oxon alone could be even greater than that from the parent. In the cumulative assessment for OP insecticides⁸, if there were no toxicity data available for a given OP-oxon, high-end adjustment factors of 10X to 100X were applied to account for the presumed increased toxicity of the oxon relative to the parent chemical. To explore the potential risk if methidathion oxon were more toxic than the parent, RQs based on these factors are reported as well in **Tables 5.16** and **5.17**, for direct effects to the aquatic and terrestrial phase CRLF, respectively.

Table 5.16 Summary of Direct Effect Acute RQs for the Aquatic-phase CRLF, based on exposure to methidathion oxon degradate				
Use	Parent Only ^a	Oxon Only (no parent) ^b		
		Equal Toxicity	10X Toxicity	100X Toxicity
Mango	0.20	0.01	0.1	1.0
Alfalfa	2.1	0.10	1.0	10
Almond	22.4	1.12	11.2	112
Nursery	52.5	2.62	26.2	262

^a Previously calculated in Table 5.1 using bluegill sunfish (*Lepomis macrochirus*) acute 96-hour LC₅₀ = 2.2 µg/L

^b Assumes EECs to be 5% of those for the parent, based on the maximum percent formation observed in laboratory studies.

Table 5.17 Summary of Direct Effect Acute Dose-based RQs for the Terrestrial-phase CRLF, based on exposure to methidathion oxon degradate				
Use	Parent Only ^a	Oxon Only (no parent) ^b		
		Equal Toxicity	10X Toxicity	100X Toxicity
Mango	12.4	0.62	6.2	60
Clover	65.4	3.3	33	330
Citrus	222.9	11.1	111	1110
Nursery	442.0	22.1	221	2210

^a Previously calculated in Table 5.5 based on mallard duck acute oral LD₅₀ = 6.7 mg/kg.

^b Assumes EECs to be 5% of those for the parent, based on the maximum percent formation observed in laboratory studies.

There is a great deal of uncertainty in these estimates because of the assumptions on which they are based, including those for formation (e.g., fate properties) and toxicity of the methidathion oxon. For the aquatic-phase CRLF, exposure estimates are based on the assumptions that all of the applied methidathion is available on the soil surface for photolysis and that all of the transformation product is available for runoff at one time. For the terrestrial-phase CRLF, exposure estimates assume that laboratory soil photolysis rates are also reflective of the photolysis rates that may occur on foliage. Additionally, the fate properties of methidathion oxon are unknown, and assuming the same fate behavior of the parent may lead to overestimation because OP oxons tend to be more

⁸ More detail on the OP cumulative assessment and the characterization of additional risk due to oxon occurrence may be found at http://www.epa.gov/pesticides/cumulative/2006-op/op_cra_appendices_part2.pdf

transient than the parent. Methidathion oxon has not been detected in surface water monitoring done in two studies, even in samples in which the parent methidathion was detected. Despite the uncertainty, the potential for acute direct effects to aquatic-phase and terrestrial-phase CRLF from exposure to methidathion oxon alone cannot be precluded, even if the toxicity is no greater than that of the parent. Exposure from the parent methidathion already leads to conclusions of risk; thus, consideration of the oxon degradate as well increases the likelihood of direct effects.

5.2.2 Indirect Effects (via Reductions in Prey Base)

5.2.2.1 Algae (non-vascular plants)

As discussed in Section 2.5.3, the diet of CRLF tadpoles is composed primarily of unicellular aquatic plants (i.e., algae and diatoms) and detritus. Due to a lack of aquatic plant toxicity data indirect effects to the CRLF via direct toxicity to aquatic plants cannot be quantitatively estimated for methidathion. There are no incident data for aquatic plants and no relevant data in the open literature. An examination of the completed CRLF assessments for nine other organophosphates indicates that nearly all of the effects determinations for aquatic non-vascular plants were either “no effect” or “not likely to adversely affect”. In addition, the mode of action for methidathion as an organophosphate insecticide is by disrupting nervous system function of exposed animals via acetylcholinesterase inhibition. Unless there is a separate herbicidal mode of action as with bensulide, a pre-emergent organophosphate herbicide which inhibits cell division in root tips and inhibits seedling growth by conjugation of acetyl co-enzyme A, this mode of action on animals is not expected to affect plants. Methidathion also has a history of being applied to a myriad of agricultural crops (as per the label), with no known incidents of adverse phytotoxic effects to aquatic plants. Therefore, based on the weight-of-evidence, the potential indirect impact to the CRLF is expected to be minimal. The effects determination is may affect, not likely to adversely affect (NLAA).

5.2.2.2 Aquatic Invertebrates

The potential for methidathion 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 range from 1.5 to 38.5, thus exceeding the acute risk LOC for non-listed species for all of the assessed uses (LOC = 0.5; see **Table 5.3**). Chronic RQs for freshwater invertebrates range from 8.9 to 145.0, thus exceeding the LOC (1.0) for all of the assessed methidathion uses (see **Table 5.4**). Only one other acute toxicity study with the same species (*daphnia magna*, MRID 00011350) provides any other data relevant to this assessment, either from submitted studies or from the open

literature. With the endpoint from that study, the lowest acute RQ would be 1.0, which still exceeds the acute LOC for non-listed species. At the lowest application rate with the default slope of 4.5 and the lowest RQ of 1.5, the probability of an individual effect is approximately 1 in 1.3 and the percentage effect to the aquatic invertebrate prey base is 79% (e.g., the percentage of the aquatic invertebrate population that is expected to be affected following exposure to methidathion). Based on the weight-of-evidence, there is a potential indirect impact to the CRLF via effects on freshwater invertebrate food items. The effects determination is LAA.

5.2.2.3 Fish and Aquatic-phase Frogs

Methidathion is very highly toxic to freshwater fish, the surrogate for the aquatic-phase CRLF. The acute LOC for non-listed species (all uses) and the chronic LOC (all uses) are exceeded; however, one study in the open literature indicates that there will be some fish species that may not be at risk following acute exposure from some of the methidathion use scenarios. At the lowest application rate, the probability of an individual effect on an acute basis at the RQ is approximately 1 in 1 and the percentage effect to the fish and aquatic-phase frog prey base is 93.4%. In addition, the incident data indicate that freshwater fish are vulnerable. Therefore, based on the weight-of-evidence, there is a potential indirect impact to the aquatic-phase CRLF based on the endpoints generated from the freshwater fish data. The effects determination is LAA.

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 RQs for both small and large insects exceed the acute LOC for non-listed species for all methidathion uses, even with only a single use. The acute RQs for small insects range from 21 (mangos) to 734 (nursery) and the acute RQs for large insects range from 2.3 (mangos) to 82 (nursery). Following a single use, the lowest RQ is 2.0 for large insects (mangos). There are insufficient studies to conduct a sensitivity analysis; however, the highest terrestrial invertebrate endpoint is from the open literature with an acute toxicity value of 0.416 µg/bee (ECOTOX reference 70351). The lowest RQ, using the endpoint from this study is 1.1 (mangos), which still exceeds the acute list species LOC of 0.05. The probability of an individual effect at the LOC of 0.5 is 1 in 4.41E+31 and the probability of an individual effect at the RQ for all uses is 1 in 1. In addition, the percentage effect to the terrestrial invertebrate prey base is 99.9 - 100%.

Two incidents were reported following application of methidathion in California. The first involved spraying of plum trees near some pollination hives. Dead bees (number not specified) were found along with residues of methidathion and diazinon. This incident was rated as possible. The second incident reported that an unknown number of bees were killed in an unknown county. The incident was attributed to accidental misuse. No residue analysis was completed; however, the certainty index for this incident is probable.

Based on the weight-of-evidence, there is a potential indirect impact to the CRLF based on this endpoint. The effects determination is LAA.

5.2.2.5 Mammals

Life history data for terrestrial-phase CRLFs indicate that large adult frogs consume terrestrial vertebrates, including mice. The acute and chronic RQs exceed the listed species acute LOC and the chronic LOC for all uses. The acute RQs range from 2.4 (mangos) to 86.8 (nursery). The chronic dietary RQs range from 117 (mangos) to 4164.5 (nursery, dose-based) and from 13.5 (mangos) to 480.0 (nursery, dietary-based). There is one study from the open literature conducted with the mouse (*mus musculus*) that appears to have a lower endpoint (E85173); however, the study is not available for review at this time. The acute LD₅₀ for this study is 10.5 mg a.i./kg. Using the endpoint from this study and assuming that a mouse weighs 20 g, the lowest RQ (mangos) would be 5.70, which exceeds the non-listed species LOC by 10 times. No other relevant studies are available in the open literature. At the lowest application rate (0.25 lb a.i./A for mangos), with the default slope of 4.5 and the lowest RQ of 2.4, the probability of an individual effect is approximately 1 in 1 and the percentage effect to the mammalian prey base is 95.6%. Based on the weight-of-evidence, the labeled uses for methidathion may indirectly impact the CRLF through effects to the mammalian prey base. The effects determination is LAA.

5.2.2.6 Terrestrial-phase Amphibians

Terrestrial-phase adult CRLFs also consume frogs. RQ values representing direct exposures of methidathion to terrestrial-phase CRLFs are used to represent exposures of methidathion to frogs in terrestrial habitats. With the exception of mangos on an acute dietary basis, the acute dose-based, acute dietary-based and/or chronic dietary-based RQs for terrestrial-phase amphibians, using avian data as a surrogate, exceed the acute non-listed species LOC and the chronic LOC for all of the assessed methidathion uses. Refining the RQ values with T-HERPS, the acute dose-based RQs still exceed the non-listed LOC (0.5) for at least one food category for all of the modeled scenarios for methidathion.

Table 5.18. Upper Bound Kenaga, Acute Terrestrial Herpetofauna Dose-Based Risk Quotients											
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 ¹	EEC	RQ ¹	EEC	RQ ¹	EEC	RQ ¹	EEC	RQ ¹
ARTICHOKE											
1.4	6.70	6.81	1.02	0.76	0.11	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	6.69	1.00	0.74	0.11	194.24	28.99	12.14	1.81	0.23	0.03
238	6.70	4.39	0.65	0.49	0.07	30.20	4.51	1.89	0.28	0.15	0.02
CITRUS											

Table 5.18. Upper Bound Kenaga, Acute Terrestrial Herpetofauna Dose-Based Risk Quotients											
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 ¹	EEC	RQ ¹	EEC	RQ ¹	EEC	RQ ¹	EEC	RQ ¹
1.4	6.70	26.46	3.95	2.94	0.44	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	26.00	3.88	2.89	0.43	754.62	112.63	47.16	7.04	0.90	0.13
238	6.70	17.04	2.54	1.89	0.28	117.31	17.51	7.33	1.09	0.59	0.09
CLOVER											
1.4	6.70	7.76	1.16	0.86	0.13	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	7.63	1.14	0.85	0.13	221.32	33.03	13.83	2.06	0.26	0.04
238	6.70	5.00	0.75	0.56	0.08	34.41	5.14	2.15	0.32	0.17	0.03
COTTON											
1.4	6.70	11.27	1.68	1.25	0.19	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	11.07	1.65	1.23	0.18	321.38	47.97	20.09	3.00	0.38	0.06
238	6.70	7.26	1.08	0.81	0.12	49.96	7.46	3.12	0.47	0.25	0.04
KIWI											
1.4	6.70	10.49	1.57	1.17	0.17	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	10.31	1.54	1.15	0.17	299.20	44.66	18.70	2.79	0.36	0.05
238	6.70	6.76	1.01	0.75	0.11	46.51	6.94	2.91	0.43	0.23	0.04
MANGO											
1.4	6.70	1.47	0.22	0.16	0.02	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	1.45	0.22	0.16	0.02	42.03	6.27	2.63	0.39	0.05	0.01
238	6.70	0.95	0.14	0.11	0.02	6.53	0.98	0.41	0.06	0.03	0.00
NURSERY											
1.4	6.70	2.62	0.39	0.29	0.04	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	2.58	0.38	0.29	0.04	74.80	11.16	4.67	0.70	0.09	0.01
238	6.70	1.69	0.25	0.19	0.03	11.63	1.74	0.73	0.11	0.06	0.01
SAFFLOWER, SUNFLOWER											
1.4	6.70	4.48	0.67	0.50	0.07	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	4.41	0.66	0.49	0.07	127.85	19.08	7.99	1.19	0.15	0.02
238	6.70	2.89	0.43	0.32	0.05	19.88	2.97	1.24	0.19	0.10	0.01
TREE FRUIT, OLIVES											
1.4	6.70	15.73	2.35	1.75	0.26	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	15.46	2.31	1.72	0.26	448.79	66.98	28.05	4.19	0.54	0.08
238	6.70	10.14	1.51	1.13	0.17	69.77	10.41	4.36	0.65	0.35	0.05
WALNUT											
1.4	6.70	25.88	3.86	2.88	0.43	N/A	N/A	N/A	N/A	N/A	N/A
37	6.70	25.44	3.80	2.83	0.42	738.28	110.19	46.14	6.89	0.88	0.13
238	6.70	16.67	2.49	1.85	0.28	114.77	17.13	7.17	1.07	0.58	0.09
^a Acute RQs that exceed the acute non-listed species LOC of 0.5 are in bold .											

In addition, on an acute dose-basis, the probability of an individual effect is 1 in 1 at the lowest RQ level and the percentage effect to the avian/terrestrial-phase amphibian prey base is 100%. Incident data indicate that birds are vulnerable. Therefore, based on the

weight-of-evidence, there is a potential indirect impact to the terrestrial-phase CRLF based on these endpoints. The effects determination is LAA.

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 as attachment sites and refugia for many aquatic invertebrates, fish, and juvenile organisms, such as fish and frogs. In addition, vascular plants also provide primary productivity and oxygen to the aquatic ecosystem. Rooted 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 would normally be assessed using RQs from freshwater aquatic vascular and non-vascular plant data; however, there are no aquatic plant data for methidathion and quantitative risk estimations cannot be conducted. There are no incident data for aquatic plants and no relevant data in the open literature. An examination of the completed CRLF assessments for nine other organophosphates indicates that nearly all of the effects determinations for aquatic vascular and non-vascular plants were either no effect or not likely to adversely affect. Not all of the other CRLF organophosphate chemicals have aquatic vascular plant data. In those cases, determinations were based on the non-vascular plant data and any available terrestrial plant data. As stated previously, the mechanism of action for methidathion as an organophosphate insecticide is by disrupting nervous system function of exposed animals via acetylcholinesterase inhibition, which is not a mode of action that is expected to affect plants. Methidathion also has a history of being applied to a myriad of agricultural crops (as per the label), with no known incidents of adverse phytotoxic effects to aquatic plants. Therefore, based on the weight-of-evidence, the potential indirect impact to the CRLF is minimal. The effects determination is may affect, NLAA.

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. Terrestrial plants also provide energy to the terrestrial ecosystem through primary production. 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.

Again, as with aquatic plants, there are no registrant-submitted terrestrial plant toxicity data for methidathion for assessment of the potential for indirect effects to the aquatic- and terrestrial-phase CRLF via effects to riparian vegetation or effects to the primary constituent elements (PCEs) relevant to the aquatic- and terrestrial-phase CRLF. However, there is limited evidence in the open literature that methidathion has the potential to elicit phototoxic effects (Ecotox ref. 64451). This paper reported that methidathion significantly affected corn photosynthesis when applied at 0.5 lbs a.i./A, a rate that is lower than many of the currently registered rates. In addition, potential phytotoxic effects have been highlighted on methidathion labels (i.e., spotting, reddening, or chlorosis of the leaves) in certain sorghum varieties. An examination of the completed CRLF assessments for nine other organophosphates indicates that the majority of the effects determinations for terrestrial plants were either “no effect” or “not likely to adversely affect”. Two were determined to be “LAA”; however, one had herbicidal activity from a known mechanism and the other used surrogate data from another pesticide. For some of these organophosphates, as with the study mentioned above, there is the potential for some damage to plants. Nevertheless, the conclusions in those cases were generally that while effects to terrestrial plants may affect the CRLF via habitat modification, they are not likely to adversely affect the CRLF based on the type and extent of damage as observed. The mode of action for methidathion as an organophosphate insecticide is not one known to affect plants. Therefore, based on the weight-of-evidence, methidathion is not likely to impact plants to an extent that is expected to adversely affect the CRLF at the labeled application rates. Although some phytotoxicity could occur, these effects are considered to be insignificant in the context of a “take”. The effects determination is may affect, NLAA.

5.2.4 Modification to Designated Critical Habitat

The risk conclusions for the designated critical habitat are based on conclusions described for indirect effects previously described. Potential habitat modification is described below.

5.2.4.1 Aquatic-Phase PCEs

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

- Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.
- Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.
- Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.*, algae).

Conclusions for potential indirect effects to the CRLF via direct effects to aquatic and terrestrial plants are used to determine whether modification to critical habitat may occur. The potential for habitat modification via impacts to aquatic plants (Sections 5.2.2.1 and 5.2.3.1) and terrestrial plants (5.2.3.2) is not considered to be significant. No habitat modification is expected.

The remaining aquatic-phase PCE is “alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.” Other than impacts to algae as food items for tadpoles (discussed above), this PCE is assessed by considering direct and indirect effects to the aquatic-phase CRLF via acute and chronic freshwater fish and invertebrate toxicity endpoints as measures of effects. Based on the potential direct impact to the aquatic-phase CRLF (Section 5.2.1.1) and impacts to freshwater invertebrates and fish as food items (Sections 5.2.2.2 and 5.2.2.3), there is a potential for habitat modification.

5.2.4.2 Terrestrial-Phase PCEs

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

- Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or drip line surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance.
- Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal.

No habitat modification is expected through impacts to terrestrial plants (5.2.3.2).

The third terrestrial-phase PCE is “reduction and/or modification of food sources for terrestrial phase juveniles and adults.” To assess the impact of methidathion on this PCE, acute and chronic toxicity endpoints for terrestrial invertebrates, mammals, and terrestrial-phase frogs are used as measures of effects. There is a potential for habitat modification based on potential reductions in prey base (Section 5.2.2.4 for terrestrial invertebrates, Section 5.2.2.5 for mammals, and 5.2.2.6 for frogs).

The fourth terrestrial-phase PCE is based on alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source. There is a potential for habitat modification based on potential direct (Section 5.2.1.2) and indirect effects (Sections 5.2.2.4, 5.2.2.5, and 5.2.2.6) to terrestrial-phase CRLFs.

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. In particular, the application rate assumed for use on nursery stock is conservative. The label does not provide explicit application rates for this use and so it is assessed based on an assumption of application at the general maximum application rate required by the labels.

6.1.2 Aquatic Exposure Modeling of Methidathion

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 m³) pond with no outlet. Exposure estimates generated using the EXAMS pond are intended to represent a wide variety of vulnerable water bodies that occur at the top of watersheds including prairie pot holes, playa lakes, wetlands, vernal pools, man-made and natural ponds, and intermittent and lower order streams. As a group, there are factors that make these water bodies more or less vulnerable than the EXAMS pond. Static water bodies that have larger ratios of pesticide-treated drainage area to water body volume would be expected to have higher peak EECs than the EXAMS pond. These water bodies will be either smaller in size or have larger drainage areas. Smaller water bodies have limited storage capacity and thus may overflow and carry pesticide in the discharge, whereas the EXAMS pond has no discharge. As watershed size increases beyond 10-hectares, it becomes increasingly unlikely that the entire watershed is planted with a single crop that is all treated simultaneously with the pesticide. Headwater streams can also have peak concentrations higher than the EXAMS pond, but they likely persist for only short periods of time and are then carried and dissipated downstream.

The Agency acknowledges that there are some unique aquatic habitats that are not accurately captured by this modeling scenario and modeling results may, therefore, under- or over-estimate exposure, depending on a number of variables. For example, aquatic-phase CRLFs may inhabit water bodies of different size and depth and/or are located adjacent to larger or smaller drainage areas than the EXAMS pond. The Agency does not currently have sufficient information regarding the hydrology of these aquatic habitats to develop a specific alternate scenario for the CRLF. CRLFs prefer habitat with perennial (present year-round) or near-perennial water and do not frequently inhabit

vernal (temporary) pools because conditions in these habitats are generally not suitable (Hayes and Jennings 1988). Therefore, the EXAMS pond is assumed to be representative of exposure to aquatic-phase CRLFs. In addition, the Services agree that the existing EXAMS pond represents the best currently available approach for estimating aquatic exposure to pesticides (USFWS/NMFS 2004).

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

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

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

In order to account for uncertainties associated with modeling, available monitoring data were compared to PRZM/EXAMS estimates of peak EECs. The surface water dataset for methidathion has several thousand samples collected from 135 sites primarily in agricultural areas, and includes several monitoring studies targeted to water bodies

receiving runoff from high OP use areas and with a high frequency of sampling, factors which reduce the uncertainty inherent in monitoring data. This monitoring detected methidathion in surface waters at concentrations of up to 15.1 ug/L. Peak modeling EECs range from 7.3 ug/L for alfalfa to 52.4 ug/L for the almond use. (The maximum modeled EEC is 114.7 ug/L for the use on nursery stock, but this result is based on conservative assumptions about application rates.) The highest monitoring detection of 15.1 ug/L is within this range, below the peak EECs for some uses but higher than EECs for others. Although the use in the location with the highest detection is unknown, the majority of the sampling was conducted in areas with high acreage of fruit and nut orchards, which had modeled EECs of 17.8 ug/L for fruit trees, 20.0 ug/L for olives, and 52.4 ug/L, suggesting that the detection is not in excess of relevant modeled EECs. Additionally, the next highest monitored detection from this relatively large dataset is 2.4 ug/L, suggesting that the modeled EECs are reasonably conservative representations of 1-in-10 year concentrations.

6.1.3 Potential Groundwater Contributions to Surface Water Chemical Concentrations

Although the potential impact of discharging ground water on CRLF populations is not explicitly delineated, it should be noted that ground water could provide a source of pesticide to surface water bodies – especially low-order streams, headwaters, and ground water-fed pools. Terrestrial field dissipation studies did not find leaching of methidathion below 12 inches, but given the fact that methidathion is soluble and classified as moderately mobile, and that its abiotic and biotic degradation in anaerobic aquatic environments may be slow, the possibility of methidathion reaching ground water cannot be precluded. Much of available ground water will eventually be discharged to the surface – often supporting stream flow in the absence of rainfall. Continuously flowing low-order streams in particular are sustained by ground water discharge, which can constitute 100% of stream flow during baseflow (no runoff) conditions. Thus, it is important to keep in mind that pesticides in groundwater may have a major (detrimental) impact on surface water quality, and on CRLF habitats.

Concentrations in a receiving water body resulting from groundwater discharge cannot be explicitly quantified, but it can be assumed that significant attenuation and retardation of the chemical will have occurred prior to discharge. Nevertheless, groundwater could still be a significant consistent source of chronic background concentrations in surface water, and may also add to surface runoff during storm events (as a result of enhanced groundwater discharge typically characterized by the ‘tailing limb’ of a storm hydrograph). In this assessment, 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, but this factor remains an uncertainty.

6.1.4 Action Area Uncertainties

An example of an important simplifying assumption that may require future refinement is the assumption of uniform runoff characteristics throughout a landscape. It is well

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

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

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

6.1.5 Usage Uncertainties

County-level usage data were obtained from California's Department of Pesticide Regulation Pesticide Use Reporting (CDPR PUR) database. Eight years of data (1999 – 2006) were included in this analysis. 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.

6.1.6 Terrestrial Exposure Modeling of Methidathion

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.

6.1.7 Spray Drift Modeling

Although there may be multiple methidathion applications at a single site, 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 methidathion from multiple applications, each application of methidathion would have to occur under identical atmospheric conditions (*e.g.*, same wind speed and – for plants – same wind direction) and (if it is an animal) the animal being exposed would have to be present directly downwind at the same distance after each application. Although there may be sites where the dominant wind direction is fairly consistent (at least during the relatively quiescent conditions that are most favorable for aerial spray applications), it is nevertheless highly unlikely that plants in any specific area would receive the maximum amount of spray drift repeatedly. It appears that in most areas (based upon available meteorological data) wind direction is temporally very changeable, even within the same day. 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 even from single applications, especially as the distance increases from the site of application, since the model does not account for potential obstructions (*e.g.*, large hills, berms, buildings, trees, *etc.*). Furthermore, conservative assumptions are often made regarding the application method (*e.g.*, aerial), release heights and wind speeds. Alterations in any of these inputs would change the area of potential effect.

6.2 Effects Assessment Uncertainties

6.2.1 Age Class and Sensitivity of Effects Thresholds

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

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

6.2.2 Use of Surrogate Species Effects Data

Guideline toxicity tests and open literature data on methidathion 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 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.

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 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.

In fish, slight spastic motions and swimming on their sides were observed following acute exposure at 1000 µg/L (MRID 00011841). Following chronic exposure, total length and wet weight are affected (MRID 00015730 or 45822701) with a NOAEC/LOAEC of 6.3/12 µg ai/L respectively. No sublethal effects were reported in the freshwater invertebrate studies.

In birds, depression, reduced reaction to external stimuli, wing droop, convulsions, and salivation were observed following acute exposure via gavage. In subacute dietary studies, toxic symptoms including depression, reduced reaction to external stimuli, wing droop, loss of coordination, lower limb weakness, ruffled appearance, prostrate posture, and loss of righting reflex were observed at 178 ppm a.i. and above. Reductions on body weight gain and food consumption were reduced at levels above 316 ppm a.i.. Following chronic exposure (MRID 44381602), none of the ducks showed symptoms of toxicity or behavioral abnormalities during the experiment. An increase in the number of eggs

cracked and a decrease in the number of eggs not cracked/eggs laid were observed at 10 ppm. Brain cholinesterase was inconsistently affected at 30 ppm.

The study used to define the action area examined the effects of subchronic methidathion administration on vascular wall damage in rats (ECOTOX ref. 80451). At the lowest dose tested (5 mg/kg/day), the levels of malondialdehyde (MDA), a biomarker for oxidative stress was significantly higher and cholinesterase activity was significantly lower. There were irregular, prominent breaks and fragmentation of the elastic fibers in the aortic wall.

In a 2-generation reproduction study in rats (MRID 40079812, -13), the parental systemic NOAEC was 5 ppm and the LOAEC was 25 ppm, based on tremors and decreased food consumption during lactation, and decreased ovarian weight. In addition, there was also a slight decrease in body weight early in the F1 growth phase at 50 ppm. The reproductive NOAEC was 5 ppm and the LOAEC was 25 ppm based on a decreased mating index and a generalized indication of pup unthriftiness while nursing. In addition, there was an increase in stillbirths and decreased pup survival at birth and during lactation at the 50 ppm treatment level.

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

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 methidathion 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 methidathion. Additionally, the Agency has determined that there is the potential for modification of CRLF designated critical habitat from the use of the chemical.

There is potential for direct risk to the aquatic- and terrestrial-phase CRLF following either acute or chronic exposure to methidathion with its current use patterns in California. The acute listed species LOC for freshwater fish (surrogate to the aquatic-

phase CRLF) is exceeded for all uses; however, there are limited data which indicate that there will be some fish species that may not be at risk following acute exposure from some of the methidathion use scenarios. The chronic LOC for freshwater fish is also exceeded for all uses. For the terrestrial-phase CRLF, using avian data as a surrogate, with the exception of mangos on a dietary basis for non-listed species, the acute dose-based, acute dietary-based and chronic dietary-based RQs for the terrestrial-phase CRLF, exceed both the acute listed and non-listed species LOC and chronic LOC for all of the assessed uses.

There is a potential for indirect effects to both the aquatic- and terrestrial-phase CRLF. For aquatic invertebrates (food source), the acute and chronic RQs exceed the acute non-listed LOC for aquatic animals and chronic LOC for aquatic animals respectively, for all assessed uses. For fish and aquatic-phase amphibians (food source), the acute non-listed species LOC is exceeded for all uses. The chronic LOC is also exceeded for all uses. No studies are available for aquatic vascular and non-vascular plants (food source and habitat); however, the weight of the evidence indicates an effect determination of may affect, not likely to adversely affect (NLAA). For terrestrial invertebrates (food source), the RQs for both small and large insects exceed acute LOC for non-listed species for all methidathion uses, even with only a single use. For terrestrial-phase amphibians (food source), see the direct effects paragraph. For mammals (food source), the acute and chronic RQs exceed the non-listed species acute LOC and the chronic LOC, respectively for all uses. No studies are available for terrestrial plants (habitat). There is some evidence for potential for terrestrial plant damage; however, the weight of the evidence indicates that the effect to terrestrial plants is may affect, not likely to adversely affect (NLAA). Habitat modification is expected for both the aquatic- and terrestrial-phase CRLF based on indirect effects from the reduction in prey base. A significant overlap is expected to exist between the use sites and CRLF habitat, particularly when the spraydrift and downstream dilution buffers are added to the methidathion use area. Spraydrift distances range from 23 to greater than 1000 feet and the downstream dilution buffer is 285 km. **Figure 5.1** references the overlap of frog habitat with NLCD land cover data for use patterns that result in LAA determinations. Given the LAA determination for the CRLF and potential modification of designated critical habitat, a description of the baseline status and cumulative effects for the CRLF is provided in **Attachment 2**.

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

Table 7.1 Effects Determination Summary for Methidathion Use and the CRLF		
Assessment Endpoint	Effects Determination ¹	Basis for Determination
Survival, growth, and/or reproduction of CRLF individuals	LAA¹	Potential for Direct Effects
		<i>Aquatic-phase (Eggs, Larvae, and Adults):</i> Highly toxic to freshwater fish. Acute listed species LOC exceeded for all uses for three tested surrogate species. Chronic effects based on survival and growth. Chronic LOC exceeded for all uses. Probability of an individual effect on an acute basis is high, both at the acute LOC and at the RQ levels. Incident data also indicate freshwater fish vulnerability, and monitoring has detected surface water concentrations above the acute endpoint. Significant overlap expected to exist between use sites and CRLF habitat, particularly when the spraydrift and downstream dilution buffers are added to the methidathion use area. Spraydrift distances range from 23 to over 1000 feet and the downstream dilution buffer is 285 km.
		<i>Terrestrial-phase (Juveniles and Adults):</i> Moderately toxic to very highly toxic to avian species. Acute dose-based, acute dietary-based and chronic dietary-based RQs for the terrestrial-phase CRLF, using avian data as a surrogate, exceed acute listed species LOC and chronic LOC for all of the assessed uses. Chronic effects based on reduction in number of normal hatchlings/live 3-week embryos. Limited data indicate that on a dose basis, the acute LOC is exceeded for all uses, even with the least sensitive species at the lowest application rate (mangos). On a dietary basis, the same is true for all uses except mangos. Probability of an individual effect on an acute basis is high at the RQ levels. Incident data indicate that birds are vulnerable. Significant overlap exists between use sites and CRLF habitat, particularly when the spraydrift buffers are applied to the methidathion use area. Spraydrift distances range from 194 to over 1000 feet.
		Potential for Indirect Effects
		<i>Aquatic prey items, aquatic habitat, cover and/or primary productivity</i> Very highly toxic to freshwater invertebrates. Acute and chronic RQs exceed acute LOC for non-listed species and chronic LOC, respectively for all assessed uses. The percentage effect to the aquatic invertebrate prey base for all uses is very high. For fish (aquatic-phase amphibians), acute non-listed species LOC exceeded for all uses. Chronic LOC exceeded for all uses. Percentage effect to the freshwater fish/aquatic-phase amphibian prey base is very high. No studies are available for aquatic vascular and non-vascular plants.
		<i>Terrestrial prey items, riparian habitat</i> RQs for both small and large insects exceed acute LOC for non-listed species for all methidathion uses, even with only a single use. Insufficient studies to conduct a sensitivity analysis; however, lowest RQ, using highest terrestrial invertebrate endpoint still exceeds the acute list species LOC. Percentage effect to the terrestrial invertebrate prey base for all uses is very high. For terrestrial-phase amphibians using avian data as a surrogate, with the exception of mangos on an acute dietary basis, the acute dose-based, acute dietary-based and chronic dietary-based RQs exceed the acute non-listed species LOC and the chronic LOC for all of the assessed methidathion uses, respectively. Percentage effect to the avian/terrestrial-phase amphibian prey base is very high. For mammals, the acute and chronic RQs exceed the non-listed species acute LOC and the chronic LOC, respectively for all uses. At the lowest RQ, the percentage effect to the mammalian prey base is very high. No studies are available for terrestrial plants.

¹ No effect (NE); May affect, but not likely to adversely affect (NLAA); May affect, likely to adversely affect (LAA)

Table 7.2 Effects Determination Summary for Methidathion Use and CRLF Critical Habitat Impact Analysis		
Assessment Endpoint	Effects Determination ¹	Basis for Determination
Modification of aquatic-phase PCE	Habitat Modification ¹	<p>No studies available for aquatic vascular and non-vascular plants and terrestrial plants.</p> <p>For the aquatic-phase CRLF, acute non-listed species LOC for freshwater fish (aquatic-phase amphibians) exceeded for all uses. Chronic LOC for freshwater fish exceeded for all uses. Probability of an individual effect and percentage effect to the freshwater fish/aquatic-phase amphibian prey base on an acute basis at the RQ are high. Incident data support freshwater fish vulnerability. For freshwater invertebrates, acute and chronic RQs exceed acute LOC (non-listed species) and chronic LOC, respectively for all assessed uses. Again, the percentage effect to the aquatic invertebrate prey base is very high. Significant overlap expected between use sites and CRLF habitat, particularly when the spraydrift and downstream dilution buffers are added to the use area. Spraydrift distances range from 23 (mangos) to over 1000 feet and the downstream dilution buffer is 285 km.</p>
Modification of terrestrial-phase PCE		<p>No studies are available for terrestrial plants.</p> <p>For the terrestrial-phase CRLF, using avian data as a surrogate, with the exception of mangos on a dietary basis for non-listed species, the acute dose-based, acute dietary-based and chronic dietary-based RQs for the terrestrial-phase CRLF, exceed both the acute listed and non-listed species LOC and chronic LOC for all of the assessed uses. Probability of an individual effect on an acute basis at the lowest RQ level and percentage effect to the avian/terrestrial-phase amphibian prey base are very high. Incident data indicate that birds are vulnerable. RQs for both small and large insects exceed acute LOC for non-listed species for all methidathion uses, even with only a single use. Percentage effect to terrestrial invertebrate prey base for all uses are very high. For mammals, the acute and chronic RQs exceed the non-listed species acute LOC and the chronic LOC, respectively for all uses. The percentage effect to the mammalian prey base is very high for all uses. Significant overlap exists between use sites and CRLF habitat, particularly when the spraydrift buffers are applied to the methidathion use area. Spraydrift distances range from 194 (mangos) to over 1000 feet.</p>

¹ Habitat Modification or No effect (NE)

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.

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