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

## Pesticide Effects Determination

**Environmental Fate and Effects Division**
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## *1.0 Executive Summary*

This ecological risk assessment evaluates the potential for the use of the herbicide metolachlor (PC#108801) to affect the California red-legged frog (*Rana aurora draytonii*), and/or modify its designated critical habitat. The California red-legged frog (CRLF) was federally listed as a threatened species by the U.S. Fish and Wildlife Service (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). Final critical habitat for the CRLF was designated by USFWS on April 13, 2006 (USFWS 2006; 71 FR 19244-19346). The CRLF is endemic to California and Baja California (Mexico) and historically inhabited 46 counties in California, including the Central Valley and both the coastal and interior mountain ranges (USFWS 1996). Its range has been reduced by approximately 70%, and it currently inhabits 22 counties in California (USFWS 1996). This assessment is being undertaken consistent with the settlement for the court case *Center for Biological Diversity (CBD) vs. EPA et al*. (Case No. 02-1580-JSW(JL)).

Metolachlor (PC#108801) is a pre-plant, pre-emergence herbicide, currently registered for use only on agricultural crops. As originally registered, metolachlor (PC#108801) is a racemic mixture of *r*- and *s*-enantiomers. Of these enantiomers, the *s*-enantiomer has been shown to be more biologically active in plants, and an enriched technical product is registered separately (PC#108800) as S-metolachlor. This assessment addresses use of racemic metolachlor, although toxicity data from both chemicals has been included.

Metolachlor is persistent and mobile in soil. It is highly persistent in water and has been detected extensively in both surface and ground water. It is a biosynthesis inhibitor, absorbed through the roots and the shoots of the plant. Metolachlor is slightly to moderately toxic to freshwater fish, amphibians, and freshwater invertebrates on an acute basis ($LC_{50}$s 1.1-26 mg/L). Toxicity to aquatic plants ($EC_{50}$s) ranges from 0.008–1.2 mg/L. Metolachlor has two major[1] degradates, metolachlor oxanilic acid (OA) and metolachlor ethane sulfonic acid (ESA) that have been identified as potentially of toxicological concern. Both degradates are less toxic than the parent metolachlor and are considered in the risk assessment.

[1] Defined as ≥10% of applied.

The California red-legged frog inhabits a mosaic of aquatic and upland habitat that it requires to complete its life history. This assessment considers direct and indirect effects on the frog and its critical habitat. For clarity and ease of understanding, the lifecycle of the frog was separated into an aquatic phase and a terrestrial phase, as the exposure and effects modeling for these two ecosystems are different. The aquatic phase includes eggs, larvae, tadpoles, juveniles, and adults. Although juveniles and adults spend a significant amount of time in terrestrial habitats, they also use the aquatic portion of their habitat, especially during breeding. The terrestrial phase evaluation includes juveniles and adults. Components of the ecosystem addressed in the assessment include aquatic plants, aquatic invertebrates, fish, terrestrial plants, terrestrial invertebrates, and terrestrial vertebrates (*e.g.* small mammals,) in addition to the various life stages of the frog itself.

Based on the screening level assessment, no LOCs were exceeded for direct effects on lifestages of the frog present in the aquatic environment. No acute LOCs were exceeded for aquatic prey. The chronic LOC for aquatic invertebrates was exceeded for all crop uses, but based on additional analysis, chronic effects to aquatic invertebrates appear unlikely to occur. LOCs for both vascular and non-vascular aquatic plants were exceeded for use on sorghum, and the LOC for non-vascular plants were exceeded for soybeans. CDPR PUR data shows no use of metolachlor on sorghum in the years for which data were available (2002-2005), and NASS data indicates soybeans are not generally grown in California. Reduction of aquatic plant biomass or populations as a result of metolachlor input are expected to be temporary, and these indirect effects are not anticipated to measurably affect frogs present in affected waterbodies. In the terrestrial phase portion of the assessment, LOCs were exceeded for small frogs, which constitutes both direct (on the California red-legged frog itself) and indirect (on frogs that may be prey) effects. Based on two refined analyses, one incorporating amphibian/reptile specific allometric equations to address dose and a drift analysis to examine the range of off-site effects, effects on the frogs appear unlikely to occur. Screening level LOCs were also exceeded for terrestrial invertebrates. Based on the toxicity data, which did not establish a definitive effects endpoint, and an analysis of the extent of possible off-site effects, effects on the frog appear unlikely.

For all uses, LOCs for terrestrial plants were exceeded. Clearance distances for the most sensitive endpoints were used to establish the action area (2,060 feet from NLCD-classified agricultural land use areas). The action area intersects with approximately 1,445 square miles of California red-legged frog habitat.[2] (Metolachlor is a biosysnthesis inhibitor, absorbed by the roots and shoots of the plant (www.syngentacropprotection-us.com/prod/herbicide/dualiimagnum). It translocates in the plant. Germinating monocots primarily absorb metolachlor through the shoot just above the seed, and germinating dicots absorb at the shoot and the roots (Zimdahl 1993).

[2] For the purposes of this assessment, CRLF habitat is defined as CNDDB occurrence sections, occupied core habitat as defined in the recovery plan, and designated critical habitat.

Terrestrial plants serve several important functions in the California red-legged frog habitat. Vegetation provides cover from predators while the frog is foraging as well as providing habitat and food sources for both its invertebrate and vertebrate prey. In the riparian and upland systems the Califronia red-legged frog occupies, there is generally some type of scrub or shrub understory providing some protection for the herbaceous plants most affected by metolachlor, and metolchlor is not expected to be deposited on bare ground or newly emerging seedlings. Effects are likely to be more closely approximated by the vegetative vigor endpoints than the seedling emergence endpoints However, EFED anticipates adverse effects that could reasonably be measured would be defined by the vegetative vigor endpoint. Thus, for ground applied metolachlor, adverse effects might reasonably be expected to occur up to 90 feet from the use site, and for aerially-applied metolachlor, adverse effects might reasonably be expected to occur up to 1000 feet from the use site.

After completing the analysis of the effects of metolachlor on the Federallly listed threatened California red-legged frog *(Rana aurora draytonii)* in accordance with methods delineated in EPA's Overview document (USEPA 2004), EFED concludes that the use of metolachlor (PC#108801) may affect, and is likely to adversely affect the California red-legged, based on indirect effects (habitat modification to terrestrial plants). EFED also concludes that these same effects constitute adverse modification to critical habitat. These effects are anticipated to occur only for those occupied core habitat areas, CNDDB occurrence sections, and designated critical habitat for the California red-legged frog that are located ≤ 1000 feet from legal use sites where metolachlor is aerially applied and ≤90 feet from ground applications. For this assessment, legal use sites are defined as NLCD-classified "agricultural lands." Potential but not anticipated indirect effects include reduction of the prey base and/or reduction of primary productivity in waters receiving runoff from fields treated with metolachlor. Table 1 describes the rationale for each component assessed. A baseline status and cumulative effects description, based on information provided by the U.S. FWS, is provided in Attachment 2.

Using ARGIS9, the NLCD classified data, and CLRF habitat information supplied by the U.S. FWS, EFED has identified the habitat areas where indirect effects are anticipated to occur (Figure 16) and designated critical habitat areas where adverse modifications are anticipated to occur (Figure 17). Indirect effects (modification of the terrestrial plant community) could potentiallyoccur in approximately 9% (~620,000 acres) of the CLRF range assessed, and adverse modification to designated critical habitat could potentially occur in 0.003% (~14 acres) of the currently designated area.

**Table 1  Effects Determination for Metolachlor**

| Assessment Endpoint | Effects determination | Basis for Determination |
|---|---|---|
| *Aquatic Phase (Eggs, larvae, tadpoles, juveniles, and adults)*[a] | | |
| *Direct Effects* | | |
| 1. Survival, growth, and reproduction of CRLF | No effect | No LOC exceedences for any life stage |
| *Indirect Effects* | | |
| 2. Reduction or modification of aquatic prey base | May affect<br>Not likely to adversely affect<br>(Discountable) | Chronic exceedences for aquatic invertebrates. No LOC exceedences for any other aquatic prey items. Based on analysis of full toxicity data set, monitoring data, and modeled EECs, chronic effects on aquatic invertebrates appear unlikely. |
| 3. Reduction or modification of aquatic plant community | May affect<br>Not likely to adversely affect<br>(Discountable) | Exceedences for both vascular and non-vascular plants for sorghum, and for non-vascular plants for soybeans. CDPR PUR data report no usage on sorghum, and soybeans are not grown in CA. |
| 4. Degradation of riparian vegetation | May affect<br>NLAA >1000 ft<br>LAA <1000ft (aerial)<br>LAA <90ft (ground)<br>Modification to critical habitat<br>Adverse <1000ft (aerial)<br>Adverse <90ft (ground) | Exceedences for both monocots and dicots in both wetlands and uplands adjacent to use site for all crops registered.<br><br>Exceedences for both monocots and dicots near use site based on spray drift alone for all crops registered. |
| *Terrestrial Phase (Juveniles and Adults)* | | |
| *Direct Effects* | | |
| 5. Survival, growth, and reproduction of CRLF | May affect<br>Not likely to adversely affect<br>(Discountable) | Screening level LOC exceedences for juveniles. Effects may be overestimated by existing toxicity data and are unlikely to occur >20 ft from use site.<br><br>No acute exceedences for adults<br>No chronic exceedences for juveniles or adults. |
| *Indirect Effects and Critical Habitat Effects* | | |
| 6. Reduction or modification of terrestrial prey base | May affect<br>Not likely to adversely affect<br>(Discountable) | Screening level LOC exceedences for terrestrial invertebrates. Effects may be overestimated by existing toxicity data and are unlikely to occur >20 ft from use site.<br><br>No exceedences for mammal or amphibian prey |
| 7. Degradation of riparian and/or upland vegetation | May affect<br>NLAA >1000 ft<br>LAA <1000ft (aerial)<br>LAA <90ft (ground)<br>Modification to critical habitat<br>Adverse <1000ft (aerial)<br>Adverse <90ft (ground) | Exceedences for both monocots and dicots in both wetlands and uplands adjacent to use site for all crops registered.<br><br>Exceedences for both monocots and dicots near use site based on spray drift alone for all crops registered. |

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 adverse modification to critical habitat.

## *2.0 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 EPA's methodologies as described in the Overview Document (U.S. EPA 2004).

## *2.1 Purpose*

The purpose of this endangered species assessment is to evaluate potential direct and indirect effects on individuals of the federallly threatened California red-legged frog *(Rana aurora draytonii*) (CRLF) arising from FIFRA regulatory actions regarding use of metolachlor on soybeans, legumes, corn, cotton, potatoes, safflower, and sorghum. In addition, this assessment evaluates whether use on these crops is expected to result in the destruction or adverse modification of the species' critical habitat. This ecological risk assessment has been prepared consistent with a settlement agreement in the case *Center for Biological Diversity (CBD) vs. EPA et al*. (Case No. 02-1580-JSW(JL)) settlement entered in Federal District Court for the Northern District of California on October 20, 2006.

In this assessment, direct and indirect effects to the CRLF and potential adverse modification to its 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, TREX, TerrPlant, AgDrift, and AgDisp, all of which are described at length in the Overview Document. Additional refinements include a modification of TREX (T-HERPS) to evaluate effects on terrestrial-phase frogs, an analysis of the usage data, and a spatial analysis. 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 metolachlor 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 OPP's Levels of Concern (LOCs). It is acknowledged that the action area for a national-level FIFRA regulatory decision associated with a use of metolachlor may potentially involve numerous areas throughout the United States and its Territories. However, for the purposes of this assessment, attention will be focused on the section of the action area, that intersects with 1) locations where CLRF is known to occur[3], 2) currently occupied core areas for the CLRF[4], and 3) designated critical habitat.

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

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

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 exceedences) upon individual CRLFs or upon the PCEs of the species' designated critical habitat, a "no effect" determination is made for use of metolachlor as it relates to this species and its designated critical habitat. If, however, direct or indirect effects to individual CRLFs are anticipated and/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 metolachlor.

If a determination is made that use of metolachlor within the action area(s) associated with the CRLF "may affect" this species and/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 metolachlor use sites) and further evaluation of the potential impact of metolachlor on the PCEs is also used to determine whether destruction or adverse 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 and/or the PCEs of its designated critical habitat.

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3 As documented in the California Natural Diversity Database (CNDDB)

4 As described in the recovery plan.

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 metolachlor is expected to directly impact living organisms within the action area (defined in Section 2.7), critical habitat analysis for metolachlor 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 destroy or adversely modify critical habitat are those that alter the PCEs and appreciably diminish the value of the habitat. Evaluation of actions related to use of metolachlor 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*

Metolachlor (PC 108801, CAS Registry #s 51218-45-2 and 87392-12-9), is an herbicide currently registered in the U.S. for agricultural uses only, on a limited number of crops. These crops include soybeans, corn, cotton, legume vegetables, potatoes, safflower and sorghum. Metolachlor has two major degradates, metolaclor ethane sulfonic acid (metolachlor ESA) and metolachlor oxanilic acid (metolachlor OA) that have been detected in both surface and ground water. Both of these degradates are considered in the assessment. This assessment does not evaluate *s*-metolachlor (PC 108800), which is a separate registration. The scope of this assessment includes exposure and effects modeling for the active ingredient metolachlor. Several current registrations include the active ingredient atrazine. Based on a comparison of formulation toxicity to mammals (Appendix G), the Agency determined an assessment of metolachor's potential effect on the CRLF when it is co-formulated with other active ingredients (as currently registered) can be based on the toxicity of metolachlor.

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

Although current registrations for metolachlor allow for use nationwide, this ecological risk assessment and effects determination addresses currently registered uses of metolachlor in portions of the action area that are reasonably assumed to be biologically relevant to the CRLF and its designated critical habitat.[5]

## 2.3 Previous Assessments

### 2.3.1 Metolachlor

The Agency has completed other assessments on metolachlor, including an evaluation of the potential effects on 26 ESUs of listed salmonids in the Pacific Northwest (PNW) (U.S. EPA 2006a) and the assessment supporting the Agency's Re-registration Eldigibility Document (RED) document (U.S. EPA 1994)[6]. The metolachlor ecological risk assessment for the RED identified an exceedence of the endangered species risk level of concern (LOC) for fish, based on runoff into a shallow (6 inch) water body from roadside use. Since 1994, EFED has incorporated the use of more advanced exposure models into the risk assessment process.

The 2006 evaluation concerning the salmonids was more comprehensive than the RED, incorporating newer exposure models and the methodologies described in the Overview document (U.S. EPA 2004). The PNW assessment considered direct effects on the growth, survival, and reproduction of the salmonids themselves, and also indirect effects, such potential impacts on prey organisms, aquatic plants, and riparian vegetation. For the salmonids, riparian vegetation was considered to serve the functions of streambank stabilization, reduction of sedimentation, temperature moderation (shading), and allochthonous input. In summary, the salmonid assessment found use of metolachlor as registered:

- Would have no (direct) effect on salmonids (survival, growth or reproduction)
- Was not likely to adversely affect salmonid prey
- Was not likely to adversely affect aquatic plants, and
- Was not likely to adversely affect riparian vegetation.

In 2007, an ecological risk assessment evaluating the risk to the Barton Springs salamander was completed. This assessment determined that metolachlor use

- Would have no (direct) effect on the Barton Springs salamander (survival, growth or reproduction)
- Was not likely to adversely affect salamander prey, and
- Was not likely to adversely affect aquatic plants.

[5] Technical labels also exist, which may include crops not listed on end use labels. Technical products are used to make formulated end use products. Because these technicals cannot be applied directly, use sites on these labels are not considered at part of the Federal action.
[6] Available via the internet at http://www.epa.gov/oppsrrd/REDs/0001.pdf

Although terrestrial plants serve important functions in riparian systems, a spatial analysis of metolachlor use sites near Barton Springs showed the closest use sites to the springs were 5-10 miles away. It appeared impacts on the terrestrial vegetation adjacent to the springs were unlikely, and they were not specifically evaluated. The no effect and not likely to adversely affect determinations were predicated, in large part, on limited use of metolachlor in areas that might result in the Barton Springs salamander being exposed to the pesticide.

### *2.3.2 California Red-legged Frog*

The Agency is currently developing a number of risk assessments for the CLRF, each addressing different pesticide active ingredients. A total of 66 chemicals will be assessed. Metolachlor is among the first group of ten chemicals to be completed. For information regarding the other chemicals in this group[7] please see the relevant document.

## *2.4 Stressor Source and Distribution*

### *2.4.1 Environmental Fate Properties*

Acceptable environmental fate data indicate that parent metolachlor appears to be moderately persistent to persistent in soil. It ranges from mobile to highly mobile in different soils and has been detected extensively in surface water and ground water. Metolachlor degradation appears to be dependent on both microbially mediated (aerobic soil metabolism $t_{1/2}$ = 66 days, 37.8 days, 37.8 days, 14.9 days, 13.9 days, and 50.3 days, anaerobic soil metabolism $t_{1/2}$ =81 days) and abiotic processes (photodegradation in water $t_{1/2}$ = 70 days under natural sunlight and photodegradation on soil $t_{1/2}$ = 8 days under natural sunlight).

The major degradates of metolachlor were initially identified as CGA-51202 (metolachlor oxanilic acid (OA)), CGA-50720, CGA-41638, CGA-37735, and CGA-13656. Subsequent studies also identified CGA-354743 (metolachlor ethane sulfonic acid (ESA)). Of these major degradates, two (metolachlor ESA and metolachlor OA) were identified as being of toxicological concern. Both have been identified in both ground water and surface water. Depending on the soil (*i.e.,* organic matter content), metolachlor has the potential to range from a moderately mobile to a highly mobile compound with $K_d$ values ranging from 0.11 to 44.8, and $K_{oc}$ values ranging from 21.6 to 367.

Field dissipation studies indicate that metolachlor is persistent in surface soil, with half lives ranging from 7 to 292 days in the upper six inch soil layer. Metolachlor was reportedly detected as deep as the 36 to 48 inch soil layer in some studies. Metolachlor OA (CGA-51202), was detected (0.11 ppm) as deep as the 30-36 inch soil depth (MRID 41335701); CGA-40172 was detected as deep as the 36-48 inch (MRID 41309802);

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[7] Other chemicals assessed in the first group include methamidaphos, methomyl, azinphos-methyl, acephate, imazpyr, aldicarb, metam sodium, diazinon and chloropicrin

CGA-40919 was detected in the 36-48 inch depth (0.21 ppm in MRID 41309802); and CGA-50720 was not detected (LOD = 0.07 ppm) in any soil segment at any interval.

### *2.4.2 Environmental Transport Mechanisms*

Potential transport mechanisms include pesticide surface water runoff, spray drift, and secondary drift of volatilized or soil-bound residues leading to deposition onto nearby or more distant ecosystems. Surface water runoff and spray drift are expected to be the major routes of exposure for metolachlor.

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 the metolachlor'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. Therefore, physicochemical properties of the metolachlor that describe its potential to enter the air from water or soil (*e.g.*, Henry's Law constant and vapor pressure), pesticide use data, modeled estimated concentrations in water and air, and available air monitoring data from the Central Valley and the Sierra Nevadas are considered in evaluating the potential for atmospheric transport of metolachlor to locations where it could impact the CRLF.

In general, deposition of drifting or volatilized pesticides is expected to be greatest close to the site of application. Computer models of spray drift (AgDRIFT and AGDISP) are used to determine potential exposures to aquatic and terrestrial organisms. Metolachlor is most toxic to monocotyledon terrestrial plants, thus the distance of potential impact away from the use sites (action area) is determined by the distance required to fall below the LOC for these organisms.

### *2.4.3 Mechanism of Action*

Metolachlor is a biosysnthesis inhibitor, absorbed by the roots and shoots of the plant (www.syngentacropprotection-us.com/prod/herbicide/dualiimagnum). It translocates in the plant. Germinating monocots primarily absorb metolachlor through the shoot just above the seed, and germinating dicots absorb at the shoot and the roots (Zimdahl 1993). Metolachlor may be active in the soil for several months following application.

A single specific biochemical target of metolachlor and other chloroacetamide herbicides has not been defined and it appears the chemicals may act via multiple pathways. Alkylation appears to be important in phytotoxicity (Jablonkai 2003) and lipophilicity has been correlated with algal reproduction effects (Junghans *et al.* 2003). Covalent inhibition of coenzyme A elongation (Schmalfuss *et al.* 2000) and covalent inhibition of very-long-chain fatty acid synthesis via chalcone synthase have been proposed as mechanisms of action in terrestrial plants (Eckermann *et al.* 2003). Inhibition of protein biosynthesis has also been proposed as a mechanism of action in plants (Pillai *et al.* 1979). Several proposed mechanisms of action in plants involve irreversible, covalent binding to cysteine residues. Consistent with cysteine reactivity, glutathione S-transerfase has been shown to be important in detoxifying chloroacetanilide herbicides in tolerant plants (Rossini *et al.* 1998).

### *2.4.3 Use Characterization*

Analysis of labeled use information is the critical first step in evaluating the federal action. The current label for metolachlor represents the FIFRA regulatory action; therefore, use sites 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. Metolachlor registrations are currently limited to agricultural crops. Specific use sites include corn, cotton, pod crops, potatoes, safflower, sorghum, and soybeans. Information on current registrations is provided in Table 2.

**Table 2 Current Metolachlor Registrations**

| Product Name | EPA Registration Number | Crops |
|---|---|---|
| Drexel Trizmet II[a] | 19713-547 | Corn |
| Drexel Me-too-lachlor | 19713-548 | Corn, cotton, pod crops, potatoes, safflower, sorghum, soybeans |
| Drexel Me-too-lachlor II | 19713-549 | Corn |
| Drexel Me-too-lachlor V Herbicide | 19713-554 | Corn |
| Drexel Me-too-lachlor IV Herbicide | 19713-555 | Corn |
| Drexel Me-too-lachlor III Herbicide | 19713-556 | Corn |
| Metolachlor 8E | 19713-591 | Corn, pod crops, safflower, sorghum, soybeans |
| Metolachlor AT[a] | 19713-593 | Corn, sorghum, soybeans |
| Metolachlor 8E Plus | 19713-595 | Corn |
| Parallel PCS | 66222-86 | Corn, pod crops, safflower |
| Metolachlor 7.8 | 60063-24 | Corn, pod crops, potatoes, safflower, soybeans |
| Triangle Herbicide[a] | 66222-131 | Corn, sorghum, soybeans |
| Parallel Plus[a] | 66222-132 | Corn, sorghum, soybeans |

Corn includes corn, corn (unspecified), field corn, popcorn, and sweet corn
Pod crops includes black-eyed peas, cowpeas, dry beans, garbanzos, lentils, lupine, succulent lima beans, succulent snap beans, and southern peas
[a] Also contains active ingredient atrazine

The Office of Pesticides Programs' Biological and Economic Analysis Division (BEAD) provides an analysis of both national- and county-level usage information using state-level usage data obtained from USDA-NASS[8], Doane[9], and the California's Department of Pesticide Regulation Pesticide Use Reporting (CDPR PUR) database[10]. CDPR PUR is considered a more comprehensive source of usage data than USDA-NASS or EPA proprietary databases, and thus the usage data reported by county in this assessment were generated using CDPR PUR data. From the CDPR PUR data, BEAD generated summaries of average and total usage by year, county, and crop for the years 2002-2005 (the most recent and best available data). Total usage is shown in Figure 1, Figure 2, and Figure 3.

Some uses reported in the CDPR PUR database may be different than those considered in the assessment. The uses considered in this risk assessment represent currently registered uses according to a review of all current labels by OPP/BEAD and OPP/SRRD. No other uses are relevant to this assessment. Any reported uses in the CA DPR database that do not reflect current labled uses may represent either historic uses that have been canceled, mis-reported uses, or cases of mis-use. Historical uses, mis-reported uses, and mis-use are not considered part of the federal action and, therefore, are not considered in this assessment.

Based on data reported in the CDPR PUR database, metolachlor use appears to be declining. In 2002, reports show a total of 3,914 lbs was applied. This has declined steadily, with only 1,068 lbs reported applied in 2005 (Figure 1). This may reflect a shift in use to *s*-metolachlor, although analysis of the existing dataset cannot confirm this supposition. Twenty-six counties in California reported some usages of metolachlor. Of the total metolachlor applied, 90% was used in only 11 counties (Figure 2). Highest usage (total usage >1,000 lbs metolachlor in 2002-2005) was reported in Contra Costa, San Joaquin, and Riverside counties. Three crop groups (beans, including soybeans and peas; corn, and cotton) accounted for 61% of the total usage (Figure 3). Usage was also reported (14%) on ornamentals including nursery and landscape uses. Presumably this reflects use of previously existing stocks, as there are no longer active registrations with these use sites. No use on sorghum was reported in 2002-2005.

[8] United States Department of Agriculture (USDA), National Agricultural Statistics Service (NASS) Chemical Use Reports provide summary pesticide usage statistics for select agricultural use sites by chemical, crop and state. See http://www.usda.gov/nass/pubs/estindx1.htm#agchem.
[9] (www.doane.com; the full dataset is not provided due to its proprietary nature)
[10] The California Department of Pesticide Regulation's Pesticide Use Reporting database provides a census of pesticide applications in the state. See http://www.cdpr.ca.gov/docs/pur/purmain.htm.

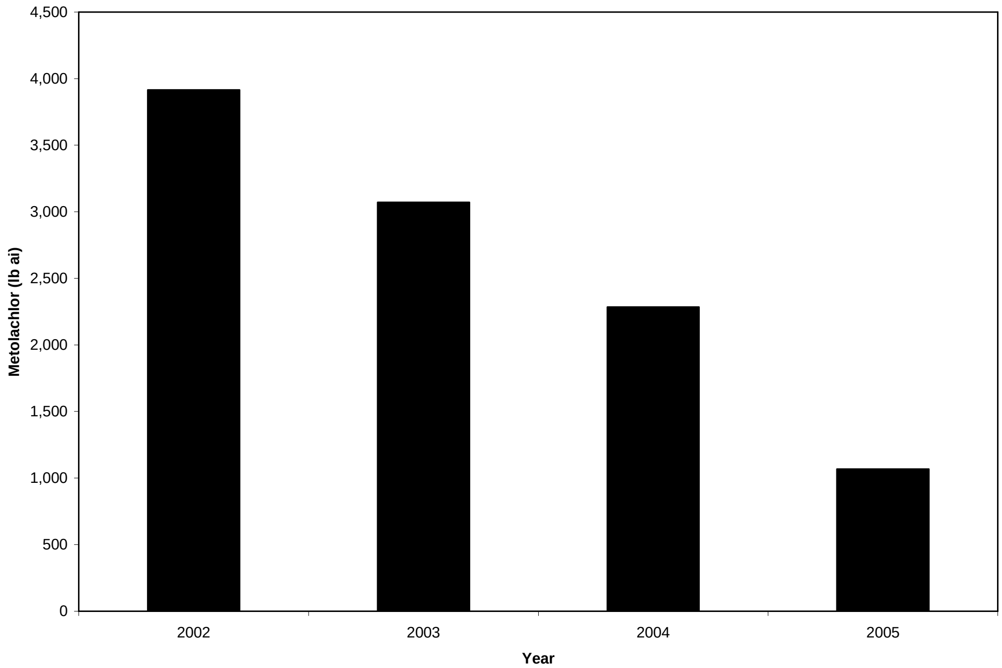



**Figure 1  Metolachlor Usage in California by Year (Total Applied, 2002-2005)**

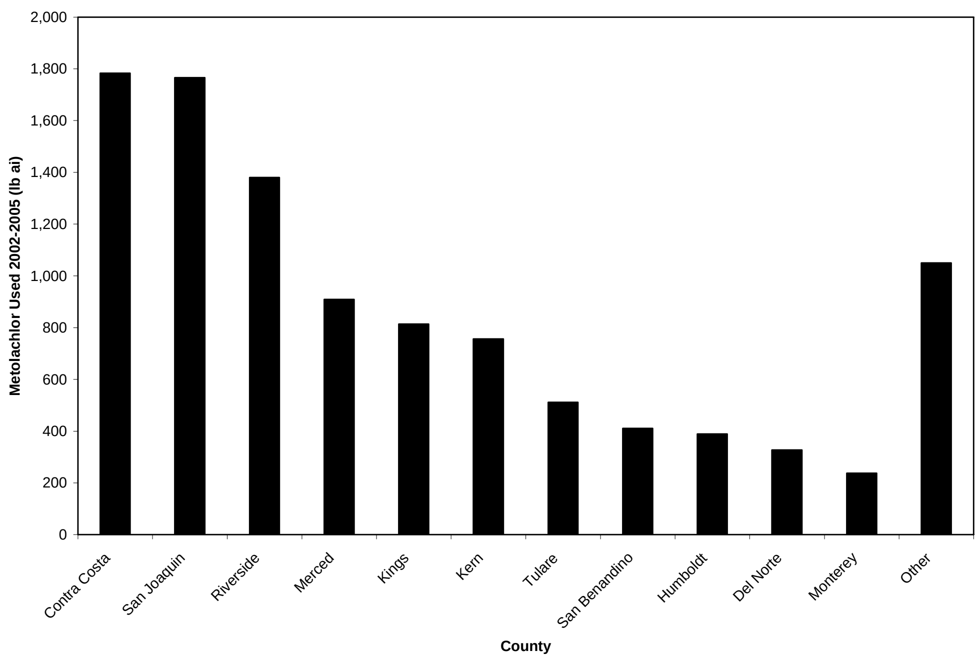



**Figure 2  Metolachlor Usage in California by County (Total Applied, 2002-2005)**

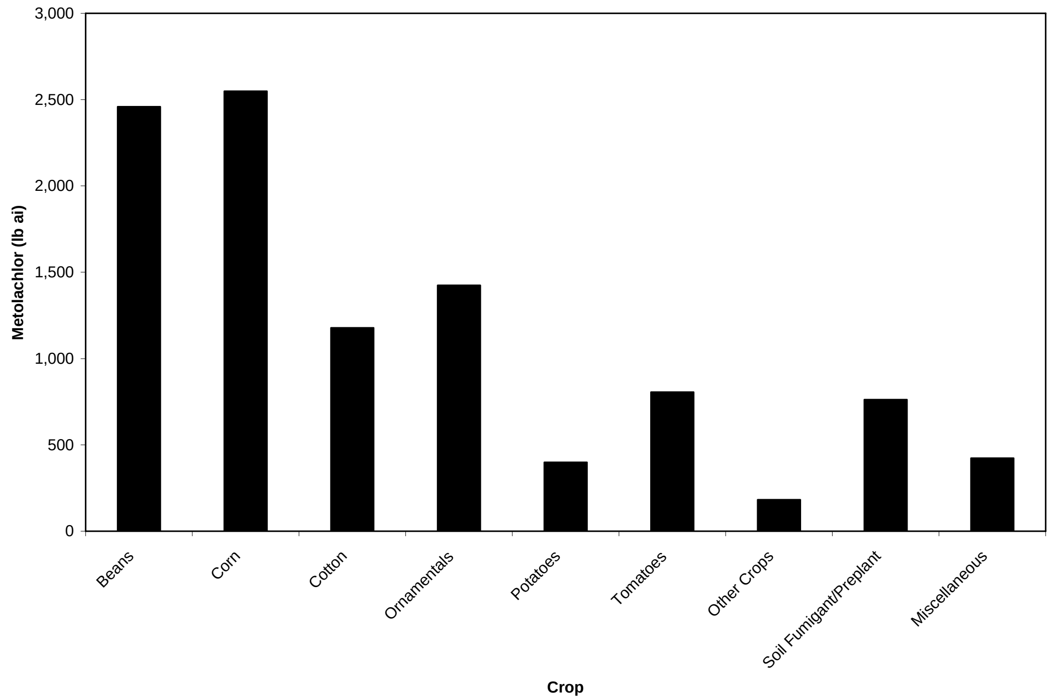



**Figure 3 Metolachlor Usage in California by Crop (Total Applied, 2002-2005)**

## *2.5 Assessed Species*

The California red-legged frog 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). Final critical habitat for the CRLF was designated by USFWS on April 13, 2006 (USFWS 2006; 71 FR 19244-19346). A brief discussion of distribution, reproduction, diet, and habitat requirements follows, with more detailed information provided in Attachment 1.

### *2.5.1 Distribution*

The frog is endemic to California and Baja California (Mexico) and historically inhabited 46 counties in California, including the Central Valley and both the coastal and interior mountain ranges (USFWS 1996). Its range has been reduced by approximately 70%, and it currently inhabits 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 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). 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. Three of these categories were designated by the USFWS in the recovery plan (recovery units, core areas, and designated critical habitat). The fourth category is known occurrences as reported in the California Natural Diversity Database (CNDDB) (Figure 4). Recovery units are large areas defined at the watershed level that have similar conservation needs and management strategies. The recovery unit is primarily an administrative designation, and land area within the recovery unit boundary is not exclusively CRLF habitat. Core areas are smaller areas within the recovery units that comprise portions of the species' historic and current range and have been determined by USFWS to be important in the preservation of the species. Designated critical habitat is generally contained within the core areas, although a number of critical habitat units are outside the boundaries of core areas, but within the boundaries of the recovery units. Additional information on CRLF occurrences from the CNDDB is used to cover the current range of the species not included in core areas and/or designated critical habitat, but within the recovery units. For purposes of this assessment, designated critical habitat, currently occupied (post-1985) core areas, and additional known occurrences of the CRLF from the CNDDB are considered the range of the species.

#### *2.5.1.1 Recovery Units*

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

#### *2.5.1.2 Core Areas*

USFWS has designated 35 core areas in which to focus recovery efforts. The core areas, which are distributed throughout portions of the historic and current range of the species, are intended to provide for long-term viability of existing populations and reestablishment of populations within historic range. These areas were selected because they: 1) contain existing viable populations; or 2) they contribute to the connectivity of other habitat areas (USFWS 2002). Core area protection and enhancement are vital for maintenance and expansion of the CRLF's distribution and population throughout its range.

### *2.5.1.3 Designated Critical Habitat*

Critical habitat was designated for the CRLF on April 13, 2006 (USFWS 2006; 71 FR 19244-19346). Critical habitat was selected for the species based on areas: 1) that are occupied by CRLFs; 2) where source populations of CRLFs occur; 3) that provide connectivity between source populations; and 4) that are ecologically significant. Designation of critical habitat is based on habitat areas that provide essential life cycle needs of the species or areas that contain primary constituent elements (PCEs) (as defined in 50 CFR 414.12(b)) The designated critical habitat areas for the CRLF are considered to have the following PCEs that justify critical habitat designation (USFWS 2006):

- Aquatic breeding habitat
- Non-breeding aquatic habitat
- Upland habitat
- Dispersal habitat

Critical habitat does not include certain areas where existing management is sufficient for CRLF protection. For the CRLF, all designated critical habitat units contain all four PCEs and were occupied by the CRLF at the time of listing.

USFWS has established adverse modification standards for designated critical habitat (USFWS 2006). Activities that may destroy or adversely modify critical habitat are those that alter the PCEs and appreciably diminish the value of the habitat. For the CRLF specifically, these include, but are not limited to, the following:

- Alteration of water chemistry or temperature
- Increased sedimentation
- Alteration of channel or pond morphology
- Elimination of upland foraging areas
- Introduction of non-native species
- Degradation of prey base

The critical habitat designation 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.

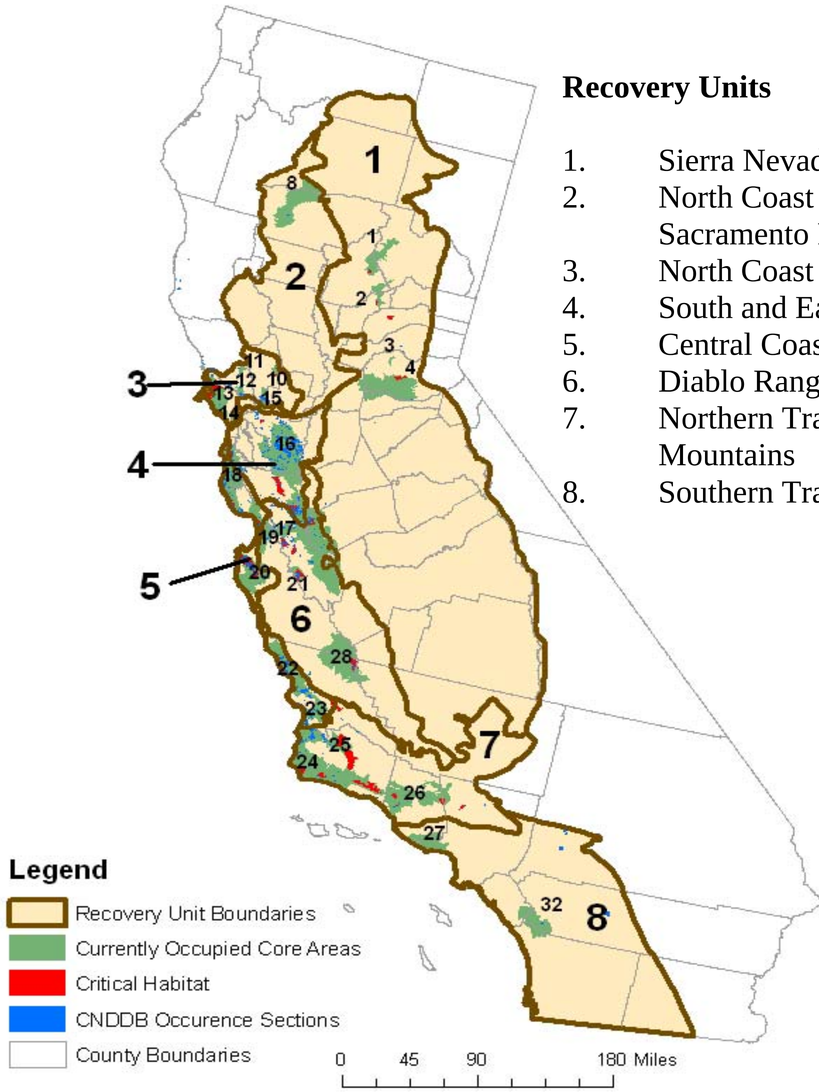
### *2.5.1.4 Other Known Occurrences from the CNDBB*

The CNDDB[11] provides location and natural history information on species found in California. It is the best available information for historical and current species location sightings.

[11] See: http://www.dfg.ca.gov/bdb/html/cnddb_info.html for additional information on the CNDDB.

## Recovery Units

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




## Core Areas

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

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

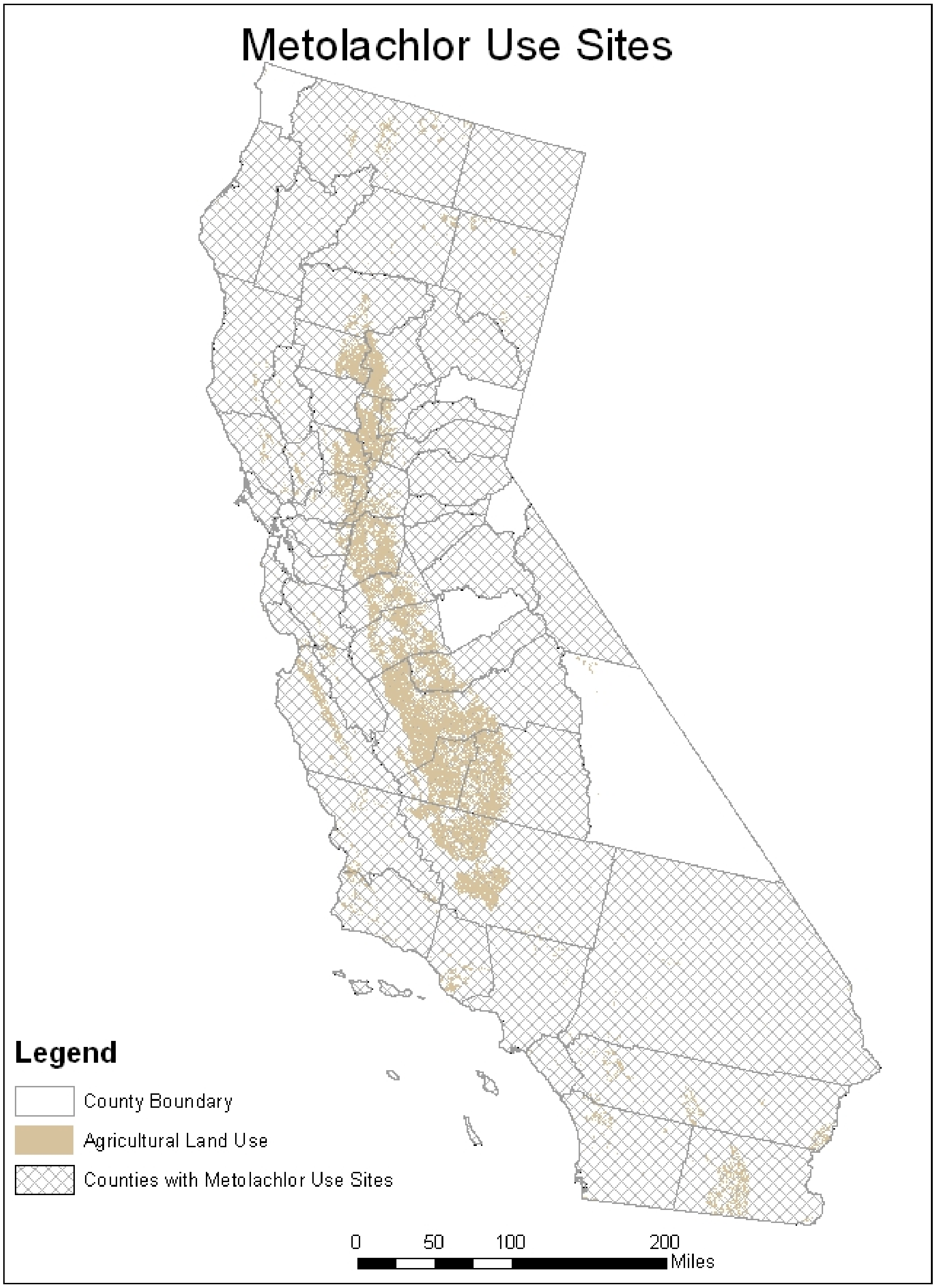
**Figure 4 California Red-legged Frog Distribution**

## *2.6 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). In the Overview Document, EPA defined exceedences of the pre-established OPP levels of concern (LOCs) as effects (USEPA 2004). The initial area of concern is delineated by the registered use sites, or some reasonable surrogate, such as a land use type. The extent of the action area is determined by the taxa for which LOCs are exceeded farthest away from the use site. This offset is added to the edge of the use sites and the total area is considered the action area. For metolachlor, the most sensitive terrestrial species are monocotyledon plants, and the offset is determined by the furthest distance from the application site that metolachlor spray drift exceeds the LOC for this taxa. For aquatic organisms, the offset is determined by the distance downstream required for the most sensitive endpoint to drop below the LOC. The CLRF-metolachlor action area is the summation of the use sites, and the offsets from the terrestrial and aquatic endpoints.

It is recognized by the Agency that the overall action area for the national registration of metolachlor includes any locations where registered uses might result in ecological effects. However, the scope of this assessment limits consideration to the areas in which applicationof the pesticide may have direct or indirect effects on California red-legged frog or its designated critical habitat.

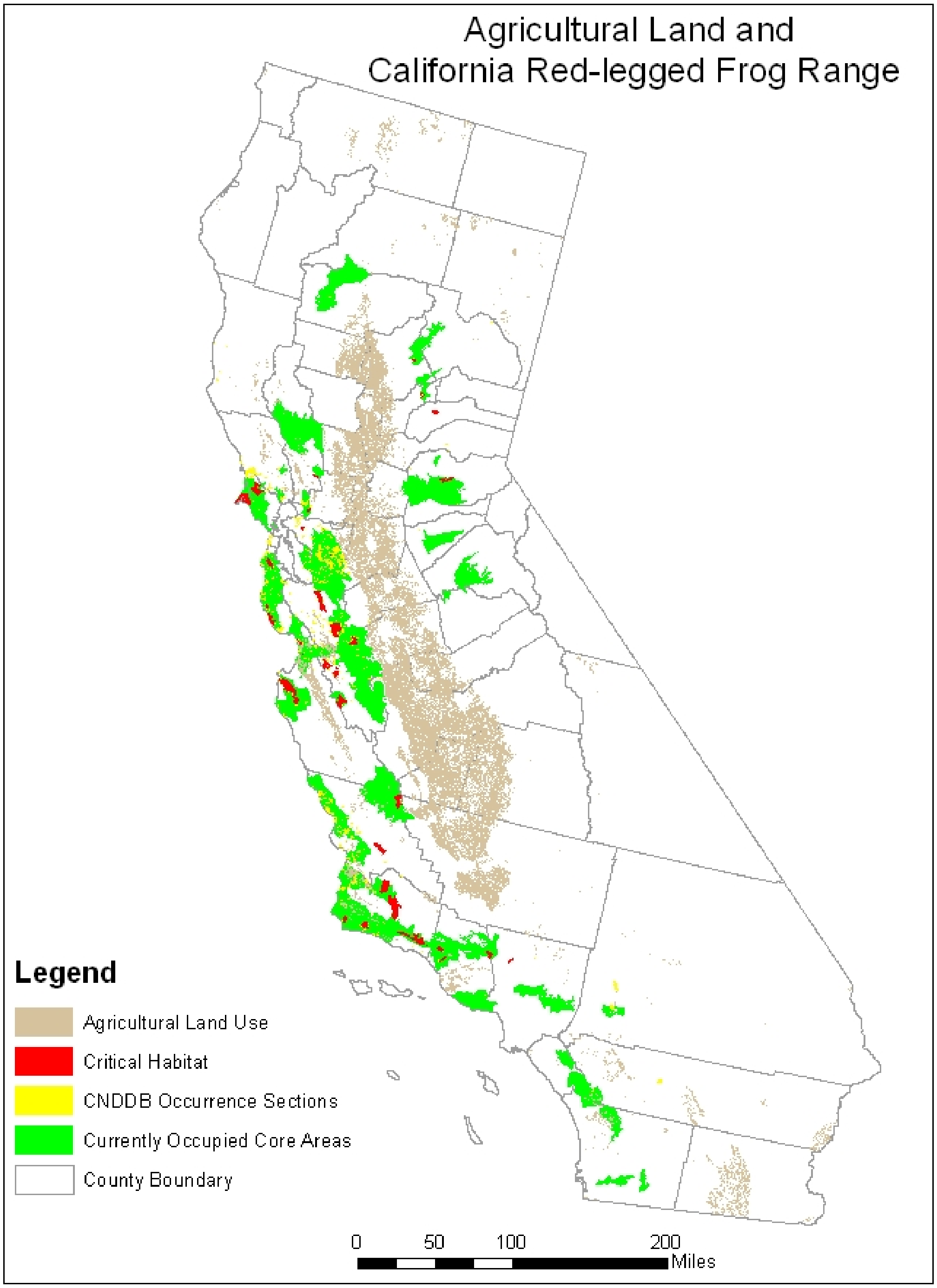
All currently registered uses of metolachlor are for agricultural crops. Figure 5 shows counties in California where NASS data shows the currently registered crops are grown. It also shows what land has been classified agricultural by the NLCD. The intersection of these two data sets comprises the initial area of concern. Essentially, the area of concern is all agricultural land within California, with the exception of two counties where the registered crops are not reported. In order to give the benefit to the species, EFED opted to define the initial area of concern as all NCLD classified agricultural land in the state of California. Given the range of the CLRF as defined for the assessments, even considering spray drift, agricultural uses in Oregon and Nevada are not expected to impact the frog. Figure 6 shows the overlap of agricultural land use and the CLRF range. Based on toxicity data metolachlor could affect terrestrial plants up to 450 feet away from the use site for ground applications, and up to 2,060 feet away from the use site for aerial applications. The extent of NCLD-classified agricultural land plus the distance associated with potential effects from aerial applications (2,060 ft) defines the extent of potential terrestrial effects. Because agricultural areas are non-contiguous, the action area, which is based on the agricultural area plus an offset, is also non-contiguous.




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

Map created by U.S. Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. May 2, 2007 (PDB).
Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983)

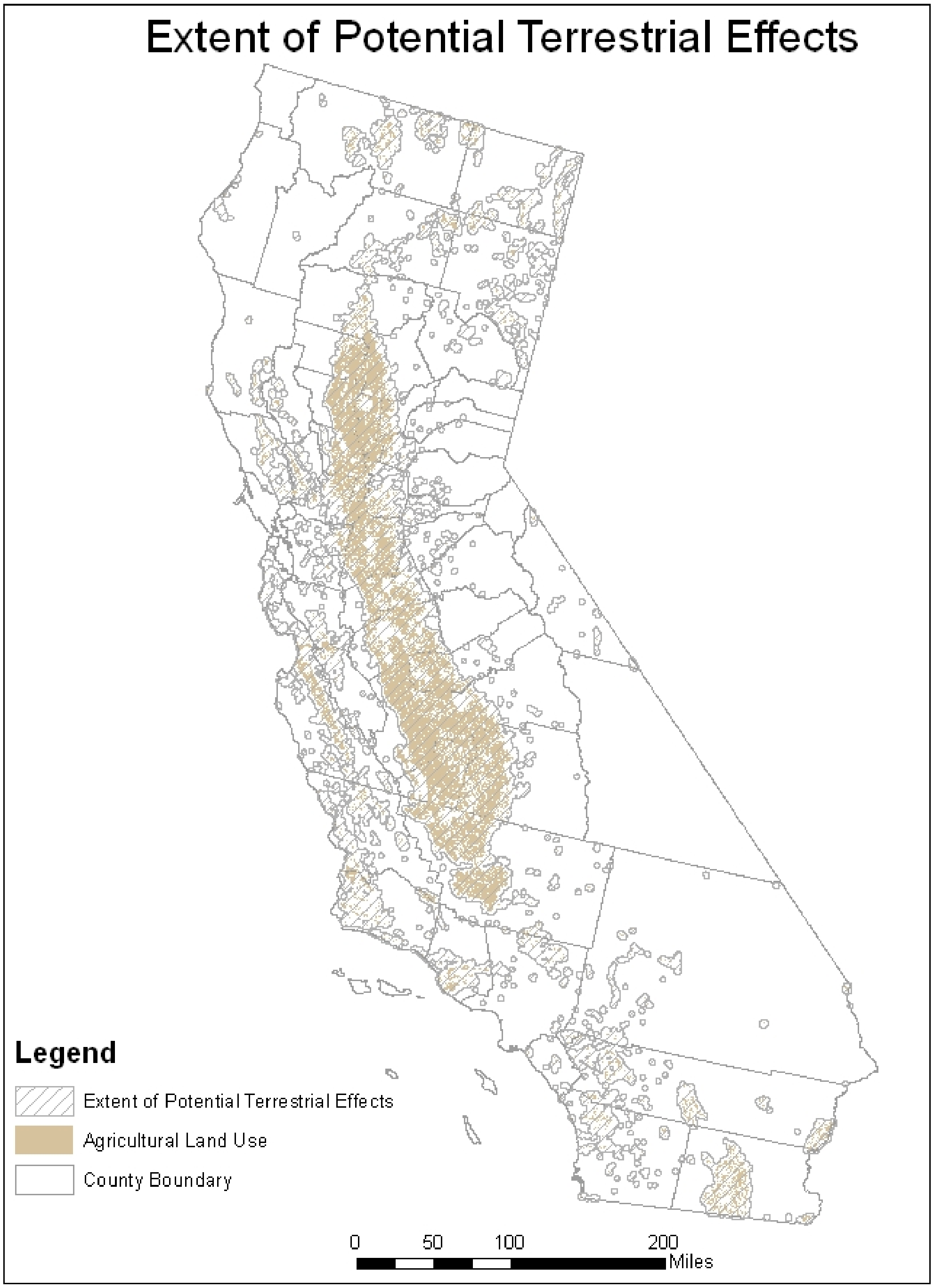
**Figure 5 Metolachalor Use Sites and Agricultural Land in California**




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

Map created by U.S. Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. May 4, 2007 (PDB). Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983)

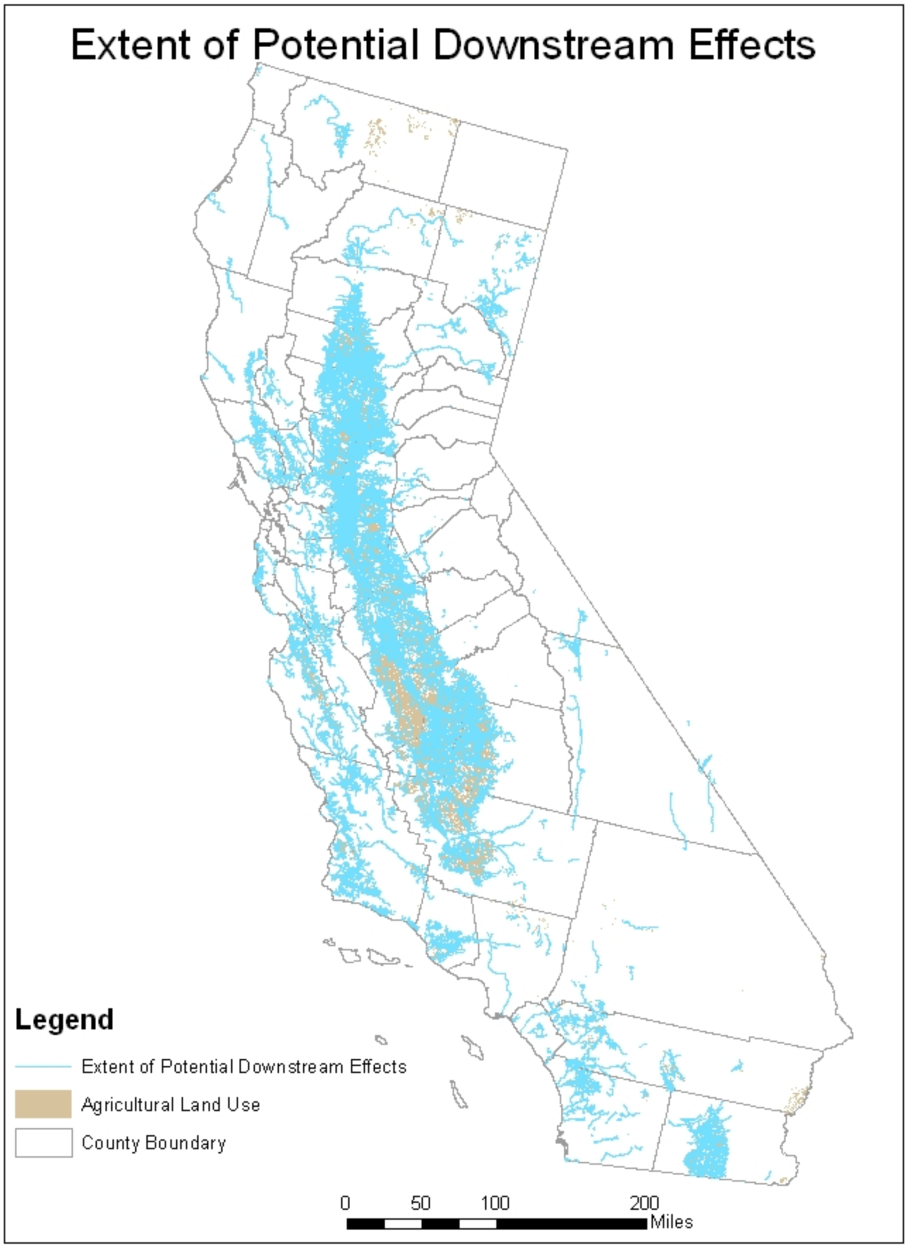
**Figure 6  Comparison of Agricultural Land with CLRF Range**




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

Map created by U.S. Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. June 7, 2007 (PDB). Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983)

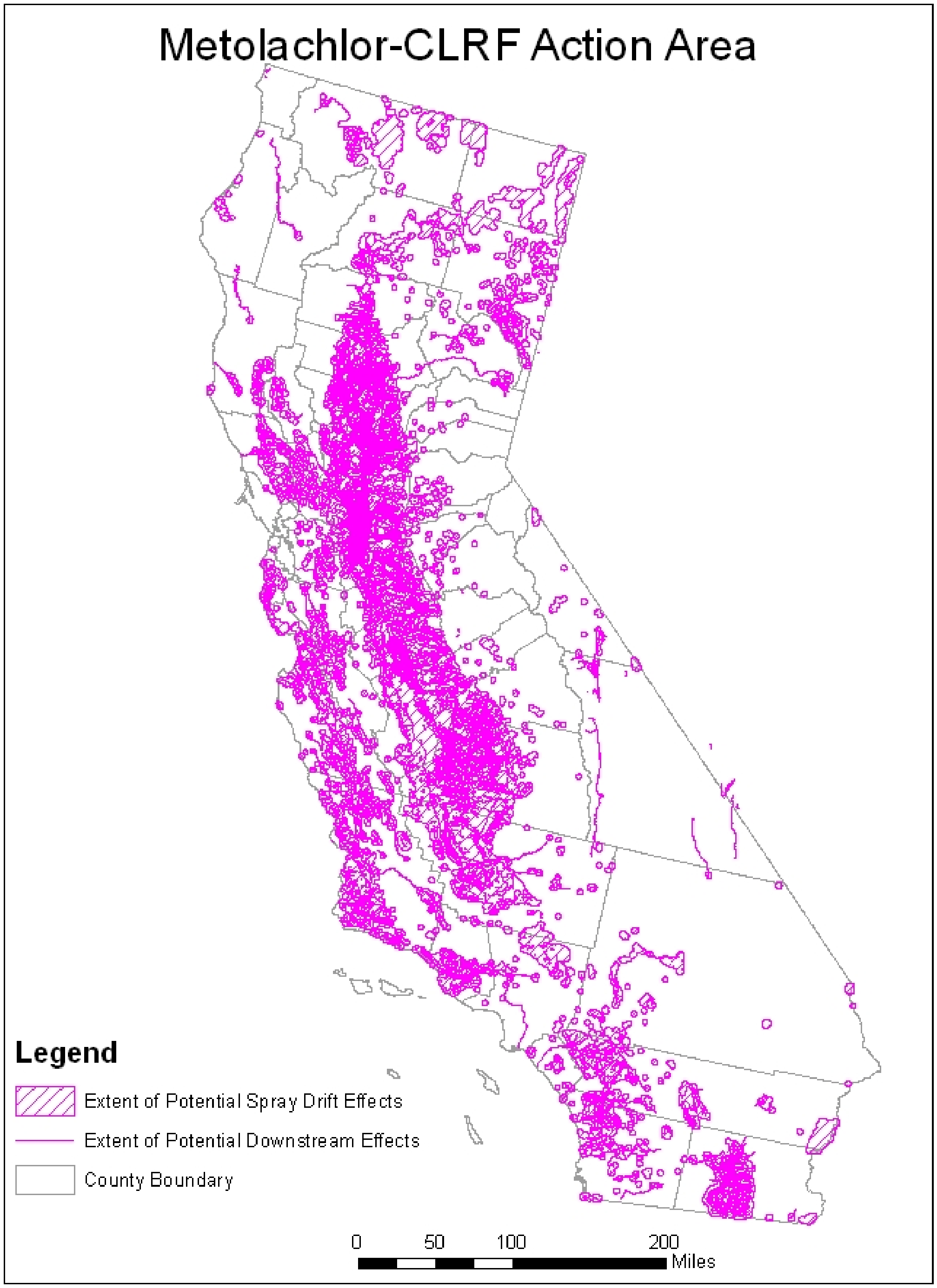
**Figure 7 Extent of Potential Terrestrial Effects**




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

Map created by U.S. Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. June7, 2007 (PDB).
Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983)

**Figure 8 Extent of Potential Downstream Effects**




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

Map created by U.S. Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. April 11, 2007.
Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983)

**Figure 9 Metolachlor-CLRF Action Area**

## 2.7 Assessment Endpoints and Measures of Ecological Effect

Assessment endpoints are defined as "explicit expressions of the actual environmental value that is to be protected."[12] 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 metolachlor (*e.g.*, runoff, spray drift, etc.), and the routes by which ecological receptors are exposed to metolachlor *(e.g.,* direct contact, *etc.).*

### 2.8.1. Assessment Endpoints for the CRLF

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

A complete discussion of all the toxicity data available for this risk assessment, including resulting measures of ecological effect selected for each taxonomic group of concern, is included in Section 4 of this document. A summary of the assessment endpoints and measures of ecological effect selected to characterize potential assessed direct and indirect CRLF risks associated with exposure to metolachlor is provided in Table 3.

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

**Table 3 Assessment Endpoints and Measures of Ecological Effects**

| Assessment Endpoint | Measures of Ecological Effects[13] |
|---|---|
| *Aquatic Phase (Eggs, larvae, tadpoles, juveniles, and adults)*[a] | |
| *Direct Effects* | |
| 1. Survival, growth, and reproduction of CRLF | 1a. Bluegill sunfish $LC_{50}$<br>1b. Fathead minnow chronic NOAEC |
| *Indirect Effects and Critical Habitat Effects* | |
| 2. Survival, growth, and reproduction of CRLF individuals via indirect effects on aquatic prey food supply (*i.e.,* fish, freshwater invertebrates, non-vascular plants) | 2a. Bluegill sunfish $LC_{50}$<br>2b. Fathead minnow chronic NOAEC<br>2c. Water flea acute $EC_{50}$<br>2d. Water flea chronic NOAEC.<br>2e. Non-vascular plant (freshwater algae) acute $EC_{50}$ |
| 3. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat, cover, food supply, and/or primary productivity (*i.e.,* aquatic plant community) | 3a. Vascular plant acute $EC_{50}$ (duckweed) [c]<br>3b. Non-vascular plant acute $EC_{50}$ (freshwater algae) [c] |
| 4. Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation | 4a. Monocot $EC_{25}$ (seedling emergence) [c]<br>4b. Dicot $EC_{25}$ (seedling emergence) [c]<br>4c. Tree and shrub LOAEC [c] |
| *Terrestrial Phase (Juveniles and adults)* | |
| *Direct Effects* | |
| 5. Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles | 5a. Bobwhite quail acute $LD_{50}$<br>5b. Bobwhite quail chronic NOAEC |
| *Indirect Effects and Critical Habitat Effects* | |
| 6. Survival, growth, and reproduction of CRLF individuals via effects on terrestrial prey (*i.e.,*terrestrial invertebrates, small mammals , and frogs) | 6a. Honey bee oral $LD_{50}$<br>6b. Rat acute $LD_{50}$<br>6b. Rat chronic NOAEC<br>6b. Bobwhite quail acute $LD_{50}$<br>6b. Bobwhite quail chronic NOAEC |
| 7. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.,* riparian and upland vegetation) | 7a. Monocot $EC_{25}$ (seedling emergence) [c]<br>7b. Dicot $EC_{25}$ (seedling emergence) |

[a] Adult frogs are no longer in the "aquatic phase" of the amphibian life cycle; however, submerged adult frogs are considered "aquatic" for the purposes of this assessment because exposure pathways in the water are considerably different that exposure pathways on land.
[b] Birds are used as surrogates for terrestrial phase amphibians.
[c] .

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

Measures of effect and assessment endpoints defined for indirect effects also apply to critical habitat. Assessment endpoints used for the analysis of designated critical habitat are based on the adverse modification standard established by USFWS (2006).

Adverse 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.

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.

## *2.8 Conceptual Model*

### *2.8.1 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 metolachlor to the environment. The following risk hypotheses are presumed for this endangered species assessment:

- Labeled uses of metolachlor within the action area may directly affect the CRLF by causing mortality or by adversely affecting growth or fecundity;
- Labeled uses of metolachlor within the action area may indirectly affect the CRLF by reducing or changing the composition of food supply;
- Labeled uses of metolachlor within the action area may indirectly affect the CRLF and/or adversely modify designated critical habitat by reducing or changing the composition of the aquatic plant community in the ponds and streams comprising the current range of the species and designated critical habitat, thus affecting primary productivity and/or cover;

- Labeled uses of metolachlor within the action area may indirectly affect the CRLF and/or adversely modify designated critical habitat by reducing or changing the composition of the terrestrial plant community (*i.e.*, riparian habitat) required to maintain acceptable water quality and habitat in the ponds and streams comprising the species' current range and designated critical habitat;
- Labeled uses of metolachlor within the action area may adversely modify the designated critical habitat of the CRLF by reducing or changing breeding and non-breeding aquatic habitat (via modification of water quality parameters, habitat morphology, and/or sedimentation);
- Labeled uses of metolachlor within the action area may adversely modify the designated critical habitat of the CRLF by reducing the food supply required for normal growth and viability of juvenile and adult CRLFs;
- Labeled uses of metolachlor within the action area may adversely modify the designated critical habitat of the CRLF by reducing or changing upland habitat within 200 ft of the edge of the riparian vegetation necessary for shelter, foraging, and predator avoidance.
- Labeled uses of metolachlor within the action area may adversely modify the designated critical habitat of the CRLF by reducing or changing dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal.
- Labeled uses of metolachlor within the action area may adversely modify the designated critical habitat of the CRLF by altering chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs.

### *2.8.2 Diagram*

The conceptual model is a graphic representation of the structure of the risk assessment. It specifies the stressor metolachlor release mechanisms, biological receptor types, and effects endpoints of potential concern. The conceptual models for aquatic and terrestrial phases of the CRLF are shown in Figure 10 and Figure 11, and the conceptual models for the aquatic and terrestrial PCE components of critical habitat are shown in Figure 11 and Figure 12. Exposure routes shown in dashed lines are not expected to be significant.

## *2.9 Analysis Plan*

The exposure and effects analysis is accordance with standard methods described in the Overview document (U.S. EPA 2004). Refinements specific to this assessment include the use of an amphibian/reptile specific terrestrial exposure model (T-HERPS), evaluation of potential effects on terrestrial invertebrates using honey bees as the surrogate, the use of AgDrift to estimate clearance distances for both plants and animals, and the use of partitioning-based estimates to consider atmospheric inputs into water bodies. All refinements have been approved within EFED and are described in the appropriate section.

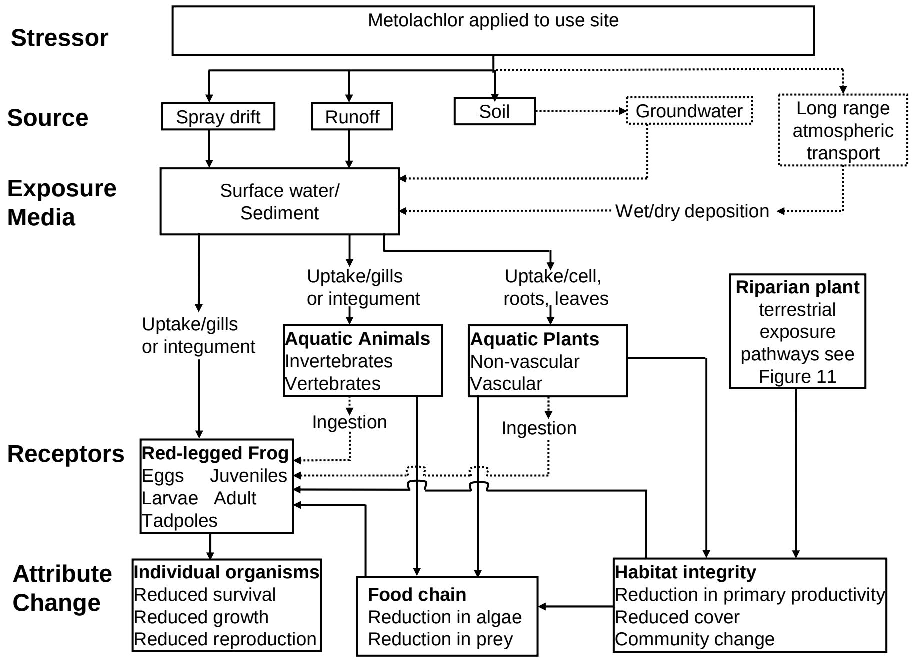



**Figure 10 Conceptual Model for Aquatic Phase of the Red-Legged Frog**

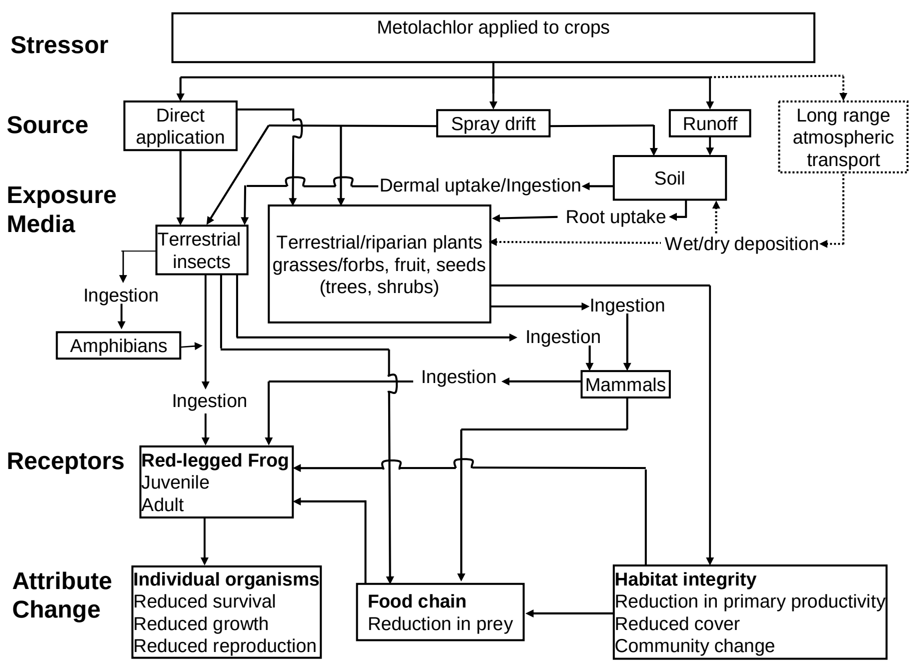



**Figure 11 Conceptual Model for Pesticide Effects on Terrestrial Phase of the Red-legged Frog**

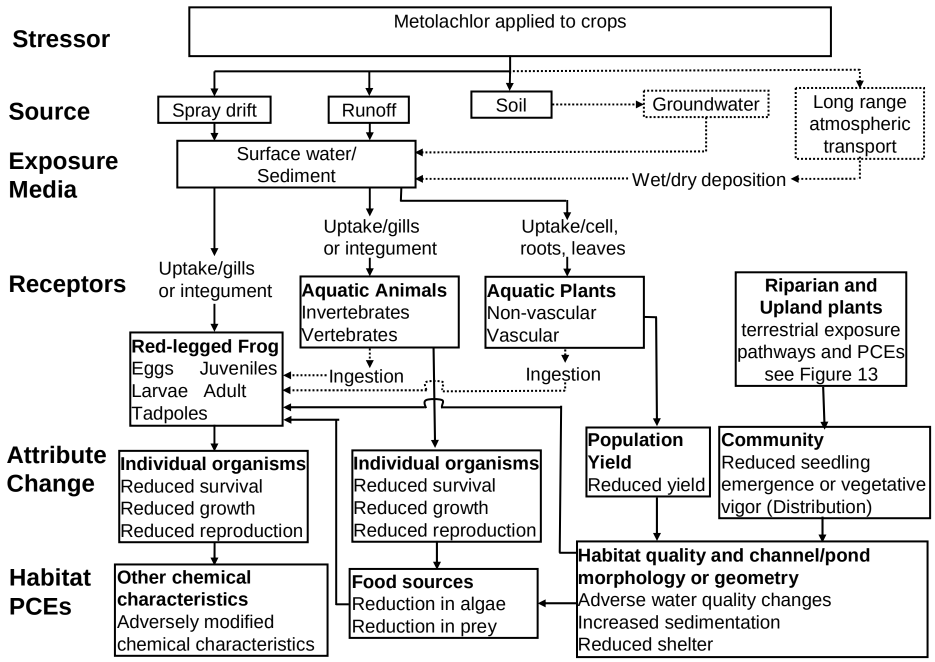



**Figure 12 Conceptual Model for Pesticide Effects on Aquatic component of Red-legged Frog Critical Habitat**

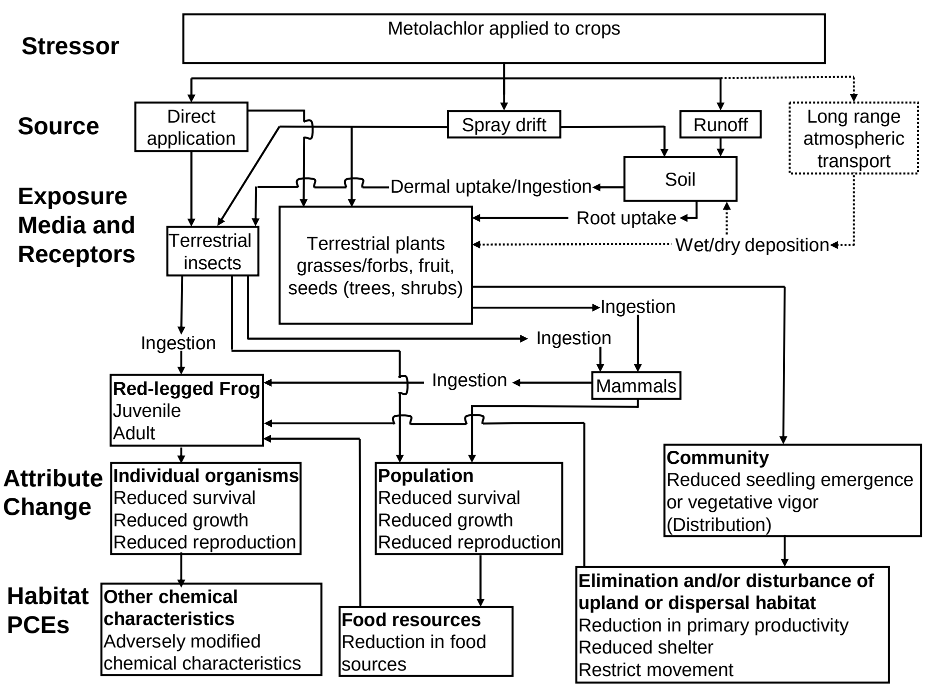



**Figure 13 Conceptual Model for Pesticide Effects on Terrestrial Component of Red-legged Frog Critical Habitat**

## 3.0 Exposure Assessment

## 3.1 Label Application Rates and Intervals

Application rates, timing, and techniques were compiled from actively registered labels. Rates used in modeling are the maximum allowed rate for that specific crop or crop group. Lower rates may exist, and/or growers may choose to apply lower concentrations than permitted by the label. Metolachlor labels permit a single application, thus intervals are not included in Table 4. It is often applied prior to planting, and in most cases is incorporated into the soil. Incorporation depths affect the concentration of pesticide in runoff, thus some crops were modeled with two separate incorporation depths.

**Table 4 Label Application Rates**

| Crop | Label | EPA Reg. No. | Application Timing (Depth of Incorporation) | Application Technique | Application Rate (lbs ai/A) |
|---|---|---|---|---|---|
| Corn | Me-Too-lachlor II | 19713-549 | Preplant Incorporated (2 inches) | Aerial | 1.63 |
| | | | | Ground | 1.63 |
| Cotton | Me-Too-lachlor | 19713-548 | Post Emergent (2 inches) | Aerial | 1.33 |
| | | | | Ground | 1.33 |
| | Me-Too-lachlor | 19713-548 | Pre Emergent (Surface Applied) | Aerial | 1.00 |
| | | | | Ground | 1.00 |
| Pod Crops (Legumes) | Metolachor 7.8 | 60063-24 | Preplant Incorporated (4-6 inches) | Aerial | 1.67 |
| | | | | Ground | 1.67 |
| Potatoes | Me-Too-lachlor | 19713-548 | Pre-emergent Incorporated (3 inches) | Aerial | 2.00 |
| | | | | Ground | 2.00 |
| Safflower | Me-Too-lachlor | 19713-548 | Preplant Incorporated (4-6 inches) | Aerial | 1.67 |
| | | | | Ground | 1.67 |
| Sorghum | Me-Too-lachlor | 19713-548 | Preplant (Surface Applied) | Aerial | 1.5 |
| | | | | Ground | 1.5 |
| | | | Preplant Incorporated (2 inches) | Aerial | 1.5 |
| | | | | Ground | 1.5 |
| Soybeans[a] | Metolachor 7.8 | 60063-24 | Preplant Incorporated (2 inches) | Aerial | 1.67 |
| | | | | Ground | 1.67 |

[a] Included in the assessment, but not grown in California.

## 3.2 Aquatic Exposure Assessment

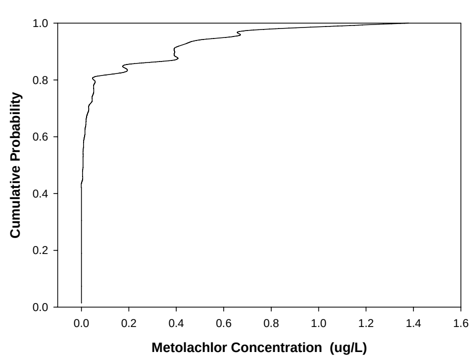
### 3.2.1 Surface Water Monitoring Data

An evaluation of the surface water monitoring data was conducted to assess the occurrence of metolachlor, metolachlor-OA, and metolachlor-ESA. Surface water data were obtained from the California Department of Pesticide Regulation (http://www.cdpr.ca.gov/docs/sw/surfdata.htm) and USGS NAWQA data warehouse. Summary statistics for each sampling site were calculated to show the maximum concentration, median concentration, and average concentration. The number of sampling years and sample frequency varied among monitoring sites. More importantly, the surface water sampling program was not targeted to the metolachlor use areas. Therefore, the monitoring data may not represent the most conservative occurrence data for metolachlor and it degradation products in California.

The maximum reported concentration of metolachlor in CADPR and USGS NAWQA monitoring data is 1.38 µg/L in the San Joaquin River near Stevinson and 3.88 µg/L in the Orestimba Creek at the River Road near Crows Landing, CA, respectively. The 90$^{th}$ percentile site metolachlor concentration is 0.394 µg/L in CADPR data and 0.666 µg/L in USGS NAWQA data.

A cumulative probability distribution of median site metolachlor concentrations are shown in Figure 14. The maximum median concentration of metolachlor in CADPR and USGS monitoring data is 0.086 µg/L in Mud Slough, a tributary of the San Joaquin River and 0.633 µg/L in the Orestimba Creek at the River Road near Crows Landing, CA, respectively. The 90$^{th}$ percentile site metolachlor concentration is 0.0162 µg/L in the CADPR data and 0.122 µg/L in the USGS NAWQA data.

**A.** **B.**



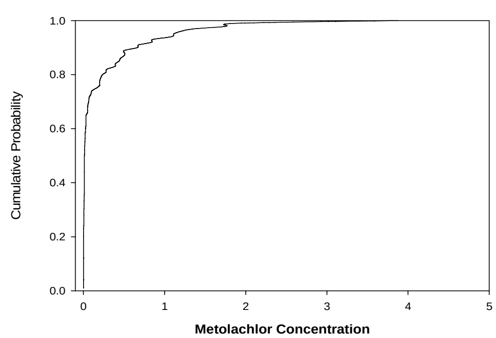




**Figure 14 Cumulative Probability Distribution of Maximum Metolachlor Concentrations for CADPR(A) and NAWQA (B)**

A. B.

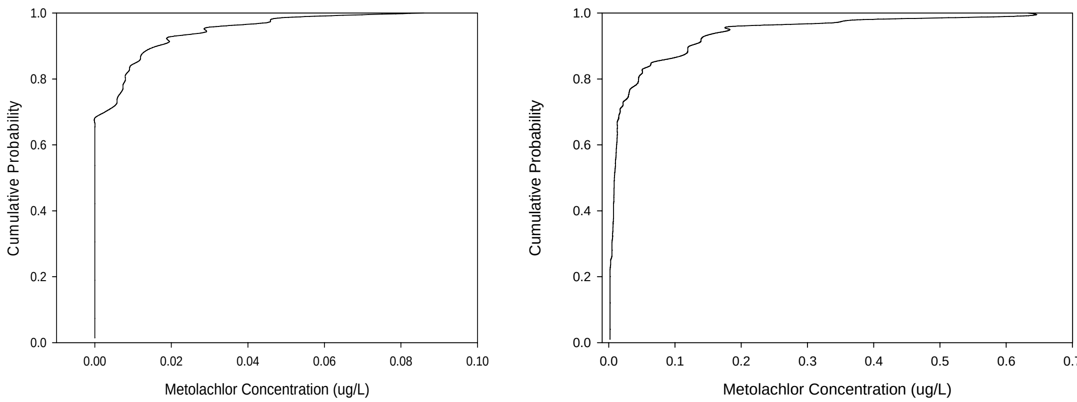



**Figure 15 Cumulative Probability Distribution of Median Metolachlor Concentrations for CADPR(A) and NAWQA (B)**

The maximum reported concentration of metolachlor OA and ESA is 0.086 and 0.0162 µg/L, respectively. A distributional analysis of metolachlor degradation product was not conducted because there were limited data on the metolachlor degradation products.

### *3.2.2 Air Monitoring*

An evaluation of air monitoring data was conducted to assess the occurrence of metolachlor, metolachlor-OA, and metolachlor-ESA. Air monitoring data were obtained from the California Department of Pesticide Regulation (Segawa, *et al*, 2003 and Kollman 2002). A review of the air monitoring data indicates that metolachlor was detected in trace quantities in an air monitoring study in Lompoc City, Santa Barbara County (Segawa, *et al*, 2003). Air concentrations of metolachlor were 1.7 ng/m$^3$ for the highest one day average, 1.01 ng/m$^3$ for the highest 3 day average, 0.54 ng/m$^3$ for the highest 18 day average concentration. Air concentrations of metolachlor were not reported in the California Pesticide Air Monitoring Results: 1986-2000 (Kollman 2002). Additionally, there was no air monitoring data for the metolachlor degradation products.

The potential impact of metolachlor air concentrations on surface water quality was assessed for the standard water body. Air concentrations above the pond are assumed to be the maximum reported air concentration in CA (1.7 ng/m$^3$). The potential contribution of volatile metolachlor was evaluated assuming different air volumes above the standard pond (10,000m$^2$). Air volumes were established according to 1 meter height increments above the pond. Air volumes ranged from 10,000 m$^3$ (1 meter height) to 655 x 10$^8$ m$^3$ (65,536 meter height). Mass loading of metolachlor was calculated assuming a complete rainwater "wash-out" from the air space above the pond. Metolachlor water concentrations from atmospheric deposition of volatile metolachlor are not expected to exceed 60 ng/L.

### *3.2.3 Aquatic Exposure Modeling*

Typically, the Agency conducts modeling using scenarios intended to represent use sites in areas that are highly vulnerable to either runoff, erosion, or spray drift. Runoff estimates predicted by the PRZM model are linked to the Exposure Analysis Modeling System (EXAMS). For ecological risk assessment, the Agency relies on a standard water body to receive the edge-of-field runoff estimates. The standard water body is of fixed geometry and includes the processes of degradation and sorption expected to occur in ponds, canals, and low order streams (*e.g.* first and second order streams). The water body is static (no outflow). The CLRF inhabits a range of water bodies, but generally prefers perennial or near perennial waters in order to complete its lifecycle (Jennings *et al.*, 1997). Generally it inhabits watersheds and drainages of 4$^{th}$ order or lower streams (Hayes and Jennings 1998).

#### *3.2.3.1 Aquatic Exposure Modeling Input and Output*

**Table 5 Input Parameters for PRZM Modeling**

| Parameter | Value | Comments | Source |
|---|---|---|---|
| Molecular Weight (grams/mole) | 283.8 | | |
| Solubility (mg/L) | 4800 | 10X reported value | product chemistry |
| Vapor Pressure (torr) | $2.8E^{-5}$ | | |
| Henry's Constant (atm $m^3$/mol) | $3.75E^{-5}$ | | |
| $K_d$ (L/kg) | 181 | Average $K_{oc}$[2] | MRID00078291<br>MRID43928935<br>MRID40430203<br>MRID40476404<br>MRID43928937<br>MRID40495602<br>MRID40495603<br>MRID40495604 |
| Aerobic Soil Metabolism Half-life (days) | 48.9 | Estimated upper[3] 90$^{th}$ percentile | MRID41309801<br>MRID43928936<br>MRID45499606 |
| Aerobic Aquatic Metabolism Half-life (days) | 141 | Based on 3X single aerobic aquatic metabolism linear first order half-life | MRID41185701 |
| Anaerobic Aquatic Metabolism Half-life (days) | 234 | Based on 2X single anaerobic aquatic metabolism linear first order half-life | MRID41185701 |
| Photodegradation in Water (days) | 70 | | MRID40430202 |
| Hydrolysis Half-life (days) | Stable | | MRID40430201 |
| Spray Drift Fraction | 5%<br>1% | Aerial<br>Ground | Default value |

[1] - Application rate given in input units for PRZM-EXAMS. Conversion is kg/ha =1.12 * lb/A

[2] - Average $K_{oc}$ using values 118.5, 303.0, 151.4, 241.4, 66.8, 21.6, 110.4, 74.4, 175.0, 333.3, 230.0, 244.7, 226.3, 367.2, 176.5, 120.7, 111.1 as per "*Guidance for Chemistry and Management Practice Input Parameters for Use in Modeling the Environmental Fate and Transport of Pesticides"* dated February 28, 2002.

[3] - Upper 90$^{th}$ Percentile based on acceptable aerobic metabolism half-lives of 66, 37.8, 37.8, 14.9, 13.9, and 50.3 days.

**Table 6  EECs in the Standard EXAMS Water Body**

| Crop (lb ai/A) | Application Timing (Depth of Incorporation) | Application Technique[a] | 1 in 10 year EEC (ug/L) | | |
|---|---|---|---|---|---|
| | | | Peak | 21 day average | 60 day Average |
| Corn (1.63) | Preplant Incorporated (2 inches) | Aerial | 7.752 | 4.510 | 2.040 |
| | | Ground | 4.551 | 2.788 | 1.261 |
| Cotton (1.33) | Post Emergent (2 inches) | Aerial | 3.722[c] | 2.004[c] | 0.897[c] |
| | | Ground | 1.052 | 0.546 | 0.256 |
| Cotton (1.00) | Pre Emergent (Surface Applied) | Aerial | 4.640 | 2.655 | 1.374 |
| | | Ground | 3.655 | 1.841 | 0.863 |
| Pod Crops (Legumes) (1.67) | Preplant Incorporated (4-6 inches) | Aerial | 5.994 | 3.566 | 1.450 |
| | | Ground | 4.088 | 1.882 | 0.953 |
| Potatoes (2.00) | Pre-emergent Incorporated (3 inches) | Aerial | 5.598 | 3.844 | 1.964 |
| | | Ground | 2.064 | 1.221 | 0.688 |
| Safflower (1.67) | Preplant Incorporated (4-6 inches) | Aerial | 4.873 | 3.049 | 1.472 |
| | | Ground | 1.944 | 1.078 | 0.597 |
| Sorghum (1.5) | Preplant (Surface Applied) | Aerial | 21.823[b] | 13.545[b] | 7.020[b] |
| | | Ground | 19.437 | 11.989 | 6.216 |
| | Preplant Incorporated (2 inches) | Aerial | 13.038 | 8.166 | 4.221 |
| | | Ground | 10.308 | 6.353 | 3.000 |
| Soybeans (1.67) | Preplant Incorporated (2 inches) | Aerial | 9.319 | 4.845 | 2.225 |
| | | Ground | 7.715 | 3.490 | 1.777 |

[a] Both aerial and ground applications were modeled.  Aerial applications typically result in higher aquatic EECs (due to greater spray drift), thus the aerial EECs are used as bounding estimates for each crop group.
[b] Used as the “highest” bounding estimate for developing risk quotients
[c] Used as the “lowest” bounding estimate for developing risk quotients.  Although some estimates for ground applications are lower, the application rate is a function of the crop, not the application method, thus it is more conservative to use the aerial EECs.

Although a complete fate data set was not available for the degradates metolachlor ESA and metolachlor OA, EFED used the PRZM-EXAMS to estimate aquatic concentrations. Data available included the soil adsorption/desorption studies for both ESA (MRID 44931722) and OA (MRID 40494605), as well as the conversion efficiency of metolachlor to the two degradates (MRIDs 43928936, 41309801). Application rates for the two degradates were determined by multiplying the metolachlor application rate by the fraction of the relevant degradate. Half-lives for the two compounds were estimated using the decline portion of the formation and decline data contained in the Comparative Aerobic Soil Metabolism Study (MRID 43928936). For other parameters where data were not available, the compounds were conservatively assumed to be stable. PRZM input parameters are shown below (Table 7) It should be noted that because of the assumptions of stability and the fact there is no outflow from the EXAMS pond, the equations in the models cause the degradates to appear to accumulate in the pond. The 1-in-10-year values used as concentration estimates are higher than they actually would be in the environment.

**Table 7 PRZM/EXAMS Parameters for Metolachlor ESA and Metolachlor OA**

| Parameter | Value | Comments | Source |
|---|---|---|---|
| Application Rate ESA (kg a.i./ha) | 0.26 | metolachlor kg ai/ha * 1.16[1]*0.12 | MRID43928936 |
| Application Rate OA (kg a.i./ha) | 0.52 | metolachlor kg ai/ha * 0.98[2]* 0.28 | MRID41309801 |
| Molecular Weight ESA (g/mole) | 329.7 | | |
| Molecular Weight OA (g/mole) | 279.4 | | |
| $K_d$ ESA (L/kg) | 0.041 | Lowest non-sandy soil, Maryland clay | MRID44931722 |
| $K_d$ OA (L/kg) | 0.079 | | MRID40494605 |
| Solubility (mg/L) | 4800 | 10X reported value of parent | product chemistry |
| Aerobic Soil Metabolism Half-life ESA (days) | 162.5 | Based on decline portion of formation and decline data | MRID4392836 |
| Aerobic Soil Metabolism Half-life OA (days) | 127.5 | | |
| Aerobic Aquatic Metabolism Half-life (days) | 0 | Assumed stable | |
| Anaerobic Aquatic Metabolism Half-life(days) | 0 | | |
| Photodegradation in Water (days) | 0 | | MRID40430202 |
| Hydrolysis Half-life (days) | 0 | | MRID40430201 |

[1] Molecular weight correction factor= MW ESA (329.7 g/mol)/MW Metolachlor (283.8 g/mol) =1.16
[2] Molecular weight correction factor= MW OA (279.4 g/mol)/ MW Metolachlor (283.8 g/mol) = 0.98

Table 8  EECs in the Standard Pond-Metolachlor ESA

| Crop | Application Timing (Depth of Incorporation) | 1 in 10 year EEC (ug/L) | | |
|---|---|---|---|---|
| | | Peak | 21 day average | 60 day Average |
| Corn | Preplant (2 inches) | 3.407 | 3.407 | 3.406 |
| Cotton | Post Emergent (2 inches) | 1.153 | 1.153 | 1.148 |
| | Pre Emergent (Surface Applied) | 3.156 | 3.155 | 3.154 |
| Pod Crops (Legumes) | Preplant (4-6 inches) | 2.998 | 2.970 | 2.948 |
| Potatoes | Pre-emergent (3 inches) | 0.386 | 0.386 | 0.386 |
| Safflower | Preplant 4-6 inches | 1.450 | 1.450 | 1.450 |
| Sorghum | Preplant (Surface Applied) | 21.328 | 21.328 | 21.328 |
| | Preplant I (2 inches) | 10.884 | 10.884 | 10.884 |
| Soybeans | Preplant (2 inches) | 5.998 | 5.942 | 5.895 |

Table 9  EECs in the Standard Pond-Metolachlor OA

| Crop | Application Timing (Depth of Incorporation) | 1 in 10 year EEC (ug/L) | | |
|---|---|---|---|---|
| | | Peak | 21 day average | 60 day Average |
| Pod Crops (Legumes) | Preplant (4-6 inches) | 6.131 | 6.073 | 6.026 |
| Corn | Preplant (2 inches) | 6.884 | 6.883 | 6.882 |
| Cotton | Post Emergent (2 inches) | 2.741 | 2.741 | 2.741 |
| | Pre Emergent (Surface Applied) | 7.900 | 7.900 | 7.900 |
| Soybeans | Preplant (2 inches) | 12.261 | 12.144 | 12.054 |
| Potatoes | Pre-emergent (3 inches) | 1.075 | 1.075 | 1.075 |
| Safflower | Preplant 4-6 inches | 2.755 | 2.754 | 2.753 |
| Sorghum | Preplant (Surface Applied) | 42.982 | 42.982 | 42.982 |
| | Preplant I (2 inches) | 21.926 | 21.926 | 21.926 |
| Soybeans | Preplant (2 inches) | 12.261 | 12.144 | 12.054 |

### 3.2.4 Aquatic Exposure Summary

Both aerial and ground applications were modeled with PRZM-EXAMS, and EECs for both are reported in Table 6. Aerial applications typically result in higher aquatic EECs (due to greater spray drift), thus the aerial EECs are used as risk quotient bounding estimates for each crop group. All current labels permit both aerial and ground application methods. Because metolachlor is incorporated into the soil for some applications, aquatic EECs for some crops may vary even though application rates are the same. Surface applications generally result in higher concentrations in field runoff than treatments incorporated into the soil. No sorghum usage was reported in the CDPR PUR database for 2002-2005.

Peak metolachlor concentrations for aerial applications (Table 6) ranged from 3.7 μg/L (cotton, post emergent, incorporated) to 21.8 μg/L (sorghum, preplant, surface applied). Peak EECs for all crops other than sorghum were < 10 μg/L. The 21-day average concentrations ranged from 2.0 μg/L (cotton, post emergent, incorporated) to 13.5 μg/L (sorghum, preplant, surface applied). The 60-day average concentrations ranged from 0.9 μg/L (cotton, post emergent, incorporated) to 7.0 μg/L (sorghum, preplant, surface applied).

Peak metolachlor concentrations for ground applications (Table 6) ranged from 1.1 μg/L (cotton, post emergent, incorporated) to 19.4 μg/L (sorghum, preplant, surface applied). Peak EECs for all crops other than sorghum were < 8 μg/L. The 21-day average concentrations ranged from 0.5 μg/L (cotton, post emergent, incorporated) to 12.0 μg/L (sorghum, preplant, surface applied). The 60-day average concentrations ranged from 0.3 μg/L (cotton, post emergent, incorporated) to 6.2 μg/L (sorghum, preplant, surface applied).

Concentrations of the degradates were not adjusted for spray drift fraction because they are assumed to form when metolchlor is in contact with soil and/or water. Estimated concentrations (Table 8 and Table 9) are higher than would actually occur in the environment because a compound that is stable to degradation "accumulates" in the modeled pond due to lack of outflow. Reported concentrations are a highly conservative estimate. Because of the "accumulation," peak concentrations, 21-day concentrations, and 60-day concentrations were approximately the same for most crops. Concentrations for metolachlor-ESA ranged from 0.4 μg/L (potatoes, pre-emergent) to 21.3 μg/L (sorghum, preplant, surface applied). Metolachlor-OA concentrations ranged from 1.1 μg/L (potatoes, pre-emergent) to 43.0 μg/L (sorghum, preplant, surface applied).

## 3.3 Terrestrial Exposure

### 3.3.1 Bird and Mammal Exposure (TREX)

EFED estimates exposure of birds and mammals to pesticides using the Terrestrial Exposure Model (TREX). TREX uses the Kenaga nomagram, as modified by Fletcher *et al.* (1994) to determine pesticide residues on several categories of food items, then calculates the potential dose an organism might receive from ingesting contaminated items using allometric equations. Dose estimates in Table 11 and Table 12 Mammal Dose Estimates are based on an upper bound dose and the assumptions that the organism exclusively eats one type of food item and forages only in the treated and/or overspray areas. Metolachlor translocates in plant tissue and the residence time of the parent compound or any degradates is unknown. Labels permit only a single application, thus residues in the plant are not expected to exceed residues on the plant.

**Table 10 Input Parameters for TREX and T-HERPS**

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

**Table 11 Bird Dose Estimates**

| Feeding Category | Dietary-based EECs (mg/kg-food item) | Dose-based EECs (mg/kg-bw) | |
|---|---|---|---|
| | | Small (20 g) | Medium (100 g) |
| *Potatoes 2 lb ai/A (highest)* | | | |
| Short grass | 480 | 547 | 312 |
| Tall grass | 220 | 251 | 142 |
| Broadleaf plants/small insects | 270 | 308 | 175 |
| Fruits/pods/seeds/large insects | 30 | 34.2 | 19.5 |
| *Cotton 1 lb ai/A (lowest)* | | | |
| Short grass | 240 | 273 | 156 |
| Tall grass | 110 | 125 | 71 |
| Broadleaf plants/small insects | 135 | 154 | 87.7 |
| Fruits/pods/seeds/large insects | 15 | 17.1 | 9.74 |

**Table 12 Mammal Dose Estimates**

| Feeding Category | Dietary-based EECs (mg/kg-food item) | Dose-based EECs (mg/kg-bw) | |
|---|---|---|---|
| | | Small (15 g) | Medium (35 g) |
| *Potatoes 2 lb ai/A (highest)* | | | |
| *Herbivores and Insectivores* | | | |
| Short grass | 480 | 458 | 316 |
| Tall grass | 220 | 210 | 145 |
| Broadleaf plants/small insects | 270 | 257 | 178 |
| Fruits/pods/seeds/large insects | 30 | 28.6 | 19.8 |
| *Granivores* | | | |
| Fruits/pods/seeds/large insects | 30 | 6.36 | 4.39 |
| *Cotton 1 lb ai/A (lowest)* | | | |
| *Herbivores and Insectivores* | | | |
| Short grass | 240 | 229 | 158 |
| Tall grass | 110 | 105 | 72.5 |
| Broadleaf plants/small insects | 135 | 129 | 89.0 |
| Fruits/pods/seeds/large insects | 15.0 | 14.3 | 9.88 |
| *Granivores* | | | |
| Fruits/pods/seeds/large insects | 15.0 | 3.18 | 2.20 |

### *3.3.2 Terrestrial Invertebrate Exposure*

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

**Table 13 Terrestrial Invertebrate Exposure**

| Application Rate (lb ai/A) | Insect Size Category | EECs (mg ai/kg insect) | Dose per Bee (μg ai/bee) |
|---|---|---|---|
| Potatoes (2 lb ai/A) Highest | Small insects | 270 | 34.6 |
| | Large insects | 30 | 3.84 |
| Cotton pre-emergent (1 lb ai/A) Lowest | Small insects | 135 | 17.3 |
| | Large insects | 15 | 1.92 |

### *3.3.3 Terrestrial Plant Exposure*

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

#### *3.3.3.1 TerrPlant*

TerrPlant has two basic exposure scenarios. The first is an adjacent upland area, which is exposed to the pesticide via drift and dissolved concentrations in sheet runoff. The second is an adjacent semi-aquatic (wetland) area, which is exposed to the pesticide via drift and to dissolved concentrations in channelized runoff. Drift is calculated as a percentage of the application rate (1% for ground, and 5% for aerial, airblast, or spray chemigation) and is not adjusted for distance from the application site. The amount of dissolved pesticide in the runoff component is estimated based on solubility of the active ingredient. TerrPlant estimates are shown in Table 14. Total loading in upland areas (runoff plus drift) ranged from 0.10 lb ai/A (sorghum, surface applied, aerial) to 0.15 lb ai/A (cotton, pre-emergent, surface applied). Total loading in wetland areas (runoff plus drift) ranged from 0.29 lb ai/A (pod crops & safflower, aerial) to 0.83 lb ai/A (sorghum, surface applied, aerial). Pesticide loading to the different areas is affected by application rate and depth of incorporation. Concentrations of metolachlor is the runoff is more important in the wetland than for the upland. Thus, the specific crops used for the bounding estimates may not be the same. Based on the TerrPlant model, spray drift to either a wetland or an upland area ranged from 0.05 lb ai/A (cotton, pre-emergent, surface applied) to 0.10 lb ai/A (potatoes, aerial). In this model, spray drift is strictly a function of application rate and method (ground vs. aerial). Loading estimates are presented in Table 14.

**Table 14 Terrestrial Plant Exposure (TerrPlant)**

| Crop and Application Rate (lb ai/A) | Total Loading (Runoff +Drift) (lb ai/A) | | Drift EEC (lb ai/A) |
|---|---|---|---|
| | Upland areas | Wetland areas | All areas |
| *Highest EECs* | | | |
| Sorghum surface applied aerial (1.5 lb ai/A) | 0.15 | 0.83 | [a] |
| Potatoes-aerial (2 lb ai/A) | [a] | [a] | 0.10 |
| *Lowest EECs* | | | |
| Cotton pre-emergent surface applied aerial (1 lb ai/A) | 0.10 | [a] | 0.05 |
| Pod crops aerial & safflower aerial (1.67 lb ai/A) | [a] | 0.29 | [a] |

[a] Total loading to adjacent areas is a function of both runoff and spray drift, which are influenced by application rate, and depth of incorporation for soil incorporated compounds. For metolachlor, the highest and lowest EECs for the various receiving compartments are not always the same crop and/or application method. Greyed squares indicate an EEC that was not used as a bounding estimate.

### *3.3.3.2 AgDrift*

Because of concerns about potential effects on non-target plants located in overspray or spray drift areas, EFED used AgDrift modeling software to estimate the clearance distances presented in the risk characterization section (Table 23). AgDrift was developed using extensive field-measured data sets, and provides a method of estimating deposition of the compound of concern at a specified distance away from the application source. The range for AgDrift Tier 1 aerial estimates, point deposition mode, is 997 ft. Deposition is heavily dependent on the method of application and droplet size. A Tier I analysis is driven primarily by these two variables. AgDrift was run in the Tier I, Terrestrial Assessment, Point Deposition mode. Droplet size evaluated was ASAE fine-medium (default), and the $90^{th}$ percentile estimate was used. For the Tier I conditions, the 1% drift assumed in TerrPlant for ground applications is equivalent to approximately 100 feet away from the application site. The 5% drift assumed for aerial applications is equivalent to approximately 200 feet away from the application site. At the extent of the range (997 ft) point deposition is approximately 1% for aerial applications.

## *4.0 Effects Assessment*

## *4.1 Aquatic Toxicity Profile*

Acute toxicity data for metolachlor used to evaluate the assessment endpoints is presented in Table 15. EFED uses the most sensitive species in each evaluation category to assess risk. The complete set of toxicity data available to EFED at the time of the assessment is contained in Appendix B. The data set consists of toxicity data from acceptable guideline tests submitted to the Agency by the registrant and open literature toxicity data that meets established acceptability criteria ("ECOTOX data"). The complete data set includes values for both racemic metolachlor (PC#108801) and S-metolachlor (PC108800). No open literature data were located for either metolachlor-ESA or metolachlor OA, thus this portion of the toxicity data only includes registrant-submitted guideline studies.

Metolachlor is slightly toxic to moderately toxic to fish ($LC_{50}$s 3.2-15.0 mg/L, Appendix B, Table 1 and Table 6) on an acute basis. Some amphibian data (Appendix B, Table 6) was located in ECOTOX. Toxicity data for two species, the African clawed frog (*Xenopus laevis*, $LC_{50}$ 13.6 mg/L) and American bullfrog (*Rana catesbeiana*, $EC_{50}$ 17.4 mg/L) indicated that mortality effects for amphibians occur in concentrations similar to lethal endpoints for fish, which serve as a surrogate for aquatic phase amphibians. Species sensitivity distributions for amphibians are not well understood at this point, thus EFED opted to use the more protective toxicity value from the fish data to calculate RQs. Metolachlor-ESA is slightly toxic to fish ($LC_{50}$ 48 mg/L) and metolachlor-OA is practically non-toxic to fish ($LC_{50}$ >93.1 mg/L). No amphibian data were located for the degradates. Sub-lethal effects noted in tests include lethargy and loss of equilibrium, occurring at concentrations of $\geq$ 3.3 mg/L. The NOAEC in chronic tests (fathead minnow) is 1 mg/L.

Metolachlor is slightly toxic to moderately toxic to freshwater invertebrates ($EC_{50}$s 1.1-26.0 mg/L, Appendix B, Table 1 and Table 6).The lowest chronic toxicity value for tests that evaluated decreases in survival, reproduction and growth was for the water flea (*Ceriodaphnia dubia*, NOAEC 0.001 mg/L). One study from open literature noted sublethal effects (behavioral modifications) in rusty crayfish *(Oronectes rusticus)* at metolachlor concentrations of 0.025 mg/L (Appendix B, Table 6). Metolachlor-ESA is practically non-toxic and metolachlor-OA is slightly toxic to the water flea (*Daphnia magna).* No chronic toxicity data were located for the degradates.

Based on registrant-submitted data, green algae are the most sensitive aquatic plants ($EC_{50}$ 0.008 mg/L, NOAEC 0.002 mg/L), and, as expected for an herbicide, plants are several orders of magnitude more sensitive than the aquatic animals. Toxicity values for various genera of aquatic plants ranged from 0.008 mg/L (green algae, *s*-metolachlor) to 1.2 mg/L (bluegreen algae, racemic metolachlor). Duckweed (*Lemna gibba*, $EC_{50}$ 0.048 mg/L) which is the surrogate for aquatic vascular plants, is less sensitive to the effects of metolachlor than the green alga, but more sensitive than any of the other non-vascular aquatic plants. For three genera (green alga, duckweed and saltwater diatoms), toxicity data were available for both racemic metolachlor and *s*-metolachlor. Based on these data, green algae and duckweed are slightly more sensitive to *s*-metolachlo*r*, and saltwater diatoms are less sensitive. The more sensitive *s*-metolachlor data are used in this assessment. Green algae and duckweed are much less sensitive to both metolachlor degradates ($EC_{50}$s >40mg/L).

**Table 15 Aquatic Toxicity Profile for Metolachlor**

| Assessment Endpoint | Surrogate Species | Toxicity Value Used | Source Citation | Comments |
|---|---|---|---|---|
| *Direct Effects* | | | | |
| Acute Toxicity to Frog | Bluegill sunfish | $LC_{50}$ = 3.2 mg/L<br>95% CI = 2.8-4.6 mg/L<br>Slope = 14.8 | MRID 43928910 | Sub-lethal effects: loss of equilibrium (3.3 ppm) |
| Chronic Toxicity to Frog | Fathead minnow | NOAEC = 1 mg/L<br>LOAEC = 2.2 mg/L | MRID 43044602 | Increase in mortality noted at ≥5 ppm<br>Hatch rate affected at 8.6 ppm |
| *Indirect Effects and Critical Habitat Effects (Prey Reduction))* | | | | |
| Acute Toxicity to Aquatic Invertebrates | Water flea | $EC_{50}$ = 1.1 mg/L | ECOTOX ref # 67777 | Endpoint measured was immobilization (*i.e.* mortality) |
| Chronic Toxicity to Aquatic Invertebrates | Water flea | NOAEC = 0.001 mg/L<br>LOAEC = 0.01 mg/L | ECOTOX ref# 83887 | Number of young per female significantly different at 0.01 mg/L. Intrinsic rate of increase (*r*) decreased at 0.01 mg/L No other parameters affected until 1 mg/L |
| *Indirect Effects and Critical Habitat Effects (Habitat Modification)* | | | | |
| Acute Toxicity to Plants (non-vascular) | Green algae | $LC_{50}$ = 0.008 ppm<br>95% CI = 0.003-0.025 ppm<br>Slope = 3 | MRID 43928929 | None |
| Acute Toxicity to Plants (vascular) | Duckweed | $LC_{50}$ = 0.021 ppm<br>95% CI = 0.019-0.023 ppm | MRID 43928931 | None |

[a] Adult frogs are no longer in the "aquatic phase" of the amphibian life cycle; however, submerged adult frogs are considered "aquatic" for the purposes of this assessment because exposure pathways in the water are considerably different that exposure pathways on land.
[b] Birds are used as surrogates for terrestrial phase amphibians.

**Table 16 Aquatic Toxicity Profile for Degradate Metolachlor OA**

| Assessment Endpoint | Surrogate Species | Toxicity Value Used | Source Citation | Comments |
|---|---|---|---|---|
| *Direct Effects* | | | | |
| Acute Toxicity to Frog | Crucian carp | $LC_{50}$ = >93.1 mg/L<br>NOAEC = >96.3 mg/L | MRID 44929502 | |
| Chronic Toxicity to Frog | No data available | | | |
| *Indirect Effects and Critical Habitat Effects (Prey Reduction)* | | | | |
| Acute Toxicity to Aquatic Invertebrate | Water flea | $LC_{50}$ = 15.4 mg/L<br>95%CI=13.0-18.4 mg/L<br>Slope = 6.1 | MRID 44929503 | |
| Chronic Toxicity to Aquatic Invertebrate | No data available | | | |
| *Indirect Effects and Critical Habitat Effects (Habitat Modification)* | | | | |
| Acute Toxicity to Plants (non-vascular) | Green algae | $LC_{50}$ = 57.1 mg/L<br>NOAEC = 29.3 mg/L | MRID 4492515 | None |
| Acute Toxicity to Plants (vascular) | Duckweed | $LC_{50}$ = >95.1 mg/L<br>NOAEC = 95.4 mg/L | MRID 4429514 | None |

**Table 17 Aquatic Toxicity Profile for Degradate Metolachlor ESA**

| Assessment Endpoint | Surrogate Species | Toxicity Value Used | Source Citation | Comments |
|---|---|---|---|---|
| *Direct Effects* | | | | |
| Acute Toxicity to Frog | Rainbow trout | $LC_{50}$ = 48 mg/L<br>95% CI = 36-64 mg/L<br>NOAEC = 36 mg/L | MRID 449931702 | Sub-lethal effects at ≥58 ppm : loss of equilibrium, erratic swimming, pigmentation changes |
| Chronic Toxicity to Frog | No data available | | | |
| *Indirect Effects and Critical Habitat Effects (Prey Reduction)* | | | | |
| Acute Toxicity to Aquatic Invertebrates | Water flea | $LC_{50}$ = >108 mg/L<br>NOAEC = 108 mg/L | MRID 44931703 | 108 ppm highest concentration tested |
| Chronic Toxicity to Aquatic Invertebrates | No data available | | | |
| *Indirect Effects and Critical Habitat Effects (Habitat Modification)* | | | | |
| Acute Toxicity to Plants (non-vascular) | Green algae | $LC_{50}$ = >99.45 mg/L<br>NOAEC = 99.45 mg/L | MRID 44931720 | None |
| Acute Toxicity to Plants (vascular) | Duckweed | $LC_{50}$ = >95.1 mg/L<br>NOAEC = 95.4 mg/L | MRID 44931719 | None |

## 4.2 *Summary of Aquatic Ecotoxicity Studies*

Information used to develop the toxicity profile for metolachlor included registrant-submitted guideline studies for both racemic metolachlor and S-metolachlor, and open literature studies that met the criteria for inclusion into ECOTOX. Open literature studies generally do not indicate whether the active ingredient tested was racemic metolachlor or S-metolachlor. The lowest values for various taxon, used to derive RQs, are reported in Table 15. Data for the degradates are reported in Table 16 and Table 17. In all cases the lowest available endpoint (based on $LC_{50}$ for acute tests, and NOAEC for chronic tests) was used in the calculation.

### *4.2.1 Toxicity to Freshwater Fish*

#### *4.2.1.1 Acute Exposure (Mortality) Studies*

A number of guideline studies evaluating the acute effects of metolachlor on freshwater fish were available. $LC_{50}$s for fish ranged from 3.2 mg/L to 15.0 mg/L, classifying metolachlor as moderately to slightly toxic to fish. Sub-lethal effects noted in several studies included loss of equilibrium and lethargy. Generally, sub-lethal effects occurred at the same concentrations as mortality. A number of different species were considered. No obvious pattern related to species sensitivity distribution was noted. (*e.g.*, warm water fish being more or less sensitive than coldwater fish.) Data from ECOTOX studies (ECOTOX #6797) gave the $LC_{50}$ as 8.0-8.4 mg/L.

An acute toxicity study assessing the effects of metolachlor-ESA (MRID 44931702) on rainbow trout *(Onchorynchus mykiss)* showed the degradate to be less toxic than the parent. The $LC_{50}$ was 48 mg/L, classifying metolachlor ESA as slightly toxic to fish. In concentrations where mortality occurred, sub-lethal effects noted included erratic swimming, loss of equilibrium, and pigmentation changes.

Acute toxicity studies were available for the effects of metolachlor-OA on two fish species, crucian carp *(Carassius carassius* MRID 44929501*)*, and rainbow trout (*Onchorynchus mykis*s, MRID 44929502). The degradate is practically non-toxic to fish on an acute basis with $LC_{50}$s of >93.1 mg/L and >96.3 mg/L, respectively.

#### *4.2.1.2 Chronic Exposure (Growth/Reproduction) Studies*

The only chronic study available for freshwater fish was a registrant-submitted study on fathead minnow (*Pimephales promela*s). The NOAEC for the most sensitive endpoint, dry weight of the larval fish, was 0.030 mg/L. The LOAEC was 0.056 mg/L.

#### *4.2.2 Toxicity to Aquatic Phase Amphibians*

No guidelines currently exist for amphibian toxicity studies. However, several studies evaluating the acute and chronic effects of metolachlor on two species of frogs met the criteria for inclusion into ECOTOX. Endpoints derived from these studies occurred at higher concentrations than the effects reported for the guideline fish studies, which are typically used as a surrogate for amphibians. Differences in the species sensitivity distributions of fish and amphibians are not well understood. Because of this fact, EFED has elected to use the more protective fish-derived toxicity values in this assessment.

##### *4.2.2.1 Acute Exposure (Mortality) Studies*

Two acute toxicity studies for amphibians were reported in ECOTOX, one for the African clawed frog (*Xenopus laevis*, $LC_{50}$ 13.6 mg/L) and one for the American bullfrog (*Rana catesbeiana*, $EC_{50}$ 17.4 mg/L). These values are higher than almost all of the $LC_{50}$s reported for fish. Based on these data, metolachlor would be classified as slightly toxic to amphibians. For the bullfrog, the test consisted of exposing tadpoles to the metolachlor-containing formulation DUAL-960E for 24 hours under static conditions (ECOTOX #20274). The study contained no mention of analyzing the solution for active ingredient, thus EFED assumes the reported concentrations are nominal. Sublethal effects reported in this study include cellular damage (LOAEL 0.272 mg/L).

The study on the African clawed frog (ECOTOX# 66376) exposed embryos from wild-collected frogs to static concentrations of metolachlor (reported purity 99%). The reported 96-hour $LC_{50}$ for metolachlor was 13.6 mg/L. The study also determined 96-hour $LC_{50}$s for two degradation products of metolachlor (2,6-diethylaniline and 2-ethyl-6-methylaniline). These $LC_{50}$s were 19.4 mg/L and 68.8 mg/L, respectively. Based on guideline fate studies, degradates evaluated in this study are not considered “major” degradates of metolachlor, and are not addresssed in this assessment. Sublethal effects in embryos exposed to metolachlor included edema, gut malformations, axial flexures, and eye abnormalities. Similar effects were noted for the degradates, although to a lesser extent.

#### *4.2.3 Toxicity to Freshwater Invertebrates*

##### *4.2.3.1 Acute Exposure (Mortality) Studies*

Registrant-submitted toxicity tests show metolachlor (MRID 00015546) and *s*-metolachlor (MRID 43928912) to be slightly toxic to daphnids on an acute basis. $LC_{50}$s for *Daphnia magna* ranged from 25-26 mg/L. NOAECs from these studies were 5.6 mg/L and 4.8 mg/L, respectively. Sublethal effects included lethargy.

Several open literature studies were available in ECOTOX for aquatic invertebrates. Some produced $EC_{50}$s in the same range (~25 mg/L) as registrant-submitted data. However, several studies contained $EC_{50}$s that were lower, in the 1.1-4.4 mg/L range. The lowest endpoint from these studies, was used to calculate RQs for this assessment (*Ceriodaphnia dubia* (water flea) ECOTOX # 67777, 48-hr $EC_{50}$ 1.1 mg/L). The next lowest endpoint from these studies was for *Chironomus plumosus* (midge fly larvae, ECOTOX #6797) In the midge fly study, both technical metolachlor (95.4% purity) and an emulsifiable concentrate (87% a.i.) were used in 48-hour static tests. The $LC_{50}$s for the tests were 3.8 mg/L (technical) and 4.4 mg/L (concentrate).

Studies on *Daphnia magna* were submitted for both major degradates. The $LC_{50}$ for metolachlor-OA is 15.4 mg/L (MRID 44929503), classifying it as slightly toxic to aquatic invertebrates. For the metolachlor-ESA, the $LC_{50}$ was >108 mg/L, (MRID 44931703), classifying it as practically non-toxic to aquatic invertebrates.

#### *4.2.3.2 Chronic Exposure (Growth/Reproduction) Studies*

The registrant submitted a full life cycle study assessing the effects of metolachlor on *Daphnia magna* (MRID 43802601). Measured concentrations were highly variable throughout the study, thus the lowest measured concentrations were used to derive conservative endpoints. Based on growth and reproduction, the NOAEC and LOAEC were 3.2 and 6.9 mg/L, respectively. ECOTOX located a chronic study on *Daphnia magna* (ECOTOX# 83887) which produced lower endpoints than the registrant-submitted study. These endpoints were used in the assessment. Authors for the study note that OECD and ISO guidelines were followed in conducting the tests. The 21-day study compared the responses of *Daphnia magna* to racemic and *s*-metolachlor. Parameters evaluated included length, longevity, days to first brood, broods per female, and number of young per female. The most sensitive parameter was number of young per female. The study established an NOAEC of 0.001 mg/L and an LOAEC of 0.01 mg/L. The reported NOAEC for *Ceriodaphnia dubia* is 6.25 mg/L (ECOTOX #13689) and a reported LOAEC for the sour paste nematode is 2 mg/L.

#### *4.2.3.3 Sublethal Effects*

One study located in the open literature evaluated effects of technical metolachlor on the behavior of rusty crayfish (*Oronectes rusticus*, ECOTOX #68515). Crayfish were collected from the wild and tested for their ability to respond appropriately to odor cues following exposure to metolachlor. Both a positive (food odor) and negative (predator odor) cue were tested. Measurements included length of time to locate the odor source, percent success in locating the odor source, and time spent motionless. Concentrations of metolachlor tested ranged from 0.025-0.075 mg/L, and included both a negative control and a solvent control. At a concentration of 0.025 mg/L, the crayfish had less success in finding the food source, took longer to find the food source, and exhibited modifications in alarm response. There appeared to be a dose-response relationship. Based on this study, the behavioral NOAEC is <0.025 mg/L and the LOAEC is 0.025 mg/L.

### 4.2.4 Toxicity to Aquatic Plants

EFED evaluated both registrant submitted studies and open literature studies for aquatic plants. Overall, based on a review of the data, the endpoints and test durations used by independent evaluators are similar to those in the guideline studies. Guideline studies provided more sensitive endpoints, and these were used in the assessment.

The registrant submitted aquatic plant studies for racemic metolachlor, s-metolachlor and for the two major degradates, metolachlor-ESA and metolachlor-OA. For the racemic metolachlor testm all five standard aquatic plant species were tested. $EC_{50}$ values ranged from 0.010 mg/L (green alga) to 1.2 mg/L (blue-green alga). NOAEC s ranged from 0.0007 mg/L (green alga) to 0.063 mg/L (blue-green alga). For *s*-metolachlor, data were submitted for the three aquatic plants most sensitive to metolachlor (green algae, duckweed, marine diatom). *S*-metolachlor $EC_{50}$s ranged from 0.008 mg/L (green alga) to 0.11 mg/L. NOAEC values ranged from 0.0015 mg/L (green alga) to 0.021 mg/L (marine diatom). Each of the two major degradates was tested with both a non-vascular (green alga) and a vascular (duckweed) plant. Both degradates are less toxic to aquatic plants than the parent compounds. Of the two plants tested, duckweed is the more sensitive to metolachlor–ESA, with an $EC_{50}$ of 43 mg/L and a NOAEC of 4 mg/L. Green alga is the more sensitive to metolcahlor-OA, with an $EC_{50}$ of 57 mg/L and a NOAEC of 29 mg/L.

## 4.3 Terrestrial Toxicity Profile

Using a dose estimate, metolachlor is practically non-toxic to birds and mammals on an acute basis. Using a dietary estimate, it is classified as slightly toxic to birds. No dietary data were available for mammals. Chronic reproductive effects for birds were noted at 1000 mg/kg diet. Chronic reproductive effects for mammals were reported at 6.9 mg/kg bw. No definitive endpoint has been established for terrestrial invertebrates. Metolachlor is an herbicide, and affects terrestrial plants at concentrations as low as 0.0048 lb ai/A. Monocots are slightly more sensitive than dicots. Effects on terrestrial plants are more pronounced in the seedling emergence tests than in the vegetative vigor tests.

**Table 18 Terrestrial Toxicity Profile for Metolachlor**

| Assessment Endpoint | Surrogate Species | Toxicity Value Used | Source Citation | Comments |
|---|---|---|---|---|
| *Direct Effects* | | | | |
| Acute Toxicity to Frog | Bobwhite quail | $LD_{50}$=>2510 mg/kg bw (dose)<br>$LC_{50}$=>4912 mg/kg bw (dietary) | MRID 43928907<br>MRID 43928908 | No treatment related mortality reported in any studies |
| Chronic Toxicity to Frog | Bobwhite quail | NOAEC=403 mg/kg diet<br>LOAEC=1010 mg/kg diet | MRID 43044602 | Reduction in egg quality (number of eggs laid) |
| *Indirect Effects and Critical Habitat Effects (Prey Reduction)* | | | | |
| Acute Toxicity to Terrestrial Invertebrates | Honey bee | $LD_{50}$ = >200 $\mu$g/bee<br>NOAEL = 200 $\mu$g/bee | MRID 44718402 | None |
| Acute Toxicity to Mouse | Rat | $LD_{50}$=2514 mg/kg bw (dose) | MRID 00015523 | Treatment related mortality at all doses tested (1670-4640 mg/kg bw) |
| Acute Toxicity to Frog | Bobwhite quail | $LD_{50}$=>2510 mg/kg bw (dose)<br>$LC_{50}$=>4912 mg/kg bw (dietary) | MRID 43928907<br>MRID 43928908 | No treatment related mortality reported in any studies |
| Chronic Toxicity to Terrestrial Invertebrates | No data available | | | |
| Chronic Toxicity to Mouse | Rat | NOAEC= 3.2 mg/kg bw<br>LOAEC= 6.9 mg/kg bw | MRID 43802601 | Reduced pup weights in $F_1$ and $F_2$ litters |
| Chronic Toxicity to Frog | Bobwhite quail | NOAEC=300 mg/kg diet<br>LOAEC=1000 mg/kg diet | MRID 000808097 | Reduction in egg quality (number of eggs laid) |
| *Indirect Effects and Critical Habitat Effects (Habitat Modification)* | | | | |
| Acute Toxicity to Terrestrial Plants (Wetland)<br>Acute Toxicity to Terrestrial Plants (Upland) | Ryegrass (Monocot)<br>Lettuce (Dicot) | $EC_{25}$=0.0048 lb ai/A<br>NOAEC=0.001 lb ai/A<br>$EC_{25}$=0.0057 lb ai/A<br>NOAEC=0.0003 lb ai/A | MRID 43928932 | Seedling emergence guideline test |

[a] Adult frogs are no longer in the "aquatic phase" of the amphibian life cycle; however, submerged adult frogs are considered "aquatic" for the purposes of this assessment because exposure pathways in the water are considerably different that exposure pathways on land.
[b] Birds are used as surrogates for terrestrial phase amphibians.

## 4.4 Summary of Terrestrial Ecotoxicity Studies

### 4.4.1 Terrestrial Vertebrates (Birds and Mammals)

Registrant-submitted data were available for both birds and mammals. No treatment-related mortality was reported in any of the bird studies. On an acute effects basis, *s*-metolachlor is practically non-toxic to birds (MRIDs 43928907, 43928906). No data for *s*-metolachlor were available for mammals, but testing of racemic metolachlor on rats (MRID 00015523) classifies it as practically non-toxic ($LD_{50}$ 2514 mg/kg bw). Treatment-related mortality occurred in rats at all doses tested (1670-4640 mg/kg bw).

Registrant submitted studies indicate metolachlor has chronic reproductive effects on birds (MRID 46508901) and mammals (MRID 000808097) at the highest tested dose. These effects, noted for both groups of organisms at approximately 1000 mg/kg diet, include a reduction in the number of eggs (birds) and reduced weight of pups (rats). No treatment-related mortality or other effects were noted in the studies.

No data regarding metolachlor or *s*-metolachlor effects on terrestrial vertebrates is currently available in ECOTOX.

### 4.4.2 Terrestrial Invertebrates

The only guideline insect tests are for honeybees. Registrant submitted studies include acute contact and acute oral toxicity studies for *s*-metolachlor (MRID 44718402, Core). The acute contact $LD_{50}$ is >200 μg a.i./bee and the oral $LD_{50}$ is >85 μg a.i./bee. NOAELS are 200 μg a.i./bee for acute contact and 85 μg a.i./bee for oral dose.

No data regarding metolachlor or *s*-metolachlor effects on terrestrial invertebrates is currently available in ECOTOX.

### 4.2.3 Terrestrial Plants

Vegetative vigor and seedling emergence guideline tests were available for *s*-metolachlor. Both monocots and dicots were more sensitive in the seedling emergence tests, which was not unexpected given *S*-metolachlor's mode of action. The seedling emergence $EC_{25}$ for the most sensitive monocot (ryegrass) was 0.0048 lb ai/A, and the $EC_{25}$ for the most sensitive dicot (lettuce) was 0.0057 lb ai/A. In terms of vegetative vigor, monocots appeared more sensitive than dicots, with a monocot (ryegrass) $EC_{25}$ of 0.021 lb ai/A, and dicot (cucumber) $EC_{25}$ of 0.27 lb ai/A. The seedling emergence NOAEC for the most sensitive monocot (ryegrass) was 0.001 lb ai/A, and the NOAEC for the most sensitive dicot (lettuce) was 0.0003 lb ai/A. Vegetative vigor NOAEC endpoints for monocots (ryegrass, 0.011 lb ai/A) and dicots (cucumber, 0.01 lb ai/A) were essentially the same.

Three plant studies evaluating effects of metolachlor and *s*-metolachlor on non-crop plant species were available in ECOTOX. Generally, these studies were conducted on mature and/or growing plants, rather than pre-emergence, thus they are more comparable to the vegetative vigor endpoints than the seedling emergence endpoints from the guideline studies. Open literature studies on crop species produced less sensitive endpoints than the registrant-submitted studies. Ecological effects data are located in Appendix B, Table 7. Plants have been grouped into two classes: herbaceous (grasses and forbs) and woody (trees and shrubs). This classification is intended to reflect both a difference in ecological function, and expected differences in sensitivity to the herbicide. In order to establish upper and lower bounds, the most sensitive and the least sensitive endpoints for each group are included in the table. For the grasses and forbs, a test concentration of 0.11 lb ai/A was applied (ECOTOX# 73233). At this concentration, results ranged from no observed effect (broomcorn) to a 90% reduction in height (barnyard grass). The most sensitive species tested in the trees and shrubs class was the Tatarian maple, which exhibited reduced growth at an application of 3.0 lb ai/A (ECOTOX# 73251). The least sensitive species tested was the European white birch, which had no observable effects at an application rate of 9.1 lb ai/A.

In a natural landscape, plants most at risk from use of metolachlor would be newly emerging plants located near the use site. Based on studies, metolachlor is absorbed by plants mostly at the roots and shoots (Zimdahl 1993, www.syngentacropprotection-us.com/prod/herbicide/dualiimagnum), thus the most effective route of exposure is when metolachlor is incorporated into or deposited onto bare soil, where it may be taken up by the growing plant (represented by the seedling emergence guideline tests). However, it is also effective against mature and growing herbaceous plants (represented by the guideline vegetative vigor tests, and most of the open literature studies) at environmentally relevant concentrations ($EC_{25}$ from 0.02-0.11 lb ai/A)
No data were located evaluating the effects of metolachlor on plants which reproduce from cut stumps and/or root propagation.

Vegetative vigor and seedling emergence guideline tests were also available for both the ESA degradate and the OA degradate. Both are less toxic than the parent compound. The Tier I studies submitted (MRIDs 44931718, 44929513) evaluated the effects of the degradates at an application rate of 0.5 lb ai/A. With the exception of the monocot seedling emergence endpoint for ESA, the $EC_{25}$s for all endpoints were greater than the amount applied. No definitive endpoint was established for monocot seedling emergence and ESA, as the NOAEC was below the concentration tested.

## *4.4 Use of Probit Slope Response Relationship*

Generally, available toxicity data provide an $LC_{50}$ or an $EC_{50}$, (the concentration at which 50% of the test populatin exhibits the designated endpoint, usually mortality). Because the Endangered Species Act (ESA) requires determination of potential effects at an individual level, this information must be extrapolated from existing data. The Agency uses the probit dose response relationship as a tool for deriving the probability of effects on a single individual (U.S. EPA, 2004). The individual effects probability associated with the acute RQ is based on the mean estimate of the probit dose response slope and an assumption of that probit model is appropriate for the data set. In some cases, probit is not the appropriate model for the data, and EFED has low confidence in extrapolations from these types of data sets. Upper and lower bound estimates of the effects probability are also provided. The upper and lower bounds of the effects probability are based on available information on the 95% confidence interval of the slope. 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). Probability of individual effects for the various assessment endpoints is provided below in Table 19.

**Table 19  Probability of Individual Effects**

| Assessment Endpoint | Surrogate Species | $LC_{50}$/ $LD_{50}$ and Slope | Fits Probit | Chance of Individual Effect |
|---|---|---|---|---|
| *Aquatic Phase<br>(Eggs, larvae, tadpoles, juveniles and adults)* | | | | |
| *Direct Effects* | | | | |
| Acute Toxicity to Frog | Bluegill sunfish | 3.2 mg/L and 2.1 (lower bound)<br>3.2 mg/L and 14.8 (slope)<br>3.2 and 27.5 (upper bound) | Yes | 1 in 318<br><1 in $10^{16}$<br><1 in $10^{16}$ |
| Chronic Toxicity to Frog | Fathead minnow | Evaluated based on no effects level,<br>chance of effects evaluation not required | | |
| *Indirect Effects (Prey Reduction)* | | | | |
| Acute Toxicity to Prey | Water flea | 1.1 mg/L and 4.5 (default slope) | Unknown (raw data not available to calculate) | 1 in $4.2 \times 10^{8}$ |
| | Bluegill sunfish | 3.2 mg/L and 2.1 (lower bound)<br>3.2 mg/L and 14.8 (slope)<br>3.2 mg/L and 27.5 (upper bound) | Yes | 1 in 318<br><1 in $10^{16}$<br><1 in $10^{16}$ |
| Chronic Toxicity to Prey | Water flea | Evaluated based on no effects level,<br>chance of effects evaluation not required | | |
| | Fathead minnow | | | |
| *Indirect Effects (Habitat Modification)* | | | | |
| Acute Toxicity to Aquatic Plants | Chance of effects evaluation not required | | | |
| Acute Toxicity to Terrestrial Plants | | | | |
| *Terrestrial Phase<br>(Juveniles and adults)* | | | | |
| Acute Toxicity to Frog | Bobwhite quail | Data set only produced a ">" value and probability of individual effects cannot be determined | | |
| Chronic Toxicity to Frog | Bobwhite quail | Evaluated based on no effects level,<br>chance of effects evaluation not required | | |
| Acute Toxicity to Prey | Honey bee | Data set only produced a ">" value and probability of individual effects cannot be determined | | |
| | Bobwhite quail | | | |
| | Rat | 2514 mg/kg bw and 2.8 (lower bound)<br>2514 mg/kg bw and 6.3 (slope)<br>2514 mg/kg and 9.8 (upper bound) | Yes | 1 in 404<br>1 in $8.2 \times 10^{9}$<br><1 in $10^{16}$ |
| | Bobwhite quail | Evaluated based on no effects level,<br>chance of effects evaluation not required | | |
| Acute Toxicity to Terrestrial Plants | Chance of effects evaluation not required | | | |

## *4.5 Incident Database Review*

The incident database contains a total of 171 reports for metolachlor. Of the reports, 150 are of plant damage mostly to agricultural crops under registered use conditions. The most commonly reported crop damage was to corn, peanuts, and soybeans. There was one reported bird kill that was rated as unlikely to be associated with metolachlor use. There are 19 reported incidents of effects on aquatic animals, primarily fish. Generally, these occurred under registered use conditions, and were rated as possibly or unlikely to be associated with the application of metolachlor. One incident, a fish kill in Minnesota, has a certainty rating of highly probable, but was also listed as accidental misuse.

Incidents are reported separately for *s*-metolachlor, but the number and type of reports are similar. There were atotal of 117 reported incidents for *s*-metolachlor. Of these, only two reports are for organisms other than plants. In one case, there is a report of three birds dying as a result of S-metolachlor use. The certainty of this incident was unrated, and legality designated as unknown. The second case was a reported fish kill of an unspecified magnitude. The legality of the use was designated unknown, and the incident was designated unlikely to be the result of the pesticide use. The remainder was damage to agricultural crops. Based on the data, it appears that most of the reports are undesired effects on the at the treatment site, when applied in accordance with registered use. The most commonly reported damaged crops were corn, cotton, and soybean. The certainty that these incidents were related to metolachlor use was generally rated as possible.

## 5.0 Risk Characterization

## 5.1 Risk Estimation

Risk is estimated by calculating the ratio of the expected environmental concentration and the appropriate toxicity endpoint. This value is the risk quotient (RQ), which is then compared to pre-established levels of concern (LOC) for each category evaluated. The RQ methodology, LOCs , and specific details of the calculations are contained in Appendix G. The highest EECs and most sensitive endpoints are used to determine the screening level RQ. Using these two values theoretically results in a conservative estimate of risk. Upper and lower bound risk quotients are presented in Table 20 (aquatic phase) and Table 21 (terrestrial phase).

**Table 20 Metolachlor Risk Quotients for Aquatic Phase**

| Assessment Endpoint | Organism or Life Stage | Concentration Estimate | RQ | LOC Exceedence[1] |
|---|---|---|---|---|
| *Aquatic Phase (Eggs, larvae, tadpoles, juvenile, and adults)[a]* | | | | |
| *Direct Effects* | | | | |
| Acute Toxicity to Frog | Juveniles, adults | Sorghum[2] (highest)<br>Cotton[3] (lowest) | <0.05<br><0.05 | No<br>No |
| Chronic Toxicity to Frog | Eggs, larvae, tadpole | Sorghum[2] (highest)<br>Cotton[3] (lowest) | <1.0<br><1.0 | No<br>No |
| *Indirect Effects and Critical Habitat Effects* | | | | |
| Acute Toxicity to Prey | Fish | Sorghum[2] (highest)<br>Cotton[3] (lowest) | <0.05<br><0.05 | No<br>No |
| | Invertebrate | Sorghum[2] (highest)<br>Cotton[3] (lowest) | <0.05<br><0.05 | No<br>No |
| Chronic Toxicity to Prey | Fish | Sorghum[2] (highest)<br>Cotton[3] (lowest) | <1.0<br><1.0 | No<br>No |
| | Invertebrate | Sorghum[2] (highest)<br>Cotton[3] (lowest) | 13.5<br>2.6 | *Yes*<br>*Yes* |
| Acute Toxicity to Aquatic Plants (Habitat, Food Source) | Duckweed | Sorghum[2] (highest)<br>Cotton[3] (lowest) | 1.0<br><1.0 | *Yes*<br>No |
| | Green algae | Sorghum[2] (highest)<br>Cotton[3] (lowest) | 2.7<br><1.0 | *Yes*<br>No |
| Acute Toxicity to Terrestrial Plants (Wetland) | Monocot | Sorghum[2] (highest)<br>Pod crops (lowest) | 172<br>60.9 | *Yes*<br>*Yes* |
| | Dicot | Sorghum[2] (highest)<br>Pod crops[4] (lowest) | 145<br>51.3 | *Yes*<br>*Yes* |
| Acute Toxicity to Terrestrial Plants (Upland) | Monocot | Sorghum[2] (highest)<br>Cotton[4] (lowest) | 31.3<br>20.8 | *Yes*<br>*Yes* |
| | Dicot | Sorghum[2] (highest)<br>Cotton[4] (lowest) | 26.3<br>17.54 | *Yes*<br>*Yes* |

[1] LOCs used in this assessment:
- Aquatic animals acute risk endangered species 0.05
- Aquatic animals chronic risk 1.0
- Aquatic plants acute risk 1.0.

[2] Sorghum preplant surface applied aerial
[3] Cotton post emergent ground
[4] Cotton pre emergent ground

**Table 21 Metolachlor Risk Quotients for Terrestrial Phase**

| Assessment Endpoint | Organism or Life Stage | Concentration Estimate | RQ | LOC Exceedence[1] |
|---|---|---|---|---|
| *Terrestrial Phase (Juveniles and adults)* | | | | |
| *Direct Effects* | | | | |
| Acute Toxicity to Frog | Juvenile (20g) | Potatoes (highest)<br>Cotton[4] (lowest) | 0.17[2]<br>0.11[2] | *Yes*<br>*Yes* |
| | Adult (100 g) | Potatoes (highest)<br>Cotton[4] (lowest) | <0.1<br><0.1 | No<br>No |
| Chronic Toxicity to Frog | All sizes | Potatoes (highest)<br>Cotton[4] (lowest) | <1.0<br><1.0 | No<br>No |
| *Indirect Effects and Critical Habitat Effects* | | | | |
| AcuteToxicity to Prey | Terrestrial Invertebrate | Potatoes (highest)<br>Cotton[4] (lowest) | 0.17[2]<br>0.09[2] | *Yes*<br>*Yes* |
| | Mouse (15 g herbivore or granivore) | Potatoes (highest)<br>Cotton[4] (lowest) | <0.1<br><0.1 | No<br>No |
| | Mouse (35 g herbivore or granivore) | Potatoes (highest)<br>Cotton[4] (lowest) | <0.1<br><0.1 | No<br>No |
| | Frog (20 g) | Potatoes (highest)<br>Cotton[4] (lowest) | 0.17[2]<br>0.11[2] | *Yes*<br>*Yes* |
| | Frog (100 g) | Potatoes (highest)<br>Cotton[4] (lowest) | <1.0<br><1.0 | No<br>No |
| Chronic Toxicity to Prey | Terrestrial Invertebrate | No data for chronic evaluation | | |
| | Mouse (herbivore or granivore all sizes) | Potatoes (highest)<br>Cotton[4] (lowest) | <1.0<br><1.0 | No<br>No |
| | Frog (all sizes) | Potatoes (highest)<br>Cotton[4] (lowest) | <1.0<br><1.0 | No<br>No |
| Acute Toxicity to Terrestrial Plants (Wetland) | Monocot | Sorghum[3] (highest)<br>Pod crops (lowest) | 172<br>60.9 | *Yes*<br>*Yes* |
| | Dicot | Sorghum[3] (highest)<br>Pod crops (lowest) | 145<br>51.3 | *Yes*<br>*Yes* |
| Acute Toxicity to Terrestrial Plants (Upland) | Monocot | Sorghum[3] (highest)<br>Cotton[4] (lowest) | 31.3<br>20.8 | *Yes*<br>*Yes* |
| | Dicot | Sorghum[3] (highest)<br>Cotton[4] (lowest) | 26.3<br>17.54 | *Yes*<br>*Yes* |

[1] LOCs used in this assessment:
- Terrestrial plants acute risk 1.0
- Terrestrial vertebrates acute risk endangered species 0.1
- Terrestrial invertebrates acute risk endangered species 0.05

[2] Toxicity tests used to evaluate the frog and terrestrial insect did not establish a definitive endpoint (*i.e.,* the value was greater than the highest concentration tested), thus these RQ values represent an upper bound

[3] Sorghum preplant surface applied aerial

[4] Cotton pre emergent surface applied

## *5.2 Risk Description*

### *5.2.1 Direct Effects*

#### *5.2.1.1 Aquatic Phase*

The aquatic phase considers life stages of the frog that are obligatory aquatic organisms, including eggs, larvae, and tadpoles. It also considers juveniles and adults, which spend a portion of their time in water bodies which may receive runoff containing metolachlor. There were no acute risk endangered species or chronic risk LOC exceedences for any life stages of the California red-legged frog based on aquatic exposure, thus no direct effects on the aquatic phase of the frog are anticipated. This results in a determination of no effect for this component of the assessment.

#### *5.2.1.2 Terrestrial Phase Adults*

For this ecological risk assessment, terrestrial phase adults are defined as frogs weighing 100g or more, based on the evaluation categories available in the T-REX model. Based on T-REX calculations, no acute risk endangered species LOCs were exceeded for the adult frogs for any application rate. There were no chronic risk exceedences for any size frog for any application rate. No direct effects on the frog are anticipated for either of these components of the assessment. This results in a determination of no effect for this component of the assessment.

#### *5.2.1.3 Terrestrial Phase Juveniles*

LOCs for juvenile frogs (20g) exceeded the endangered species acute risk LOCs for all application rates >1.0 lb ai/A based on the screening level estimate (T-REX). It is important to note the toxicity endpoint used in the evaluation was not definitive. The most sensitive test established that the $LC_{50}$ and $LD_{50}$ were both greater than the highest concentration tested (4912 mg/kg bw and 2510 mg/kg bw, respectively. Thus, the RQs calculated based on these endpoints are an upper bound estimate. RQs for a definitive endpoint would be lower, but how much lower cannot be determined from this study. No treatment related mortality was reported in any of the studies. Based on the concentrations tested, metolachlor is classified as practically non-toxic to birds, and thus, using birds as a surrogate for terrestrial phase amphibians, as practically non-toxic to terrestrial phase amphibians.

However, because the surrogate for juvenile frogs exceeded the LOCs for all application rates >1.0 lb ai/A using a dose-based estimate, two additional evaluation methods were used to better evaluate potential risk.

One method was use of T-HERPS, a modification of T-REX which includes amphibian/reptile specific allometric equations, weight classes appropriate for the CLRF, and prey items specific to the CLRF. It is important to note that while the allometric equations and prey items are more specific to the frog, the toxicity data used in this assessment are that for a surrogate species (bobwhite quail). It is unknown what direction use of the surrogate toxicity data might bias the estimate is unknown. T-HERP groups the frogs into three classes: small (1.4g), medium (37g), and large (238g). The two smaller weight classes most closely approximate the 20g juvenile that exceeded LOCs using the T-REX model. Based on T-HERPS, the two smaller weight classes do not exceed the endangered species acute risk LOCs for any food group. The chronic LOC is exceeded for the short grass food group (RQ = 1.19). However, it is unlikely this food group is a significant component of the juvenile diet. CLRFs in the terrestrial phase are primarily carnivores, thus they would be unlikely to ingest short grass.

The second method was to evaluate how far away from the use site juvenile frogs might be able to consume contaminated food items. To evaluate this, T-REX was first used to determine the application rate at which the LOC was cleared for all food items for the juvenile frog (20g). The clearance application rate was 0.62 lb ai/A. To determine how far away from the use site this "application rate" could occur for each crop, AgDrift was used to estimate the deposition. The AgDrift model was parameterized using fractions of the application collected on deposition cards, which would most closely approximate the "short grass" category. For the highest application rate of 2.0 lb ai/A (potatoes), off-site deposition dropped below 0.62 lb ai/A at a distance of 20 ft from the use site. For all other application rates, it fell below the clearance application rate at a distance of 10 ft from the use site. When estimating clearance distance, an important consideration is the foraging distance of the organism (T-REX is based on the assumption that the animal evaluated forages exclusively in the treated area). Thus, only CLRFs foraging exclusively within 20 feet of the treatment site would be at risk. EFED recognizes the potential for off-site movement of the pesticide via biological vectors (*i.e.*, the residue deposited on or accumulated in the body of an animal leaving the field that is then consumed by the frog), however at this time there is no standard method to evaluate it. EFED anticipates biological vectors will not be an important exposure pathway for metolachlor because it is not bioaccumlative, a slow-acting poision, or potentially more toxic to a predator consuming the contaminated organism.

Considering the facts that the toxicity endpoints are an upper bound estimate, refined analysis suggests that CLRFs are unlikely to receive a toxic dose, and drift analysis indicates effects would likely be confined to the use site plus a 20 foot drift zone, direct effects from metolachlor to the terrestrial phase juveniles appears unlikely. The evaluation of the direct effects on terrestrial phase CLRF results in a determination of may affect, not likely to adversely affect (discountable).

### *5.2.2 Indirect Effects and Critical Habitat Effects (Reduction in Prey Base)*

#### *5.2.2.1 Terrestrial Invertebrates*

LOCs for terrestrial invertebrates exceeded the endangered species acute risk LOCs for all application rates based on the screening level estimate. It is important to consider that the toxicity endpoint used in the evaluation was not definitive. The most sensitive test established that the acute contact $LD_{50}$ was greater than the highest concentration tested (200 μg ai/bee). How much higher the endpoint is cannot be determined from the study. No other terrestrial invertebrate data were available.

RQs for terrestrial invertebrates exceeded the LOC for all application rates. In order to determine how far from the use site an insect would have to be before the RQ dropped below the LOC, a clearance distance was calculated in a fashion similar to that used for the juvenile frogs. The concentration of pesticide on the T-REX category representing the insects (broadleaf plants and small insects) that would be equivalent to the LOC was determined (78.125 ppm). T-REX was then used to determine what application rate would result in this concentration, or one slightly lower. At an application rate of 0.55 lb ai/A, the concentration of metolachlor on broadleaf plants and small insects is estimated to be 74.25 ppm.

To determine how far away from the use site this "application rate" could occur for each crop, AgDrift was used to estimate the deposition. AgDrift was parameterized using fractions of the application collected on deposition cards, which would most closely approximate the short grass category, thus using it to estimate concentrations for broadleaf plants and small insects is expected to be more conservative. For the highest application rate of 2.0 lb ai/A (potatoes), off-site deposition dropped below 0.55 lb ai/A at a distance of 20 ft from the use site. For most other application rates, it fell below the clearance application rate at a distance of 10 ft from the use site. For the lowest application rate of 1.0 lb ai/A (potatoes), off-site deposition dropped below 0.55 lb ai/A at a distance of <10 ft from the use site.

Based on the data and risk estimation methods available, risk to terrestrial invertebrates appears unlikely. However, this conclusion must be considered in light of the fact that very little data are available to represent a vast, diverse, and ecologically important taxon. The conclusion is consistent with other toxicity data that shows metolachlor to be primarily toxic to plants. While future ecotoxicity studies could affect the conclusion, based on the best available information at the time of the assessment no effects on terrestrial invertebrates are expected. The evaluation of the potential effects on terrestrial invertebrates results in a determination of may affect, not likely to adversely affect (discountable).

#### *5.2.2.2 Small Mammals*

No endangered species acute risk LOCs were exceeded for small mammals such as mice or voles that might be part of the prey base for the CLRF.

#### *5.2.2.3 Terrestrial Phase Amphibians*

Although endangered species acute risk LOCs were exceeded for small amphibians (20g), direct effects from metolachlor on these organisms appears unlikely. For a more detailed discussion, see the discussion in Section 5.2.1.3.

#### *5.2.2.4 Aquatic Invertebrates*

No endangered species acute risk LOCs were exceeded for aquatic invertebrates. Chronic risk LOCs (based on the NOAEC) were exceeded for all application rates. This toxicity study used *Ceriodaphnia dubia*, one of the most sensitive aquatic invertebrate species.  The lowest test concentration was 0.001mg/L, (the NOAEC), and the next test concentration (the LOAEC) was 0.010 mg/L, thus effects would be expected to occur somewhere within this range.  Based on this particular study, it cannot be determined if the effects occur closer to the NOAEC or the LOAEC.

Data from other studies generally show effects at higher concentrations.  Another study (ECOTOX# 13689) determined a chronic reproductive NOAEC of 6.3 mg/L for *Ceriodaphnia dubia* ,which is 3 orders of magnitude higher than the study used to calculate the risk quotients.  The registrant submitted guideline study for *Daphnia magna* resulted in a chronic reproductive NOAEC of 3.2 mg/L (MRID 43802601) .  Behavioral studies regarding the olfaction response of the rusty crayfish produced a LOAEL of 0.025 mg/L (ECOTOX# 68515).  Modeled peak concentrations of metolachlor ranged from 0.0027 mg/L to 0.0135 μg/L.  Maximum reported concentrations for monitoring sites in California ranged from 0.0014 mg/L to 0.0039 mg/L.

Based on the full toxicity data set and both the modeled and monitored concentrations of metolachlor in surface water, it appears unlikely that aquatic invertebrates will be measurably affected by currently registered uses of metolachlor.

##### *5.2.2.5 Fish*

No endangered species acute risk LOCs or chronic LOCs were exceeded for fish.  No indirect effects are anticipated based on this component of the ecological risk assessment.

### *5.2.3 Indirect Effects and Critical Habitat Effects (Habitat Degradation)*

#### *5.2.3.1 Aquatic Plants (Vascular and Non-vascular)*

Aquatic plants serve several important functions.  They are primary producers, and provide the autochthonous energy base for the aquatic system, especially the non-vascular plants.  Typically, vascular plants provide structure to the system rather than energy, providing attachment sites for many aquatic invertebrates, and refugia for juvenile organisms, such as fish and frogs.  Emergent plants help reduce sediment loading, and provide some stability to nearshore areas and lower streambanks.  For the CLRF, vascular aquatic plants provide an attachment site for egg masses.

Presence of herbicides in the water bodies supporting the CLRF could reduce populations of sensitive non-vascular plants, and/or cause a shift in phytoplankton community dynamics.  Generally, green algae are considered a good food source for herbivorous zooplankton, as are diatoms, which are considerably more resistant to metolachlor.  Blue-green algae are typically considered to be less palatable.  These food generalizations may also apply to larval CLRFs.

Typically, algal populations are relatively dynamic, and the presence of herbicides in the water may result in an overall reduction of biomass, and/or a shift in community composition as more sensitive species are eliminated. Herbicides may also modify timing of maximum algal growth. Often, although not always, an algal community will recover when the stressor is removed.

Table 22 shows acute risk quotients for all crops for all taxa of freshwater plants tested, and for the one vascular plant tested. Acute risk LOCs were exceeded for two crops, sorghum (both aerial and ground applications) and soybeans (aerial applications). No other crops exceeded LOCs for any plant evaluated. CDPR PUR data, presented in Section 2.4.3, reports no sorghum usage in 2002-2005. The only other exceedence is for soybeans, when metolachlor is applied aerially. For soybeans, the LOC is exceeded for freshwater algae, but not for any of the other plants shown in Table 22. RQs in Table 22 were calculated based on the most sensitive endpoint for the respective plant types. Reported annual use on bean crops ranged from 72 lbs (2005) to 1,236 lbs (2004), but based on NASS data, soybeans are not grown in California. Aquatic concentrations of metolachlor resulting from use on other crops in the CDPR PUR "bean" category are represented by the pod crops category in Table 22.

**Table 22 Aquatic Plant Risk Quotients for All Crops**

| Crop and Application Method | Peak EEC (mg/L) | RQs | | | | |
|---|---|---|---|---|---|---|
| | | Non-vascular Plants | | | Vascular Plants | LOC Exceedence |
| | | FW Alga | FW Diatom | Blue-green Alga | Duckweed | |
| *Aerial Applications* | | | | | | |
| Corn | 0.0078 | <1 | <1 | <1 | <1 | No |
| Cotton (Post emergent) | 0.0046 | <1 | <1 | <1 | <1 | No |
| Pod Crops | 0.0060 | <1 | <1 | <1 | <1 | No |
| Potatoes | 0.0056 | <1 | <1 | <1 | <1 | No |
| Safflower | 0.0049 | <1 | <1 | <1 | <1 | No |
| Sorghum (Surface applied) | 0.0218 | 2.7 | <1 | <1 | 1.0 | *Yes* |
| Soybean | 0.0093 | 1.2 | <1 | <1 | <1 | *Yes* |
| *Ground Applications* | | | | | | |
| Corn | 0.0046 | <1 | <1 | <1 | <1 | No |
| Cotton (Post emergent) | 0.0011 | <1 | <1 | <1 | <1 | No |
| Pod Crops | 0.0041 | <1 | <1 | <1 | <1 | No |
| Potatoes | 0.0021 | <1 | <1 | <1 | <1 | No |
| Safflower | 0.0011 | <1 | <1 | <1 | <1 | No |
| Sorghum[a] (Surface applied) | 0.0194 | 2.4 | <1 | <1 | <1 | *Yes* |
| Soybean[b] | 0.0077 | <1 | <1 | <1 | <1 | No |

[a] No reported usage of metolachlor on sorghum in 2002-2005 CDPR PUR data
[b] Not grown in California

Based on modeled concentrations for various crops and concentrations, concentrations of metolachlor in aquatic systems near use sites could be high enough to affect sensitive algal species. Based on the distribution of modeled concentrations, monitoring data, and usage data, effects on sensitive species are not anticipated to occur frequently. While the possibility of changes in the algal community exists, and these changes could potentially affect the CLRF, the changes are not expected to measurably affect the viability of CLRF individuals. This results in a determination of may affect, not likely to adversely affect (discountable) for this component of the assessment.

#### *5.2.3.2 Terrestrial Plants*

Terrestrial plants serve several important functions in the California red-legged frog habitat. Vegetation provides cover from predators while the frog is foraging as well as providing habitat and food sources for both its invertebrate and vertebrate prey. Upland vegetation provides cover during dispersal and riparian vegetation helps to maintain the integrity of aquatic systems by providing bank stability and, in some cases, allochthonous input.

Loss, destruction, and alteration of habitat were identified as a threat in the CLRF Recovery Plan (USFWS 2002). Herbicides can affect habitat in a number of ways. The first, and most extreme, is that herbicide deposition could kill all or a substantial amount of the vegetation in the area, thus removing the structure which defines the habitat, and reducing the functions (*e.g.,* cover, food supply for prey base) provided by the vegetation.

Metolachlor is absorbed through the roots and the shoot of the plant, and is most efficacious when applied to the soil from which the plant absorbs it. This is demonstrated by the difference in response to the two guideline studies. The $EC_{25}$ for seedling emergence tests is 0.005 lb ai/A for monocots and 0.006 lb ai/A for dicots. For the vegetative vigor tests (more correlative to what would occur if metolachlor was deposited on a plant that was actively growing, as opposed to one that had just emerged) the $EC_{25}$ is 0.02 lb ai/A for monocots and 0.27 lb ai/A for dicots, a difference of an order of magnitude.

In a healthy riparian system, there is often a three-tier vegetation system, with trees as an overstory, shrubs as an understory, and grasses and forbs forming the ground cover. Some evaluations show the CLRF occupies waterbodies with dense riparian vegetation such as willows (*Salix spp.)* (Hayes and Jennings 1988). Upland habitat often includes scrub and shrub (USFWS 2002). While no guideline data are available for trees and shrubs, open literature data in ECOTOX indicates these woody species are far less sensitive to metolahclor, with effects noted in the 3.0-9.1 lb ai/A range. It is reasonable to presume that the shrub species in both types of habitats will intercept some of the metolachlor which might otherwise be deposited on the more sensitive herbaceous species. Additionally, in a natural system, senescent plants, fallen leaves, and other debris often provide a litter layer which might also serve to protect newly emerging herbaceous plants. Areas of bare soil in the CRLF habitat are expected to be relatively small in comparision to the total habitat area. Thus, effects in a natural system are likely to be more closely approximated by the vegetative vigor endpoints than the seedling emergence endpoints.

Table 23 shows clearance distances, where the RQ drops below the LOC, for all combinations of application rates, guideline endpoints, and application methods (ground versus aerial). Clearance distances for the more sensitive seedling emergence endpoints were used to establish the action area (2,060 feet from NLCD-classified agricultural land use areas). However, EFED anticipates adverse effects that could reasonably be measured would be defined by the vegetative vigor endpoint. Thus, for ground-applied metolachlor, adverse effects might reasonably be expected to occur up to 90 feet from the use site, and for aerially-applied metolachlor, adverse effects might reasonably be expected to occur up to 1000 feet from the use site for aerial applications and up to 90 feet for ground applications. In some cases, topography (such as an intervening ridge) or weather conditions (such as prevailing winds towards or away from the frog habitat) could affect these estimates. Analysis of site-specific details is beyond the scope of this assessment.

**Table 23 Required Distance from Application to Fall Below LOC**

| Crop | Application Rate (lb ai/A) | Clearance Distance (ft) | | | | |
|---|---|---|---|---|---|---|
| | | Seedling Emergence | | Vegetative Vigor | | Tree/Shrub[3] |
| | | Monocot | Dicot | Monocot | Dicot | |
| *Ground applications*[1] | | | | | | |
| Corn | 1.63 | 400 | 350 | 80 | 10 | 0 |
| Cotton | 1.33 | 300 | 250 | 60 | 10 | 0 |
| | 1.00 | 250 | 200 | 50 | 10 | 0 |
| Pod Crops | 1.67 | 400 | 350 | 80 | 10 | 0 |
| Potatoes | 2.00 | 450 | 400 | 90 | 10 | 0 |
| Safflower | 1.67 | 400 | 350 | 80 | 10 | 0 |
| Sorghum | 1.50 | 350 | 300 | 70 | 10 | 0 |
| Soybeans | 1.67 | 400 | 350 | 80 | 10 | 0 |
| *Aerial applications*[2] | | | | | | |
| Corn | 1.63 | <900 | <900 | 900 | 100 | 0 |
| Cotton | 1.33 | <900 | <900 | 700 | 100 | 0 |
| | 1.00 | <900 | <900 | 500 | 100 | 0 |
| Pod Crops | 1.67 | <900 | <900 | 900 | 100 | 0 |
| Potatoes | 2.00 | <900 | <900 | >900[4] | 100 | 0 |
| Safflower | 1.67 | <900 | <900 | 900 | 100 | 0 |
| Sorghum | 1.50 | <900 | <900 | 800 | 100 | 0 |
| Soybeans | 1.67 | <900 | <900 | 900 | 100 | 0 |

[1] Based on AgDrift estimate, low boom, 90% percentile, to the nearest 10ft if <100ft, to the nearest 50ft if >100 ft
[2] Based on AgDrift estimate, aerial 90% percentile, to the nearest 100 ft
[3] Based on most sensitive endpoint for tree/shrub type plant (Tartarian maple LOAEC, 3.0 lb ai/A, ECOTOX 73251)
[4] Approximately equal to LOC at 990 ft, limit of AgDrift.

## *5.3 Risk Conclusions*

After completing the analysis of the effects of metolachlor on the federally listed threatened California red-legged frog *(Rana aurora draytonii)* in accordance with methods delineated in the Overview document (USEPA 2004), EFED concludes that the use of metolachlor (PC#108801) may affect, and is likely to adversely affect the California red-legged, based on indirect effects (habitat modification to terrestrial plants). EFED also concludes that these same effects constitute adverse modification to critical habitat. These effects are anticipated to occur only for those occupied core habitat areas, CNDDB occurrence sections, and designated critical habitat for the California red-legged frog that are located ≤ 1000 feet from legal use sites where metolachlor is application aerially, and ≤ 90 feet where metolachlor is applied with ground-based equipment. For the purpose of this assessment, legal use sites are defined as NLCD-classified agricultural lands. Potential but not anticipated indirect effects include reduction of the prey base and/or reduction of primary productivity in waters receiving runoff from fields treated with metolachlor. Rationale for each component assessed is provided in Table 24.

Using ARGIS9, the NLCD classified data, and CLRF habitat information supplied by the U.S. FWS, EFED has identified the habitat areas where indirect effects are anticipated to occur (Figure 16) and designated critical habitat areas where adverse modifications are anticipated to occur (Figure 17). Indirect effects (modification of the terrestrial plant community) could potentially occur in approximately 9% (~620,000 acres) of the CLRF range assessed, and adverse modification to designated critical habitat could potentially occur in 0.003% (~14 acres) of the currently designated area. Specific core areas, and designated critical habitat units which could be adversely affected by use of metolachlor are listed in Table 25. Table 25 also lists the counties in which the units occur. In some cases, core areas and/or critical habitat units may be located in more than one county or recovery unit, and will be listed in both.

**Table 24 Effects Determination for Metolachlor**

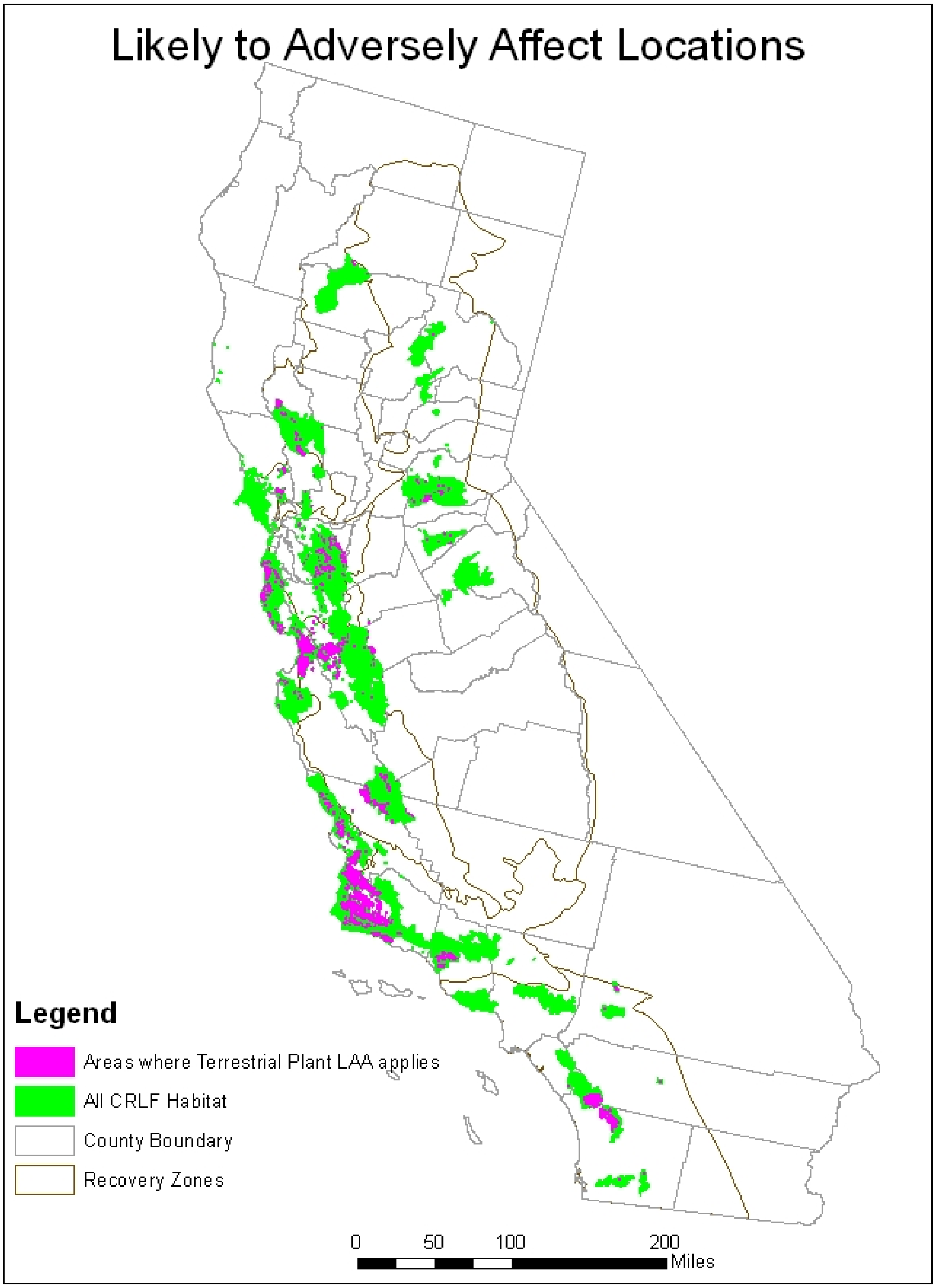
| Assessment Endpoint | Effects determination | Basis for Determination |
|---|---|---|
| *Aquatic Phase (Eggs, larvae, tadpoles, juveniles, and adults)*[a] | | |
| *Direct Effects* | | |
| 1. Survival, growth, and reproduction of CRLF | No effect | No LOC exceedences for any life stage |
| *Indirect Effects* | | |
| 2. Reduction or modification of aquatic prey base | May affect<br>Not likely to adversely affect<br>(Discountable) | Chronic exceedences for aquatic invertebrates. No LOC exceedences for any other aquatic prey items. Based on analysis of full toxicity data set, monitoring data, and modeled EECs, chronic effects on aquatic invertebrates appear unlikely. |
| 3. Reduction or modification of aquatic plant community | May affect<br>Not likely to adversely affect<br>(Discountable) | Exceedences for both vascular and non-vascular plants for sorghum, and for non-vascular plants for soybeans. CDPR PUR data report no usage on sorghum, and soybeans are not grown in CA. |
| 4. Degradation of riparian vegetation | May affect<br>NLAA >1000 ft<br>LAA <1000ft (aerial)<br>LAA <90ft (ground)<br>Modification to critical habitat<br>Adverse <1000ft (aerial)<br>Adverse <90ft (ground) | Exceedences for both monocots and dicots in both wetlands and uplands adjacent to use site for all crops registered.<br><br>Exceedences for both monocots and dicots near use site based on spray drift alone for all crops registered. |
| *Terrestrial Phase (Juveniles and Adults)* | | |
| *Direct Effects* | | |
| 5. Survival, growth, and reproduction of CRLF | May affect<br>Not likely to adversely affect<br>(Discountable) | Screening level LOC exceedences for juveniles. Effects may be overestimated by existing toxicity data and are unlikely to occur >20 ft from use site.<br><br>No acute exceedences for adults<br>No chronic exceedences for juveniles or adults. |
| *Indirect Effects and Critical Habitat Effects* | | |
| 6. Reduction or modification of terrestrial prey base | May affect<br>Not likely to adversely affect<br>(Discountable) | Screening level LOC exceedences for terrestrial invertebrates. Effects may be overestimated by existing toxicity data and are unlikely to occur >20 ft from use site.<br><br>No exceedences for mammal or amphibian prey |
| 7. Degradation of riparian and/or upland vegetation | May affect<br>NLAA >1000 ft<br>LAA <1000ft (aerial)<br>LAA <90ft (ground)<br>Modification to critical habitat<br>Adverse <1000ft (aerial)<br>Adverse <90ft (ground) | Exceedences for both monocots and dicots in both wetlands and uplands adjacent to use site for all crops registered.<br><br>Exceedences for both monocots and dicots near use site based on spray drift alone for all crops registered. |

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.

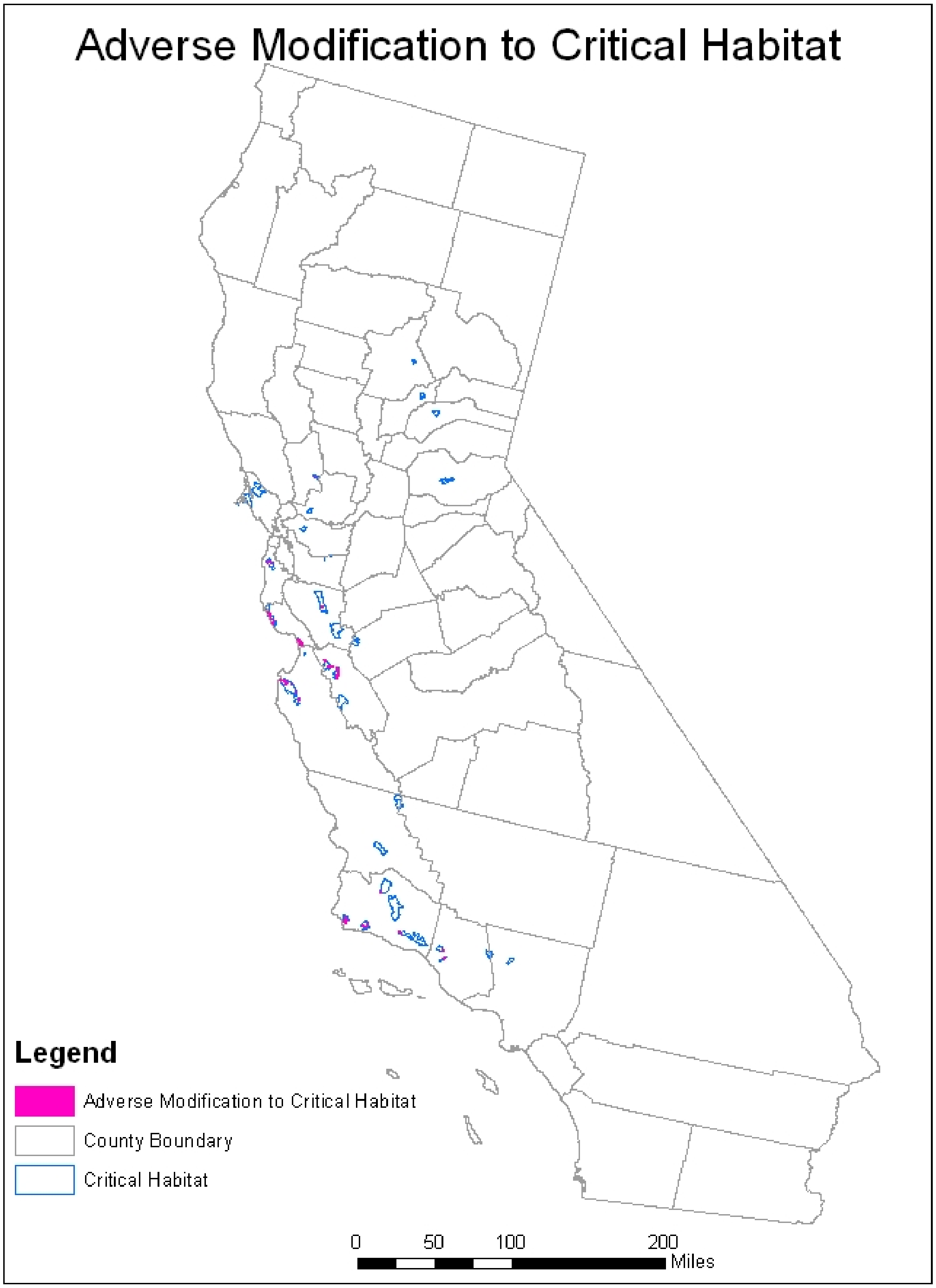
- Information on population responses of prey base organisms to the pesticide. Currently, methodologies are limited to predicting exposures and likely levels of direct mortality, growth or reproductive impairment immediately following exposure to the pesticide. The degree to which repeated exposure events 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 adverse modification to critical habitat.




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

Map created by U.S. Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. June 7, 2007 (PDB).
Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983)

**Figure 16 Locations where Metolachlor Use is Likely to Adversely Affect the CLRF**




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

Map created by U.S. Environmental Protection Agency, Office of Pesticides Programs, Environmental Fate and Effects Division. June 7, 2007 (PDB). Projection: Albers Equal Area Conic USGS, North American Datum of 1983 (NAD 1983)

**Figure 17 Locations where Metolachlor Use could Adversely Modify CLRF Critical Habitat**

**Table 25  Areas of Possible Adverse Effects and Adverse Modification by Recovery Unit**

| Recovery Unit | Likely to Adversely Affect Core Area | Adverse Modification Critical Habitat Unit | County [b] |
|---|---|---|---|
| 1 Sierra Nevada Foothills and Central Valley | 1 Feather River<br>2 Consumnes River<br>5 South Fork Calaveras River<br>6 Tuolumne River | None | Amador<br>Calaveras<br>El Dorado<br>Plumas<br>Tuolumne |
| 2 North Coast Range Foothills and Western Sacramento River Valley | 8 Cottonwood Creek<br>9 Putah Creek-Cache Creek[a] | None | Shasta<br>Lake<br>Napa<br>Yolo |
| 3 North Coast and North San Francisco Bay | 9 Putah Creek-Cache Creek[a]<br>10 Lake Berryessa Tribs<br>11 Upper Sonoma Creek<br>12 Petaluma-Sonoma Creek<br>15 Jameson Canyon-Lower Napa River | NAP-1 | Napa<br>Sonoma<br>Solano |
| 4 South and East San Francisco Bay | 16 East San Francisco Bay | SNM-1A<br>STC-1A | Alameda<br>Contra Costa<br>San Joaquin<br>San Mateo<br>Santa Clara<br>Stanislaus |
| 5 Central Coast | 18 South San Francisco River<br>19 Watsonville Slough-Elkhorn Slough[a]<br>20 Carmel River-Santa Lucia<br>22 Estero Bay<br>23 Arroyo Grande Creek | SNM-1A<br>SCZ-1<br>SCZ-2<br>MNT-2 | Monterey<br>San Luis Obispo<br>San Mateo<br>Santa Cruz |
| 6 Diablo Range and Salinas Valley | 16 East San Francsico Bay<br>19 Watsonville Slough-Elkhorn Slough<br>28 Estrella River | STC-1B<br>SNB-1<br>SNB-2 | Kern<br>Merced<br>Monterey<br>San Benito<br>San Luis Obispo<br>Santa Clara |
| 7 Northern Transverse Range and Tehachapi Mountains | 24 Santa Maria River-Sanata Inez River<br>25 Sisquoc River<br>26 Ventura River-Santa Clara River | STB-1<br>STB-3<br>STB-4<br>STB-5<br>STB-7<br>VEN-1<br>VEN-2 | Santa Barbara<br>Ventura |
| 8 Southern Transverse and Peninsular Ranges | 27 Santa Monica Bay-Ventura Coastal<br>30 Forks of the Mojave<br>32 Santa Rosa Plateau<br>33 San Luis Rey<br>34 Sweetwater<br>35 Laguna Mountain | None | Los Angeles<br>Orange<br>Riverside<br>San Diego |

[a] Some core areas are in two recovery units, thus they are listed twice.
[b] Counties listed may contain either affected core areas, designated critical habitat, or both.

## 6.0 Uncertainties

Risk assessment, by its very nature, is not exact, and requires the risk assessor to make assumptions regarding a number of parameters, to use data which may or may not accurately reflects the species of concern, and to use models which are a simplified representation of complex ecological processes. In this risk assessment, EFED has used the best available data regarding such important parameters as the life history of the California red-legged frog, typical environmental conditions in the proximity of frog habitat, toxicity of metolachlor, and usage of metolachlor in the action area. Frequently, such information is better expressed as ranges rather then points, and when this is the case, EFED has opted to use the end of range resulting in a conservative estimate of risk, in order to provide the benefit of doubt to the frog. These uncertainties, and the directions in which they may bias the risk estimate, are described below.

## 6.1 Exposure Assessment Uncertainties

Overall, the uncertainties inherent in the exposure assessment tend to result in over-estimation of exposures. This is apparent when comparing modeling results with monitoring data. In particular, estimated peak exposures are generally an order of magnitude above 90$^{th}$ percentile site concentrations in the surface water monitoring data. In general, the monitoring data should be considered a lower bound on exposure, while modeling represents an upper bound.

### 6.1.1 Modeling Assumptions

The uncertainties incorporate in the exposure assessment cannot be quantitatively characterized. However, given the available data and the EFED's reliance on conservative modeling assumptions, it is expected that the modeling results in an over-prediction of exposure. Qualitatively, conservative assumptions which may affect exposure include the following:

- Modeling for each use site assumes that the entire 10-hectare watershed is taken up by the respective use pattern.
- The assessment assumes all applications have occurred concurrently on the same day at the exact same application rate.
- The assessment assumes all applications are at maximum labeled rate.

### 6.1.2 Maximum Use Scenarios

The screening-level risk assessment focuses on characterizing potential ecological risks resulting from a maximum use scenario, which is determined from label 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 pesticide resistance, timing of applications, cultural practices, and market forces.

### 6.1.4 Modeling Inputs

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).

### 6.1.5 Aquatic Exposure Estimates

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 a farmer's field on a day-to-day basis. It considers factors such as rainfall and plant transpiration of water, as well as how and when the pesticide is applied. It has two major components: hydrology and chemical transport. Water movement is simulated by the use of generalized soil parameters, including field capacity, wilting point, and saturation water content. The chemical transport component can simulate pesticide application on the soil or on the plant foliage. Dissolved, adsorbed, and vapor-phase concentrations in the soil are estimated by simultaneously considering the processes of pesticide uptake by plants, surface runoff, erosion, decay, volatilization, foliar wash-off, advection, dispersion, and retardation.

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

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

#### *6.1.6 Usage Uncertainties*

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

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.

## 6.2 *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 California Red Legged Frog.

### *6.2.2 Use of Surrogate Species Data*

Currently, there are no FIFRA guideline toxicity tests for amphibians. Therefore, in accordance with the Overview Document (U.S. EPA 2004), data for the most sensitive freshwater fish are used as a surrogate for aquatic-phase amphibians such as the California red-legged frog. Available open literature information on metolachlor toxicity to aquatic-phase amphibians (African clawed frog) shows this species approximately is 3 to 4 times less sensitive than the freshwater fish endpoint EFED used in the assessment. Species sensitivity distribution data for amphibians indicates the range of sensitivity is similar to that of freshwater fish (Birge *et al.*, 2000). The African clawed frog appears to be less sensitive than some of the native species. Therefore, the endpoint based on freshwater fish ecotoxicity data is assumed to be protective. Extrapolation of the risk conclusions from the most sensitive tested species to the California red-legged frog is more likely to overestimate the potential risks than to underestimate the potential risk. Information to indicate were the California red-legged frog may fall in a species sensitivity distribution was not located.

### *6.2.3 Extrapolation of Effects*

Length of exposure and concurrent environmental stressors (*e.g,* urban expansion, habitat modification, predators) will likely affect the response of the California red-legged frog to metolachlor. Because of the complexity of an organism’s response to multiple stressors, the overall “direction” of the response is unknown. Additional environmental stressors may decrease or increase the sensitivity to the herbicide. Timing, peak concentration, and duration of exposure are critical in terms of evaluating effects. These factors will vary both temporally and spatially within the action area. Overall, the effect of this variability may result in either an overestimation or underestimation of risk

### 6.2.4 *Acute LOC Assumptions*

The risk characterization section of this assessment includes an evaluation of the potential for individual effects. The individual effects probability associated with the acute RQ is based on the assumption that the dose-response curve fits a probit model. It uses the mean estimate of the slope and the $LC_{50}$ to estimate the probability of individual effects.

### 6.2.5 *Residue Levels Selection*

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.

### 6.2.6 *Dietary Intake*

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.

### *6.2.7 Mixtures*

The California red-legged frog and various components of its ecosystem may be exposed to multiple pesticides, introduced into its environment either via a multiple active ingredient formulated product, a tank mixture, or transport from independently applied active ingredients. Multiple pesticides may act in an additive, synergistic, or antagonistic fashion. Quantifying reasonable environmental exposures and establishing reasonable corresponding toxicological endpoints for the myriad of possible situations is beyond the scope of this document, and in some cases, beyond the current state of ecotoxicological practice. Mixtures could affect the CLRF in ways not addressed in this assessment. Exposure to multiple contaminants could make organisms more or less sensitive to the effects of metolachlor, thus the directional bias associated with environmental mixtures is unknown, and may vary on a case-by-case basis.

### *6.2.8 Sublethal Effects*

For an acute risk assessment, 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 assessment 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.

Sublethal effects, including behavioral effects, have been linked to metolachlor. Where quantitative data existed, these effects were considered in the assessment, and appear to occur at concentrations higher than the frank effects used as assessment endpoints. Thus, based on data available at the time of the assessment, risk conclusions in the assessment are anticipated to be adequately protective in regards to sublethal effects.

### *6.2.9 Location of Wildlife Species*

For this baseline terrestrial 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.

*References*

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

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

Birge, WJ, AG Westerman, and JA Spromberg. (2000) Comparative Toxciology and Risk Assessment of Amphibians, Chap 14 in Ecotoxicology of Amphibians and Reptiles, Sparling, DW, G Linder, and CA Bishop, (Eds.), SETAC Press, Pensacola, FL.

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

Eckermann C, B. Matthes, M. Nimtz, V. Reiser, B. Lederer, P. Boger P, J. Schroder. (2003). Covalent binding of chloroacetamide herbicides to the active site cysteine of plant type III polyketide synthases. *Phytochemistry* **64**(6):1045-54.

Fairchild, JF, LC Sappington, DS Ruessler (1999) An Ecological Risk Assessment for Herbicide Impacts on Primary Productivity of the Lower Missouri River. U.S. Geological Survey Toxic Substances Hydrology Program-Proceedings of the Technical Meeting, Charleston, South Carolinia, March 8-12, 1999. U.S. Geological Survey Water Resources Investigations Report 99-40188, Vol 2 of 3.

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

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

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

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

Fletcher, JS, JE Nelesson, and TG Pfleeger. (1994)Literature Review and Evaluation of the EPA Food-chain (Kenaga) Nomogram , an Instrument for Estimating Pesticide Residues on Plants.  Environmental Toxicology and Chemistry 13 (9) 1383-1391.

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

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

Jablonkai I. (2003).  Alkylating reactivity and herbicidal activity of chloroacetamides. *Pest Manag Sci*. **59**(4):443-50.

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

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

Junghans, M., T. Backhaus, M. Faust, M. Scholze, and L.H. Grimme. (2003). Predictability of combined effects of eight chloroacetanilide herbicides on algal reproduction.  *Pest Manag Sci.* **59**(10):1101-10.

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

Kollman, Wynetta S. 2002. Summary of Bill 1807/3219: Pesticide Air Monitoring Results: Conducted by the California Air Resources Board 1986-2000. California Department of Pesticide Regulation, Sacramento, CA.

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

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

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

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

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

Pillai P., D.E. Davis, and B. Truelove (1979). Effects of metolachlor on germination, growth, leucine uptake, and protein synthesis. *Weed Science* **27**(6):634-637.

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

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

Rossini L., C. Frova, M. E. Pè, L. Mizzi, and M. Sari Gorla. (1998). Alachlor Regulation of Maize Glutathione S-Transferase Genes. *Pesticide Biochemistry and Physiology*. **60**(3):205-211.

Segawa, R., Schreider, J., and P. Wofford. 2003. *Ambient Air Monitoring for Pesticides in Lompoc, California*. Vol. 1: Executive Summary. California Department of Pesticide Regulation, Sacramento, CA.

Schmalfuss J., B. Matthes, K. Knuth, P. Böger (2000). Inhibition of Acyl_CoA Elongation by Chloroacetamide Herbicides in Microsomes from Leek Seedlings. *Pesticide Biochemistry and Physiology*, **67**(1):25-35.

Sparling, DW, Fellers, GM, and LL McConnell. 2001. Pesticides and amphibian population declines in California, USA. *Environmental Toxicology & Chemistry* 20(7): 1591-1595.

U.S. EPA (1995). Reregistration Eligibility Decision (RED): Metolachlor. Prevention, Pesticides and Toxic Substances, Washington, D.C. EPA 738-R-95-006.

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

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

U.S. EPA (2006a). *Risks of Metolachlor Use to 26 Evolutionarily Significant Units of Endangered and Threatened Pacific Northwest Salmonids*, June 19, 2006, Environmental Fate and Effects Division, Office of Pesticide Programs, Washington, DC.

U.S. EPA (2007) *Risks of Metolachlor Use to Federally Listed Endangered Barton Springs Salamanders (Eurycea sosorum)*. May 3, 2007, Environmental Fate and Effects Division, Office of Pesticide Programs, Washington, DC.

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

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

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

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

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

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

Zimdahl, R.L. (1993). *Fundamentals of Weed Science.* Academic Press. San Diego California USA.

## *ECOTOX References*

Ecotox references listed below are discussed in the text of this document, and/or tabulated in Appendix B, Ecological Effects. Bibliographies for other toxicological studies located by ECOTOX are listed in Appendix D, ECOTOX Bibliographies. Studies not tabulated in the Ecological Effects Appendix contained endpoints less sensitive than the ones already tabulated.


6797 Mayer, FLJ and Ellersieck, MR. (1986). *Manual of Acute Toxicity: Interpretation and Data Base for 410 Chemicals and 66 Species of Freshwater Animals*. Resour. Publ. No. 160, U.S. Dept of Interior, Fish & Wildlife Service, Washington, DC, 505 p.

19186 Day, KE and Hodge V. (1996). The Toxicity of the Herbicide Metolachlor, Some Transformation Products and a Commercial Safener to an Alga (Selenastrum capricornutum), a Cyanophyte. *Water Qual. Res. J. Can.* 31:197-214.

19461 Fairchild, JF, Ruessler, DS, Haverland, PS, and Carlson, AR. (1997). Comparative Sensitivity of *Selenastrum capricornutum* and *Lemna* minor to Sixteen Herbicides. *Arch. Environ. Contam. Toxicol*, 32:353-357.

20274 Clements, C, Ralph, S., and Petras, M. (1997). Genotoxicity of Select Herbicides in *Rana catesbeiana* Tadpoles using the Alkaline Single-Cell Gel DNA Elcetrophoresis (Comet) Assay. *Environ. Mol. Mutagen*. 29:277-288.

66376 Osano, O, Admiraal, W, and Otieno, D. (2002). Developmental Disorders in Embryos of the Frog *Xenopus laevis* Induced by Chloroacetanilide Herbicides and Their Degradation Products. *Environ. Toxicol. Chem*. 21:375-379.

67700 Ronco, A, Sobrero, C, Grassi, Vkaminski, L, Massolo, L, and Mina, L. (2000) WaterTox Bioassy Intercalibration Network: Results from Argentina. *Environ. Toxicol.* 15:287-296.

68515 Wolfe, MC and Moore, PA. (2002). Effects of the Herbicide Metolachlor on the Perception of Chemical Stimuli by Oronectes rusticus. *J.N.Am. Benthol. Soc* 21:457-74.

73233 Hatton, PJ, Dixon, D, Cole, DJ, Edwards, R. (1996). Glutathione Transferase Activities and Herbicide Selectivity in Maize and Associated Weed Species. Pestic. Sci.46: 267-275.

73249 Warren, SL and Skroch, WA. (1991). Evaluation of Six Herbicides for Potential use in Tree Seed Beds. *J. Environ. Hortic*. 9:160-163.

73251 Hood, LR and Klett, JE. (1992). Preemergent Weed Control in Container-Grown Herbaceous and Woody Plants. *J. Environ. Hortic*. 10:8-11.

83887 Liu, H, Ye, W, Zhan, X, Liu, W. (2006). A Comparative Study of *rac*- and *S*-*metolachlor* Toxicity to *Daphnia magna*. *Ecotox. Environ. Safety*. 63: 451-455.

*Appendix A – Exposure and Monitoring*

*Appendix B – Ecological Effects*

*Appendix C – ECOTOX Bibliography*

*Appendix D – RQ Method and LOCs*

*Appendix E – Analysis Summary*

*Appendix F – GIS Summary and Maps*

*Appendix G – Information on Registered Products with Multiple Active Ingredients*