

**Potential Risks of Atrazine Use to Federally
Threatened California Red-legged Frog
(*Rana aurora draytonii*) and Delta Smelt
(*Hypomesus transpacificus*)**

Pesticide Effects Determinations

**Environmental Fate and Effects Division
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1. Executive Summary

1.1. Purpose of Assessment

The purpose of this assessment is to evaluate potential direct and indirect effects on the California red-legged frog (*Rana aurora draytonii*) (CRLF) and the delta smelt (*Hypomesus transpacificus*) (DS) arising from FIFRA regulatory actions regarding use of atrazine on agricultural and non-agricultural sites. In addition, this assessment evaluates whether these actions can be expected to result in modification of designated critical habitat for the CRLF and the DS.

The CRLF was listed as a threatened species by USFWS in 1996. The species is endemic to California and Baja California (Mexico) and inhabits both coastal and interior mountain ranges. The DS was listed as threatened on March 5, 1993 (58 FR 12854) by the U.S. Fish and Wildlife Service (USFWS) (USFWS, 2007 a). It is only found in Suisun Bay and the Sacramento-San Joaquin estuary near San Francisco Bay.

1.2. Assessed Chemicals

Atrazine is an herbicide that inhibits photosynthesis by stopping electron flow in Photosystem II. Triazine herbicides associate with a protein complex of the Photosystem II in chloroplast photosynthetic membranes (Schulz et al., 1990). The result is an inhibition in the transfer of electrons that in turn inhibits the formation and release of oxygen.

Potential risks from exposure to atrazine degradates were not quantified for aquatic organisms because degradates have been shown to be orders of magnitude less toxic than atrazine to aquatic organisms and are presumed to be less toxic than atrazine to terrestrial plants (Section 4). However, some degradates, including DEA, DIA, and DACT have been shown to be more toxic than atrazine to mammals and birds. Therefore, potential risks to terrestrial animals were considered for these degradates. However, potential risks from exposure to the degradates for terrestrial animals were considerably lower than risks described for parent atrazine.

1.3. Assessment Procedures

This assessment was completed in accordance with the U.S. Fish and Wildlife Service (USFWS) and National Marine Fisheries Service (NMFS) *Endangered Species Consultation Handbook* (USFWS/NMFS, 1998) and is consistent with procedures and methodology outlined in the Agency's Overview Document (U.S. EPA, 2004).

1.3.1. Toxicity Assessment

The assessment endpoints include direct toxic effects on survival, reproduction, and growth of individuals, as well as indirect effects, such as reduction of the food source and/or modification of habitat. Federally designated critical habitat has been established for the CRLF and the DS. Primary constituent elements (PCEs) were used to evaluate whether atrazine has the potential to modify designated critical habitat. Atrazine has been extensively studied, and there are a large number of toxicity studies available in the open literature. The Agency evaluated registrant-submitted studies and data from the open literature to characterize atrazine toxicity. The lowest toxicity value available from acceptable or supplemental studies that evaluated toxicity endpoints that are quantitatively associated with assessment endpoints of growth, reproduction, and survival were used to estimate potential risks to the assessed species and taxonomic groups on which the CRLF and DS depend for sustenance.

Previous risk assessments used threshold concentrations based on the Comprehensive Aquatic Systems Model (CASM) to evaluate the potential for atrazine to affect aquatic plant communities. However, a Scientific Advisory Panel (SAP) was convened in December 2007 to discuss the use of CASM to interpret targeted atrazine monitoring data relative to the robust set of micro/mesocosm data available for atrazine. In its 2007 report on the proceedings of the SAP meeting, the SAP commended the Agency on its use of community-level modeling; however, the Panel expressed concern with issues of model validation, transparency, and the analysis of sensitivity. In order to address the recommendations of the SAP relative to CASM, the Agency is currently working on a number of tasks, including a Quality Assurance (QA) evaluation, an expanded sensitivity analysis, and validation of the model as a tool for interpreting monitoring data.

Given the ongoing work to address the recommendations of the SAP regarding CASM, the Agency acknowledges that threshold concentrations used to evaluate potential indirect effects to listed species via community-level effects to aquatic plants may change. The threshold concentrations used in previous atrazine effects determinations were based on exposures relative to the atrazine micro/mesocosm studies that were unlikely to exceed a CASM-based index. Pending further work to address concerns related to QA evaluation, sensitivity analysis, and validation of CASM, the Agency will not rely on a CASM-based index for this assessment, but will instead use results from the laboratory aquatic plant studies and the thresholds for community level effects that were described in the atrazine IRED (U.S. EPA, 2003), which was 10 to 20 µg/L on a recurrent basis or over a prolonged period of time. However, once the further work on CASM and subsequent modification of threshold concentrations is completed if appropriate, the Agency may revisit the effects determinations presented in this assessment if necessary.

1.3.2. Exposure Assessment

1.3.2.1. Aquatic Exposures

Tier-II aquatic exposure models were used to estimate high-end exposures of atrazine in aquatic habitats resulting from runoff and spray drift from different uses. Peak model-estimated environmental concentrations resulting from different atrazine uses range from 17 to 64 µg/L. The maximum reported monitoring value from surface water data from California evaluated in this assessment were all less than 1.0 ppb. Frequency of detections ranged from 10% to 43%.

1.3.2.2. Terrestrial Exposures

The T-REX model was used to estimate potential atrazine exposures to terrestrial species including birds (surrogate species for terrestrial phase CRLFs), mammals (CRLF prey), and invertebrates (CRLF prey). The AgDRIFT model was also used to estimate deposition of atrazine on terrestrial and aquatic habitats from spray drift and to determine the distance from atrazine use sites the CRLF and the DS may be at risk of direct or indirect effects. The TerrPlant model was used to estimate atrazine exposures to terrestrial-phase CRLF habitat, including plants inhabiting semi-aquatic and dry areas, resulting from uses involving foliar atrazine applications. The T-HERPS model was used to allow for further characterization of the dietary exposures of terrestrial-phase CRLFs relative to birds, which were used as a surrogate species for the CRLF.

1.3.3. Measures of Risk

Acute and chronic risk quotients (RQs) are compared to the Agency's Levels of Concern (LOCs) to identify instances where atrazine use within the action area has the potential to adversely affect the CRLF or DS or adversely modify designated critical habitat. When RQs for a particular type of effect are below LOCs, the pesticide is considered to have "no effect" on the species and its designated critical habitat. Where RQs exceed LOCs, a potential to cause adverse effects or habitat modification is identified, leading to a conclusion of "may affect". If atrazine use "may affect" the assessed species, and/or may cause modification to designated critical habitat, the best available additional information is considered to refine the potential for exposure and effects, and distinguish actions that are NLAA (not likely to adversely affect) from those that are LAA (likely to adversely affect).

1.4. Atrazine Uses Assessed

All potential uses of atrazine in California were evaluated as part of this assessment. Currently registered non-agricultural uses of atrazine include residential areas such as playgrounds and home lawns, turf (golf courses and recreational fields), and forestry. Agricultural uses include corn, sorghum, macadamia nuts, and guava. Several uses (fallow and rights of way) have geographic restrictions that restrict use to areas

outside of California and are not considered relevant to this assessment. In addition, sugarcane is a registered atrazine use and is grown in limited acreage in Imperial county. Currently, this acreage does not overlap with CRLF or DS occurrence however, exposure estimates have been provided to account for any future expansion of sugarcane into CRLF or DS ranges.

Atrazine is used throughout the United States on a number of agricultural commodities (primarily corn and sorghum). Relative to the major atrazine use areas in the Midwest corn/sorghum belt, atrazine use in California is moderate based on data available from the CDPR. In California, the major uses of atrazine were on forestry followed by corn and Bermuda grass. It is typically applied as a spray by air or ground, but residential use products also include a granular formulation.

1.5. Summary of Conclusions

Based on the best available information, the Agency makes a May Affect, and Likely to Adversely Affect determination for the CRLF and the DS from the labeled use of atrazine as described in Table 1.1. The effects determination is based on potential direct effects to terrestrial phase CRLFs and potential indirect effects to the CRLF and the DS. The LAA determination applies to the following uses for both species: corn, sorghum, macadamia nuts, guava, residential uses, forestry, and turf.

Additionally, the Agency has determined that there is the potential for modification of designated critical habitat of the CRLF and the DS from the use of the chemical. A summary of the risk conclusions and effects determinations for each listed species assessed and their designated critical habitat is presented in Tables 1.1 and 1.2. Further information on the results of the effects determination is included as part of the Risk Description in Section 5.2. Given the LAA determination for the CRLF and the DS and potential modification of designated critical habitat for both species, a description of the baseline status and cumulative effects for the CRLF is provided in **Attachment 2** and the baseline status and cumulative effects for the DS is provided in **Attachment 4**.

Table 1.1 Effects Determination Summary for Effects of Atrazine on the CRLF and the DS

Species	Effects Determination ¹	Basis for Determination
California red-legged frog (<i>Rana aurora draytonii</i>)	LAA ¹	Potential for Direct Effects
		<i>Aquatic-phase (Eggs, Larvae, and Adults):</i> No acute or chronic LOCs were exceeded for aquatic phase amphibians based on available fish and amphibian data.
		<i>Terrestrial-phase (Juveniles and Adults):</i> Acute and chronic LOCs were exceeded for birds. Considering factors such as lower food intake of terrestrial phase amphibians relative to birds reduces EECs and RQs, but does not reduce RQs to levels that are below LOCs.
		Potential for Indirect Effects
		<i>Aquatic prey items, aquatic habitat, cover and/or primary productivity</i> Atrazine could potentially impact terrestrial and aquatic plants to an extent that could result in indirect effects to the CRLF or modification of critical habitat.
Delta Smelt (<i>Hypomesus transpacificus</i>)	LAA ¹	<i>Terrestrial prey items, riparian habitat</i> CRLFs could be affected as a result of potential impacts to grassy/herbaceous vegetation. Food item abundance such as terrestrial invertebrates and terrestrial phase amphibians are not expected to be impacted to an extent that is expected to adversely affect the CRLF. However, potential impacts to herbivorous mammal abundance to an extent that could indirectly affect terrestrial phase CRLFs could not be precluded.
		Potential for Direct Effects No LOC exceedances occurred for acute or chronic effects to fish.
		Potential for Indirect Effects Labeled uses of atrazine have the potential to adversely affect the delta smelt and modify critical habitat either by reducing available food (marine copepods and aquatic plants), by impacting the riparian habitat of grassy and herbaceous riparian areas, or by impacting water quality via effects to aquatic vegetation.

¹ May affect, likely to adversely affect (LAA)

Table 1.2 Effects Determination Summary for Atrazine Use and CRLF and DS Critical Habitat Impact Analysis

Assessment Endpoint	Effects Determination ¹	Basis for Determination
Modification of aquatic-phase PCEs (DS and CRLF)	Habitat Modification¹	As described in Table 1.1., the effects determination for the potential for atrazine to affect aquatic phase CRLFs and the DS is LAA. This determination is based on the potential for atrazine to affect the DS and CRLF's food and habitat. Potential effects to aquatic invertebrates, aquatic plants, and terrestrial (riparian) plants identified in this assessment could result in aquatic habitat modification.
Modification of terrestrial-phase PCE (CRLF)		As described in Table 1.1., the effects determination for the potential for atrazine to affect terrestrial phase CRLFs is LAA. This determination is based on the potential for atrazine to directly affect terrestrial phase CRLFs and indirectly affect CRLFs by potentially impacting food supply and vegetative habitat. These potential effects could result in modification of critical habitat.

Based on the conclusions of this assessment, a formal consultation with the U. S. Fish and Wildlife Service under Section 7 of the Endangered Species Act should be initiated.

When evaluating the significance of this risk assessment's direct/indirect and adverse habitat modification effects determinations, it is important to note that pesticide exposures and predicted risks to the listed 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.

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 and the DS life stages within the action area and/or applicable designated critical habitat. 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 assessed species.
- Quantitative information on prey base requirements for the assessed species. While existing information provides a preliminary picture of the types of food sources utilized by the assessed species, 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 species and potential modification to critical habitat.

2. Problem Formulation

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

2.1 Purpose

The purpose of this endangered species assessment is to evaluate potential direct and indirect effects on individuals of the federally threatened California red-legged frog (*Rana aurora draytonii*) (CRLF) and the delta smelt (*Hypomesus transpacificus*) (DS) arising from FIFRA regulatory actions regarding labeled uses of atrazine. In addition, this assessment evaluates whether labeled atrazine use is expected to result in modification of designated critical habitat for the CRLF and the DS. This ecological risk assessment has been prepared consistent with the settlement agreement in *Center for Biological Diversity (CBD) vs. EPA et al.* (Case No. 02-1580-JSW(JL)) entered in Federal District Court for the Northern District of California on October 20, 2006. This assessment also addresses the DS for which atrazine was alleged to be of concern in a separate suit (*Center for Biological Diversity (CBD) vs. EPA et al.* (Case No. 07-2794-JCS)).

In this assessment, direct and indirect effects to the CRLF and the DS and potential modification to their designated critical habitat are evaluated in accordance with the methods described in the Agency's Overview Document (U.S. EPA 2004). The effects determinations for each listed species assessed is based on a weight-of-evidence method that relies heavily on an evaluation of risks to each taxonomic group relevant to assess both direct and indirect effects to the listed species and the potential for modification of their designated critical habitat (*i.e.*, a taxon-level approach). Screening level methods include use of standard models such as PRZM-EXAMS, T-REX, TerrPlant, and AgDRIFT, all of which are described at length in the Overview Document. In addition, THERPS has been used to refine estimates of exposure and risk to amphibians. 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 atrazine is based on an action area. The action area is the area directly or indirectly affected by the federal action, as indicated by the exceedance of the Agency’s Levels of Concern (LOCs). It is acknowledged that the action area for a national-level FIFRA regulatory decision associated with a use of atrazine may potentially involve numerous areas throughout the United States and its Territories. However, for the purposes of this assessment, attention will be focused on relevant sections of the action area including those geographic areas associated with locations of the CRLF and DS and their designated critical habitat within the state of California. As part of the “effects determination,” one of the following three conclusions will be reached separately for each of the assessed species in the lawsuits regarding the potential use of atrazine in accordance with current labels:

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

The CRLF and the DS have designated critical habitats associated with them. Designated critical habitat identifies specific areas that have the physical and biological features, (known as primary constituent elements or PCEs) essential to the conservation of the listed species. The PCEs for the CRLF are aquatic and upland areas where suitable breeding and non-breeding aquatic habitat is located, interspersed with upland foraging and dispersal habitat. PCEs for the DS include characteristics required to maintain habitat for spawning, larval and juvenile transport, rearing, and adult migration.

If the results of initial screening-level assessment methods show no direct or indirect effects (no LOC exceedances) upon individuals or upon the PCEs of the species’ designated critical habitat, a “no effect” determination is made for use of atrazine as it relates to each species and its designated critical habitat. If, however, potential direct or indirect effects to individuals of each species are anticipated or effects may impact the PCEs of the designated critical habitat, a preliminary “may affect” determination is made for the FIFRA regulatory action regarding atrazine.

If a determination is made that use of atrazine “may affect” a listed species or its designated critical habitat, additional information is considered to refine the potential for exposure and for effects to each species and other taxonomic groups upon which these species depend (*e.g.*, prey items). Additional information, including spatial analysis (to determine the geographical proximity of the assessed species’ habitat and atrazine use sites) and further evaluation of the potential impact of atrazine on the PCEs is also used to determine whether modification of designated critical habitat may occur. Based on the refined information, the Agency uses the best available information to distinguish those

actions that “may affect, but are not likely to adversely affect” from those actions that “may affect and are likely to adversely affect” the assessed listed species and/or result in “no effect” or potential modification to the PCEs of its designated critical habitat. This information is presented as part of the Risk Characterization in Section 5 of this document.

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

2.2 Scope

Atrazine is currently registered as an herbicide in the U.S. to control annual broadleaf and grass weeds in corn, sorghum, sugarcane, and other crops. In addition to food crops, atrazine is also used on a variety of non-food crops, forests, residential/industrial uses, golf course turf, recreational areas, and rights-of-way.

The end result of the EPA pesticide registration process 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, acceptable methods of application, approved use sites, and any restrictions on how applications may be conducted. Thus, the use or potential use of atrazine in accordance with the approved product labels is “the action” being assessed.

This ecological risk assessment is for currently registered uses of atrazine in portions of the action area reasonably assumed to be biologically relevant to the CRLF or the DS and their designated critical habitat. Further discussion of the action area and designated critical habitat is provided in Section 2.4 and 2.5.

2.2.1. Evaluation of Degradates

This ecological risk assessment includes all potential ecological stressors resulting from the use of atrazine, including atrazine and its potential degradates of concern. Degradates of concern may include those that are found at significant (>10% by weight relative to parent) concentrations in available degradation studies and those that are of toxicological concern. Degradates of atrazine include hydroxyatrazine (HA), deethylatrazine (DEA),

deisopropylatrazine (DIA), and diaminochloroatrazine (DACT). Atrazine degradates and their routes of formation are summarized in Table 2.1 below.

Table 2.1. Summary of Formation Pathway of Atrazine Degradates of Concern

Degradate	Formation Pathway				
	Photolysis in Water	Photolysis in Soil	Aerobic Metabolism in Soil	Anaerobic Metabolism in Soil	Anaerobic Metabolism in Water
Deethylatrazine (DEA)	X (18%) ^a	X (18%) ^a	X	X	X
Deisopropylatrazine (DIA)	X	X	X	X	X
Diaminochlorotriazine (DACT)	X (15%) ^a	X	X	X	X
Hydroxyatrazine (HA)		X	X	X	X

^a Values in parentheses are percentage of parent formed; only values for major (>10%) degradates are shown. See U.S. EPA, 2003a for additional discussion on these degradates.

Comparison of available toxicity information for the degradates of atrazine indicates lesser aquatic toxicity than the parent for fish, aquatic invertebrates, and aquatic plants (Table 2.2). Specifically, the available degrade toxicity data for HA indicate that it is not toxic to freshwater fish and invertebrates at the limit of its solubility in water. In addition, no adverse effects were observed in fish or daphnids at DACT concentrations up to 100 mg/L. Acute toxicity values for DIA are 3- and 36-fold less sensitive than acute toxicity values for atrazine in fish and daphnids, respectively. In addition, available aquatic plant degrade toxicity data for HA, DEA, DIA, and DACT report non-definitive EC₅₀ values (i.e., 50% effect was not observed at the highest test concentrations) at concentrations that are at least 700 times higher than the lowest reported aquatic plant EC₅₀ value for parent atrazine. Although degrade toxicity data are not available for terrestrial plants, lesser or equivalent toxicity is assumed, given the available ecotoxicological information for other taxonomic groups including aquatic plants and the likelihood that the degradates of atrazine may lose efficacy as an herbicide. Therefore, given the lesser aquatic toxicity of the degradates as compared to the parent, and the relatively small proportion of the degradates expected to be in the environment and available for exposure relative to atrazine, the focus of this assessment is parent atrazine for aquatic organisms.

Table 2.2 Comparison of Acute Freshwater Toxicity Values for Atrazine and Degradates

Substance Tested	Fish LC₅₀ (µg/L)	Daphnid EC₅₀ (µg/L)	Aquatic Plant EC₅₀ (µg/L)
Atrazine	5,300	3,500	1
HA	>3,000 (no effects at saturation)	>4,100 (no effects at saturation)	>10,000
DACT	>100,000	>100,000	No data
DIA	17,000	126,000 (NOAEC: 10,000)	2,500
DEA	No data	No data	1,000

The DEA degradate has been shown to be of similar toxicity to birds and mammals on an acute oral basis. Other dealkylatrazine degradates have been shown to be more acutely toxic to rats and more developmentally toxic to gestating rat pups than the parent atrazine (Table 2.3 below). Acute avian studies suggest that DIA is less toxic than atrazine to birds on an acute oral basis. No avian toxicity data for DACT are available; therefore, based on the equivalent toxicity in mammals, DACT may also be of toxicological concern in birds, which are used as surrogate species for terrestrial phase CRLFs. Additional details on available toxicity data for the degradates are provided in Section 4 of this assessment and in Appendix A.

Table 2.3. Summary of Available Degradate Toxicity Data in Birds and Mammals

Chemical	Acute Bird LD50 (mg/kg-bw)	Acute Mammal LD50 (mg/kg-bw)	Mammal Developmental NOAEL (mg/kg-bw)
Atrazine	940 (MRID 00024721)	1900 (MRID 00024706)	10 (MRID 40566302)
HA	>2000 (MRID 46500008)	Not available	25 (MRID 41065202)
DEA	768 (MRID 46500009)	1240 (MRID 43013201)	5 (MRID 43013209)
DIA	>2000 (MRID 46500007)	1100 (MRID 43013202)	5 (MRID 43013208)
DACT	Not available	Not available	2.5 (MRID 41392402)

Appendix B includes RQs for degradates for terrestrial animals. High-end RQs were calculated by assuming degradate exposure occurs at 18% of parent atrazine levels (highest detected amount in available fate studies) and the lowest toxicity value across all degradates for birds and mammals. Under these assumptions, RQs derived for the atrazine degradates for birds and mammals are considerably lower than RQs used to characterize potential risks in this assessment for parent atrazine, although potential risks from exposure to degradates exceeded concern levels. Therefore, risk conclusions for atrazine are protective of potential risks from exposure to degradates as well.

2.2.2. Evaluation of Mixtures

The Agency does not routinely include an evaluation of mixtures of active ingredients (either those mixtures of multiple active ingredients in product formulations or those in the applicator's tank) in its risk assessments. In the case of product formulations of active ingredients (registered product containing more than one active ingredient) each active ingredient is subject to an individual risk assessment for regulatory decision regarding the active ingredient on a particular use site. If effects data are available for a formulated

product containing more than one active ingredient, they may be used qualitatively or quantitatively in accordance with the Agency's Overview Document and the Services' Evaluation Memorandum (U.S. EPA, 2004; USFWS/NMFS, 2004).

The results of available toxicity data for environmental mixtures of atrazine with other pesticides are presented in Appendix A. According to the available data, other pesticides may combine with atrazine to produce additive, more than additive, or less than additive effects. Based on the results of the available data, study authors claim that more than additive effects with atrazine may occur for a number of organophosphate insecticides including diazinon, chlorpyrifos, and methyl parathion, as well as herbicides including alachlor. If chemicals that show more than additive effects with atrazine are present in the environment in combination with atrazine, the toxicity of atrazine may be increased, offset by other environmental factors, or even reduced by the presence of antagonistic contaminants if they are also present in the mixture. The variety of chemical interactions presented in the available data set suggests that the toxic effect of atrazine, in combination with other pesticides used in the environment, can be a function of many factors including but not necessarily limited to: (1) the exposed species, (2) the co-contaminants in the mixture, (3) the ratio of atrazine and co-contaminant concentrations, (4) differences in the pattern and duration of exposure among contaminants, and (5) the differential effects of other physical/chemical characteristics of the receiving waters (e.g., organic matter present in sediment and suspended water). Quantitatively predicting the combined effects of all these variables on mixture toxicity to any given taxa with confidence is beyond the capabilities of the available data. However, a qualitative discussion of implications of the available pesticide mixture effects data involving atrazine on the confidence of risk assessment conclusions is addressed as part of the uncertainty analysis for this effects determination.

Atrazine has registered products that contain multiple active ingredients. Analysis of the available open literature and acute oral mammalian LD₅₀ data for multiple active ingredient products relative to the single active ingredient is provided in **Appendix C**. The results of this analysis show that an assessment based on the toxicity of the single active ingredient of atrazine is appropriate.

2.3 Previous Assessments

Atrazine has been the subject of a number of ecological risk assessments conducted by U.S. EPA. Several assessments have recently been conducted on the potential for atrazine to affect a number of listed species as part of the Natural Resources Defense Counsel settlement agreement and one listed species included in a second settlement agreement with the Center for Biological Diversity and Save Our Springs Alliance. These effects determinations, which are available on the web at www.epa.gov/espp, review atrazine's potential direct and indirect effects to the following listed species:

- 1) Barton Springs salamander (*Eurycea sosorum*) (U.S. EPA, 2006c);
- 2) Shortnose sturgeon (*Acipenser brevirostrum*), dwarf wedgemussel (*Alasmidonta heterodon*), loggerhead turtle (*Caretta caretta*), Kemp's ridley

- turtle (*Lepidochelys kempii*), leatherback turtle (*Dermochelys coriacea*), and green turtle (*Chelonia mydas*) in the Chesapeake Bay (U.S. EPA, 2006d);
- 3) Alabama sturgeon (*Scaphirhynchus suttkusi*) (U.S. EPA, 2006e).
- 4) Pink mucket pearly mussel, Rough pigtoe mussel, Shiny pigtoe pearly mussel, Fine-rayed pigtoe mussel, Heavy pigtoe mussel, Ovate clubshell mussel, Southern clubshell mussel, and Stirrupshell mussel (U.S. EPA, 2007a).
- 5) Fat pocketbook pearly mussel (*Potamilus capax*), Purple cat's paw pearly mussel (*Epioblasma obliquata*), and Northern riffleshell (*Epioblasma torulosa rangiana*) endangered species assessment (U.S. EPA 2007b)
- 6) Topeka shiner and pallid sturgeon (U.S. EPA, 2008)

In addition, the Agency completed a refined ecological risk assessment for potential aquatic impacts of atrazine use in January 2003 (U.S. EPA, 2003a). This assessment was based on laboratory ecotoxicological data as well as microcosm and mesocosm field studies found in publicly available literature, a substantial amount of monitoring data for freshwater streams, lakes, reservoirs, and estuarine areas, and incident reports of adverse effects on aquatic and terrestrial organisms associated with the use of atrazine.

The results of this ecological assessment for atrazine is fully discussed in the January 31, 2003, Interim Reregistration Eligibility Decision (IRED)¹. In that assessment the need for the following information related to potential ecological risks was established: 1) a monitoring program to identify and evaluate potentially vulnerable water bodies in corn, sorghum, and sugarcane use areas; and 2) further information on potential amphibian gonadal developmental responses to atrazine. On October 31, 2003, EPA issued an addendum that updated the IRED issued on January 31, 2003 (U.S. EPA, 2003b). This addendum described new scientific developments pertaining to monitoring of watersheds and potential effects of atrazine on endocrine-mediated pathways of amphibian gonadal development. As of the writing of this assessment, preliminary data from the ecological monitoring study have been submitted and are used to characterize potential exposures. However, analyses of the data are ongoing.

Finally, On August 1, 2003, EPA released an assessment of the potential effects of atrazine to 26 listed Evolutionarily Significant Units (ESUs) of Pacific salmon and steelhead. That assessment concluded that registered uses of atrazine would have “no effect”, directly or indirectly to the 26 ESUs nor to designated critical habitat. While potential effects to riparian vegetation were noted, the extent of atrazine use in the large geographic areas comprising the relevant watersheds, lead to a conclusion that use would have no effect on the species from any potential effects to riparian areas.

As discussed in the October 2003 IRED, the Agency also conducted an evaluation of the submitted studies regarding the potential effects of atrazine on amphibian gonadal development and presented its assessment in the form of a white paper for external peer review to a FIFRA Scientific Advisory Panel (SAP) in June 2003². In the white paper

¹ The 2003 Interim Reregistration Eligibility Decision for atrazine is available at the following Web site: <http://www.epa.gov/oppsrrd1/REDs/0001.pdf>.

² The Agency's May 2003 White Paper on Potential Developmental Effects of Atrazine on Amphibians is available at <http://www.epa.gov/oscpmont/sap/2003/june/finaljune2002telconfreport.pdf>.

dated May 29, 2003, the Agency summarized seventeen studies consisting of both open literature and registrant-submitted laboratory and field studies involving both native and non-native species of frogs (U.S. EPA, 2003d). The Agency concluded that none of the studies fully accounted for environmental and animal husbandry factors capable of influencing endpoints that the studies were attempting to measure. The Agency also concluded that the current lines-of-evidence did not show that atrazine produced consistent effects across a range of exposure concentrations and amphibian species tested.

Based on this assessment, the Agency concluded and the SAP concurred that there was sufficient evidence to formulate a hypothesis that atrazine exposure may impact gonadal development in amphibians, but there were insufficient data to confirm or refute the hypothesis (<http://www.epa.gov/oscpmont/sap/2003/June/junemeetingreport.pdf>). Because of the inconsistency and lack of reproducibility across studies and an absence of a dose-response relationship in the currently available data, the Agency determined that the data did not alter the conclusions reached in the January 2003 IRED regarding uncertainties related to atrazine's potential effects on amphibians. The SAP supported EPA in seeking additional data to reduce uncertainties regarding potential risk to amphibians. Subsequent data collection occurred following a multi-tiered process outlined in the Agency's white paper to the SAP (U.S. EPA, 2003d). These data were submitted, and a second SAP was held to discuss results of these studies (U.S. EPA 2007). These data suggested that gonadal development of larval *Xenopus laevis* was not affected by atrazine exposure. Based on these data, it was concluded that atrazine does not consistently affect amphibian gonadal development. It is acknowledged, however, that there is uncertainty in using *Xenopus laevis* to represent all amphibians.

Finally, in December 2007 a SAP was convened to address the potential for community level effects to aquatic plants in Midwestern streams (http://www.epa.gov/scipoly/sap/meetings/2007/120407_mtg.htm). This SAP involved the review of a community level model as well as discussion of ongoing surface water monitoring, identification of sites exceeding levels of concern determined with the community model, identification of factors contributing to exposures deemed to be above levels of concern, and discussion of a proposed approach for extrapolation of surface water results to non-sampled watersheds.

2.4 Stressor Source and Distribution

2.4.1 Environmental Fate Properties

The following fate and transport description for atrazine was summarized based on information contained in the 2003 IRED (U.S. EPA, 2003a). In general, atrazine is expected to be mobile and persistent in the environment. The main route of dissipation is microbial degradation under aerobic conditions. Because of its persistence and mobility, atrazine is expected to reach surface and ground water. This is confirmed by the widespread detections of atrazine in surface water and ground water. Atrazine is persistent in soil, with a half-life (time until 50% of the parent atrazine remains) exceeding 1 year under some conditions (Armstrong et al., 1967). Atrazine can

contaminate nearby non-target plants, soil and surface water via spray drift during application. Atrazine is applied directly to target plants during foliar application, but pre-plant and pre-emergent applications are generally far more prevalent.

The resistance of atrazine to abiotic hydrolysis (stable at pH 5, 7, and 9) and to direct aqueous photolysis (stable under sunlight at pH 7), and its only moderate susceptibility to degradation in soil (aerobic laboratory half-lives of 3-4 months) indicates that atrazine is unlikely to undergo rapid degradation on foliage. Likewise, a relatively low Henry's Law constant (2.6×10^{-9} atm-m³/mol) indicates that atrazine is not likely to undergo rapid volatilization from foliage. However, its relatively low octanol/water partition coefficient ($\text{Log } K_{ow} = 2.7$), and its relatively low soil/water partitioning (Freundlich K_{ads} values < 3 and often < 1) may somewhat offset the low Henry's Law constant value, thereby possibly resulting in some volatilization from foliage. In addition, its relatively low adsorption characteristics indicate that atrazine may undergo substantial washoff from foliage. It should also be noted that foliar dissipation rates for numerous pesticides have generally been somewhat greater than otherwise indicated by their physical chemical and other fate properties.

In terrestrial field dissipation studies performed in Georgia, California, and Minnesota, atrazine dissipated with half lives of 13, 58, and 261 days, respectively. The inconsistency in these reported half-lives could be attributed to the temperature variation between the studies in which atrazine was seen to be more persistent in colder climates. Long-term field dissipation studies also indicated that atrazine could persist over a year in such climatic conditions. A forestry field dissipation study in Oregon (aerial application of 4 lb ai/A) estimated an 87-day half-life for atrazine on exposed soil, a 13-day half-life in foliage, and a 66-day half-life on leaf litter.

Atrazine is applied directly to soil during pre-planting and/or pre-emergence applications. Atrazine is transported indirectly to soil due to incomplete interception during foliar application, and due to washoff subsequent to foliar application. The available laboratory and field data are reported above. For aquatic environments, reported half-lives were much longer. In an anaerobic aquatic study, atrazine overall (total system), water, and sediment half-lives were given as 608, 578, and 330 days, respectively.

A number of degradates of atrazine were detected in laboratory and field environmental fate studies as previously summarized in Table 2.1. Deethyl-atrazine (DEA) and deisopropyl-atrazine (DIA) were detected in all studies, and hydroxy-atrazine (HA) and diaminochloro-atrazine (DACT) were detected in all but one of the listed studies. Deethylhydroxy-atrazine (DEHA) and deisopropylhydroxy-atrazine (DIHA) were also detected in one of the aerobic studies.

All of the chloro-triazine and hydroxy-triazine degradates detected in the laboratory metabolism studies were present at less than the 10% of applied that the Agency uses to classify degradates as "major degradates" (U.S. EPA, 2004); however, several of these degradates were detected at percentages greater than 10% in soil and aqueous photolysis studies. Insufficient data are available to estimate half-lives for these degradates. The

dealkylated degradates are more mobile than parent atrazine, while HA is less mobile than atrazine and the dealkylated degradates.

2.4.2 Mechanism of Action

Atrazine inhibits photosynthesis by stopping electron flow in Photosystem II. Triazine herbicides associate with a protein complex of the Photosystem II in chloroplast photosynthetic membranes (Schulz et al., 1990). The result is an inhibition in the transfer of electrons that in turn inhibits the formation and release of oxygen.

2.4.3 Use Characterization

Nationally, atrazine is used on a variety of terrestrial food crops, non-food crops, forests, residential/industrial uses, golf course turf, recreational areas and rights-of-way. Atrazine yields season-long weed control in corn, sorghum and certain other crops. Nationally, the major atrazine uses include corn (83 percent of total ai produced per year - primarily applied pre-emergence), sorghum (11 percent of total ai produced), sugarcane (4 percent of total ai produced) and others (2 percent ai produced). Atrazine formulations include dry flowable, flowable liquid, liquid, water dispersible granule, wettable powder and coated fertilizer granule. Nationally, the maximum registered use rate for atrazine is 4 lbs ai/acre; and 4 lbs ai/acre is the maximum, single application rate for the following uses: sugarcane, forest trees (softwoods, conifers), forest plantings, guava, macadamia nuts, ornamental sod (turf farms), and ornamental and/or shade trees.

As a selective herbicide, atrazine is applied pre-emergence and post-emergence. Figure 2.1 presents the national distribution of use of atrazine (Kaul and Jones, 2006). Table 3.1 presents a summary of atrazine uses being assessed quantitatively in this assessment.

National Distribution of Atrazine Use (total lbs)

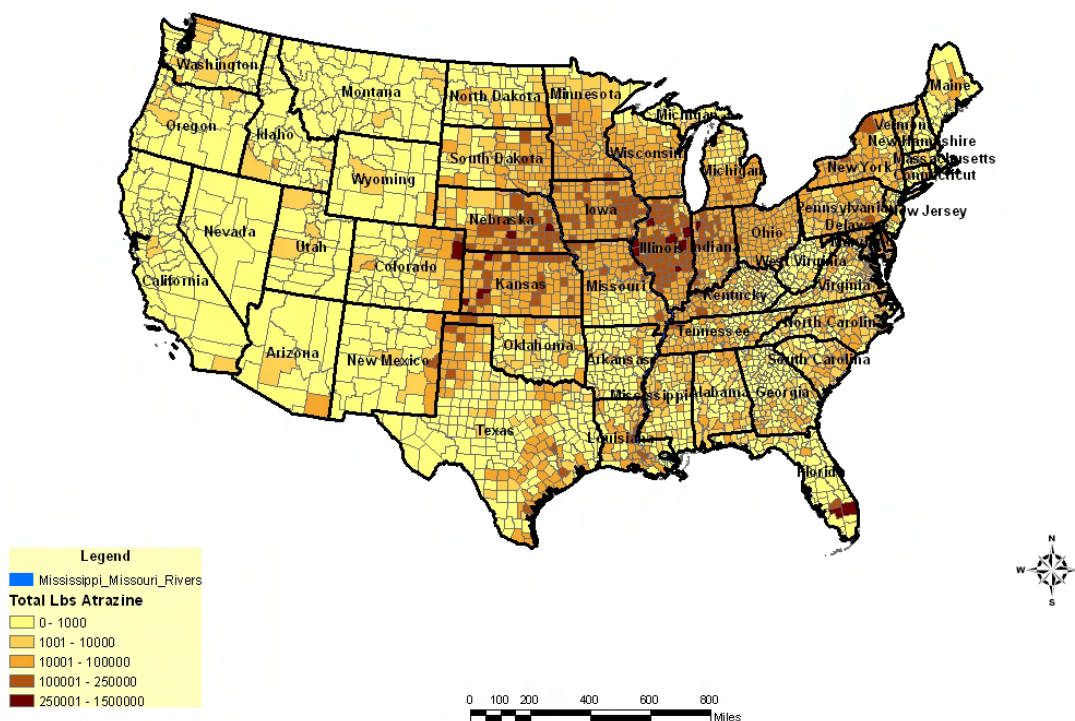


Figure 2.1 National Extent of Atrazine Use (lbs)

The Agency's Biological and Economic Analysis Division (BEAD) provides an analysis of both national- and county-level usage information (Kaul and Jones, 2006) using state-level usage data obtained from USDA-NASS³, Doane (www.doane.com; the full dataset is not provided due to its proprietary nature) and the California's Department of Pesticide Regulation Pesticide Use Reporting (CDPR PUR) database⁴. CDPR PUR is considered a more comprehensive source of usage data than USDA-NASS or EPA proprietary databases, and thus the usage data reported for atrazine by county in this California-specific assessment were generated using CDPR PUR data. Eight years (1999-2006) of usage data were included in this analysis (Carter and Kaul, 2008). Data from CDPR PUR were obtained for every pesticide application made on every use site at the section level (approximately one square mile) of the public land survey system. BEAD summarized these data to the county level by site, pesticide, and unit treated (Carter and Kaul, 2008). Calculating county-level usage involved summarizing across all applications made within

³ 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>.

⁴ 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>.

a section and then across all sections within a county for each use site and for each pesticide. The county level usage data that were calculated include: average annual pounds applied, average annual area treated, and average and maximum application rate across all eight years. The units of area treated are also provided where available.

Relative to the major atrazine use areas in the Midwest corn/sorghum belt, atrazine use in California is moderate based on data available from the CDPR. Total use between 1999 and 2006 declined from roughly 72,000 lbs total to almost 33,000 lbs in 2006 suggesting an overall decline in atrazine use. In California, the major uses of atrazine were on forestry with an average of 20,780 lbs between 1999 and 2006, followed by corn with an average total of 15,437 lbs, and Bermuda grass at 5,556 lbs. All other registered uses were less than 1,000 lbs annually across this time span. A summary of atrazine usage for all California use sites is provided below in **Table 2.4**. Average annual pounds of atrazine applied from 1999 to 2004 by county is depicted in Figure 2.2.

Table 2.4 Summary of California Department of Pesticide Registration (CDPR) Pesticide Use Reporting (PUR) Data from 1999 to 2006 for Currently Registered Atrazine Uses

Site Name	Average Pounds Applied	Avg App Rate (lb/acre) All Uses	Avg 95th% App Rate (lb/acre)	Avg 99th% App Rate (lb/acre)	Avg Max App Rate (lb/acre)
Bermuda grass	5556	1.4	2.0	3.2	13.7
Christmas Trees	72	1.4	3.1	3.3	3.3
Corn (all uses)	15,437	1.5	2.7	3.6	4.6
Forest, Timberland	20,780	3.2	4.0	4.8	5.4
Landscape Maintenance	492	NA	NA	NA	NA
Sorghum (all uses)	779	1.3	1.5	1.5	1.5
Sugarcane	341	2.1	2.9	3.9	3.9
Turf/Sod	85	0.7	0.8	1.8	1.8

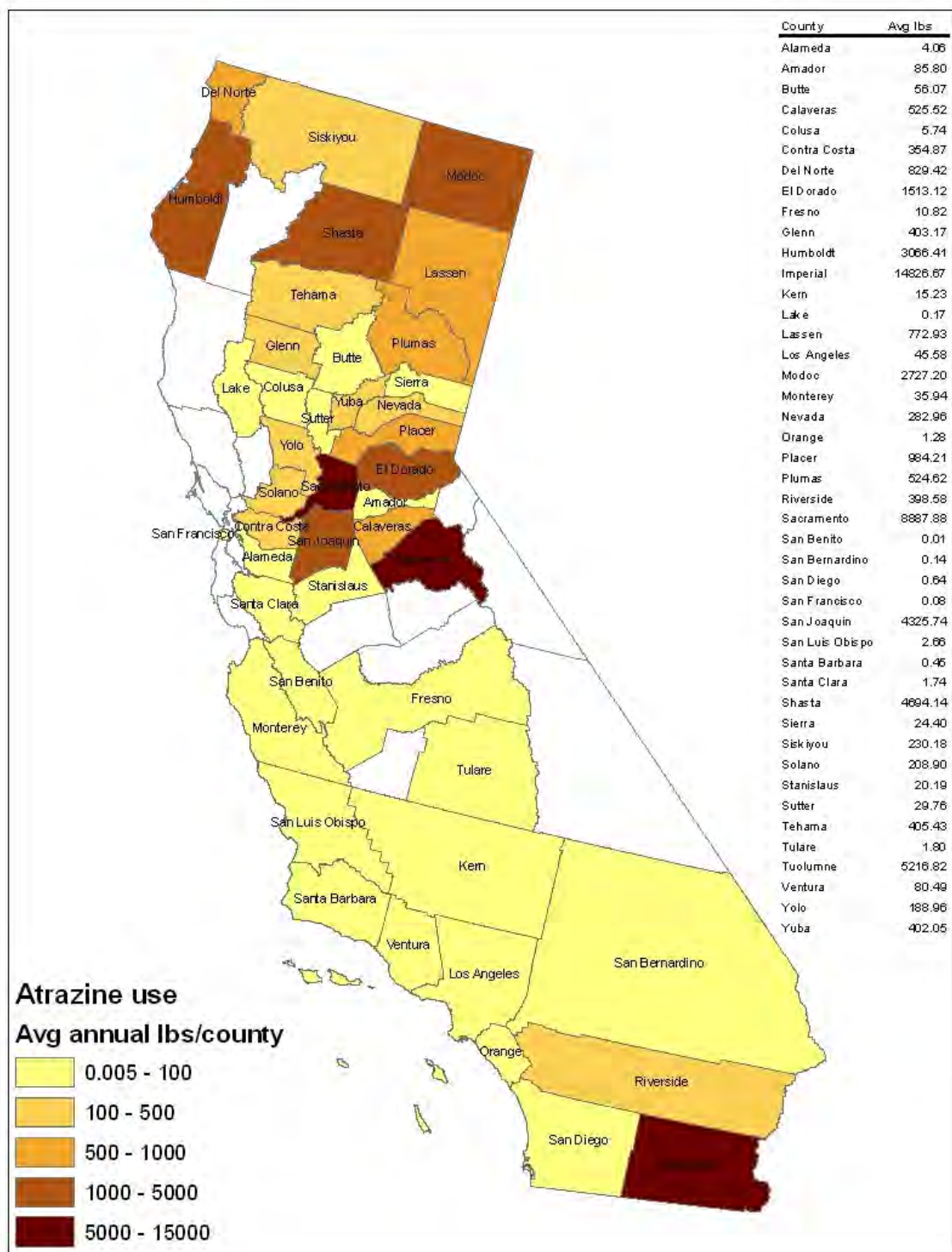


Figure 2.2. Average annual pounds of atrazine applied from 1999 to 2004 across all uses

2.5 Assessed Species

Table 2.5 provides a summary of the current distribution, habitat requirements, and life history parameters for the listed species being assessed. More detailed life-history and distribution information can be found in Attachments 1 and 3. See Figures 2.2 and 2.3 for a map of the current range and designated critical habitat of the assessed listed species.

Table 2.5. Summary of Current Distribution, Habitat Requirements, and Life History Information for the Assessed Listed Species¹

Assessed Species	Size	Current Range	Habitat Type	Designated Critical Habitat?	Reproductive Cycle	Diet
California red-legged frog (<i>Rana aurora draytonii</i>)	Adult (85-138 cm in length), Females – 9-238 g, Males – 13-163 g; Juveniles (40-84 cm in length)	Northern CA coast, northern Transverse Ranges, foothills of Sierra Nevada, and in southern CA south of Santa Barbara	Freshwater perennial or near-perennial aquatic habitat with dense vegetation; artificial impoundments; riparian and upland areas	Yes	<u>Breeding</u> : Nov. to Apr. <u>Tadpoles</u> : Dec. to Mar. <u>Young juveniles</u> : Mar. to Sept.	<u>Aquatic-phase</u> ² : algae (tadpoles only), freshwater aquatic invertebrates and fish <u>Terrestrial-phase</u> : terrestrial invertebrates, small mammals, and frogs
Delta smelt (<i>Hypomesus transpacificus</i>)	Up to 120 mm in length	Suisun Bay and the Sacramento-San Joaquin estuary (known as the Delta) near San Francisco Bay, CA	The species is adapted to living in fresh and brackish water. They typically occupy estuarine areas with salinities below 2 parts per thousand (although they have been found in areas up to 18ppt). They live along the freshwater edge of the mixing zone (saltwater-freshwater interface).	Yes	They spawn in fresh or slightly brackish water upstream of the mixing zone. Spawning season usually takes place from late March through mid-May, although it may occur from late winter (Dec.) to early summer (July-August). Eggs hatch in 9 – 14 days.	They primarily eat planktonic copepods, cladocerans, amphipods, and insect larvae. Larvae feed on phytoplankton; juveniles feed on zooplankton.

¹ For more detailed information on the distribution, habitat requirements, and life history information of the assessed listed species, see Attachment 3

² For the purposes of this assessment, tadpoles and submerged adult frogs are considered “aquatic” because exposure pathways in the water are considerably different than those that occur on land.

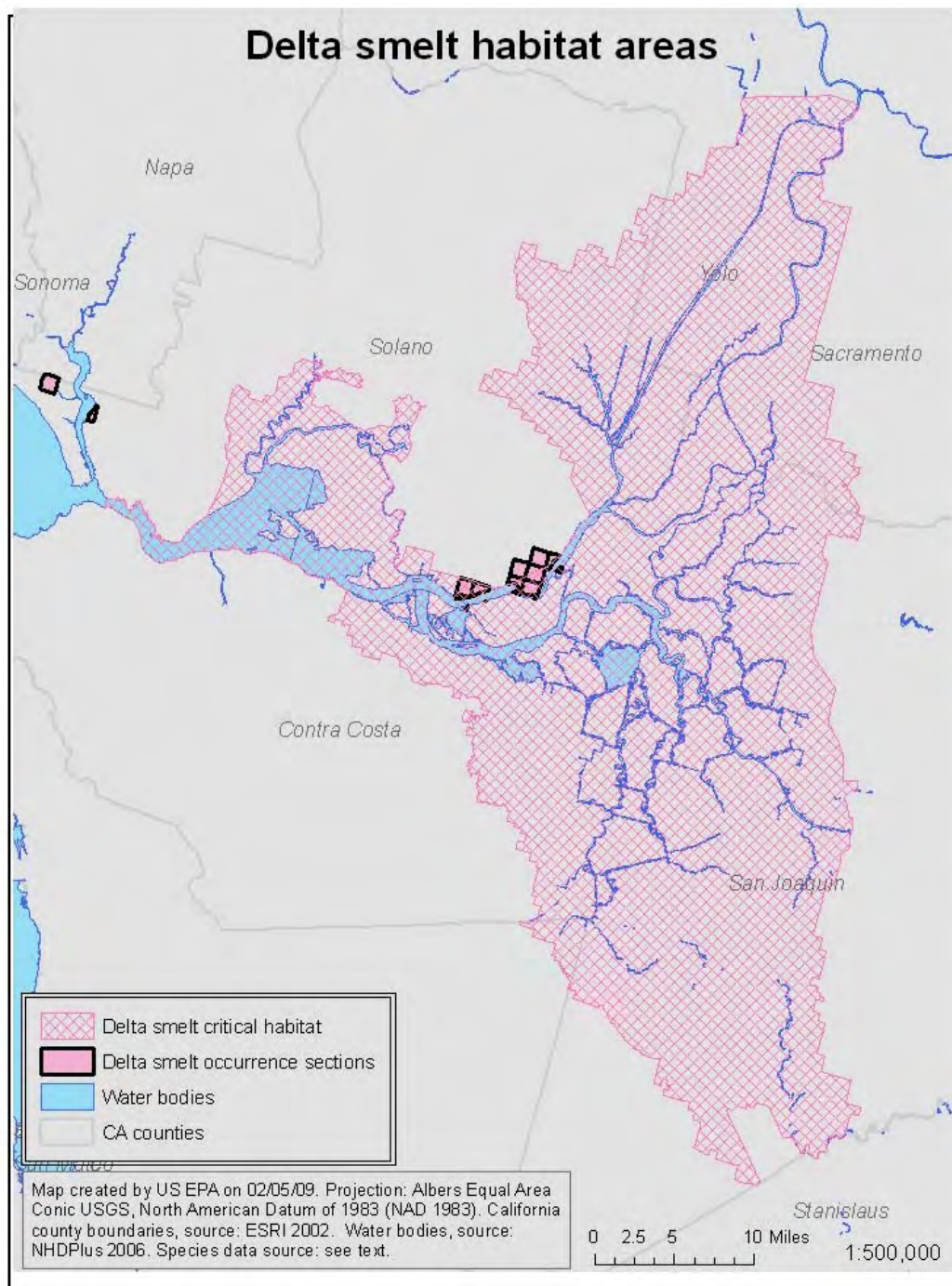


Figure 2.2 DS Habitat Areas

Other Known Occurrences from the CNDDb

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

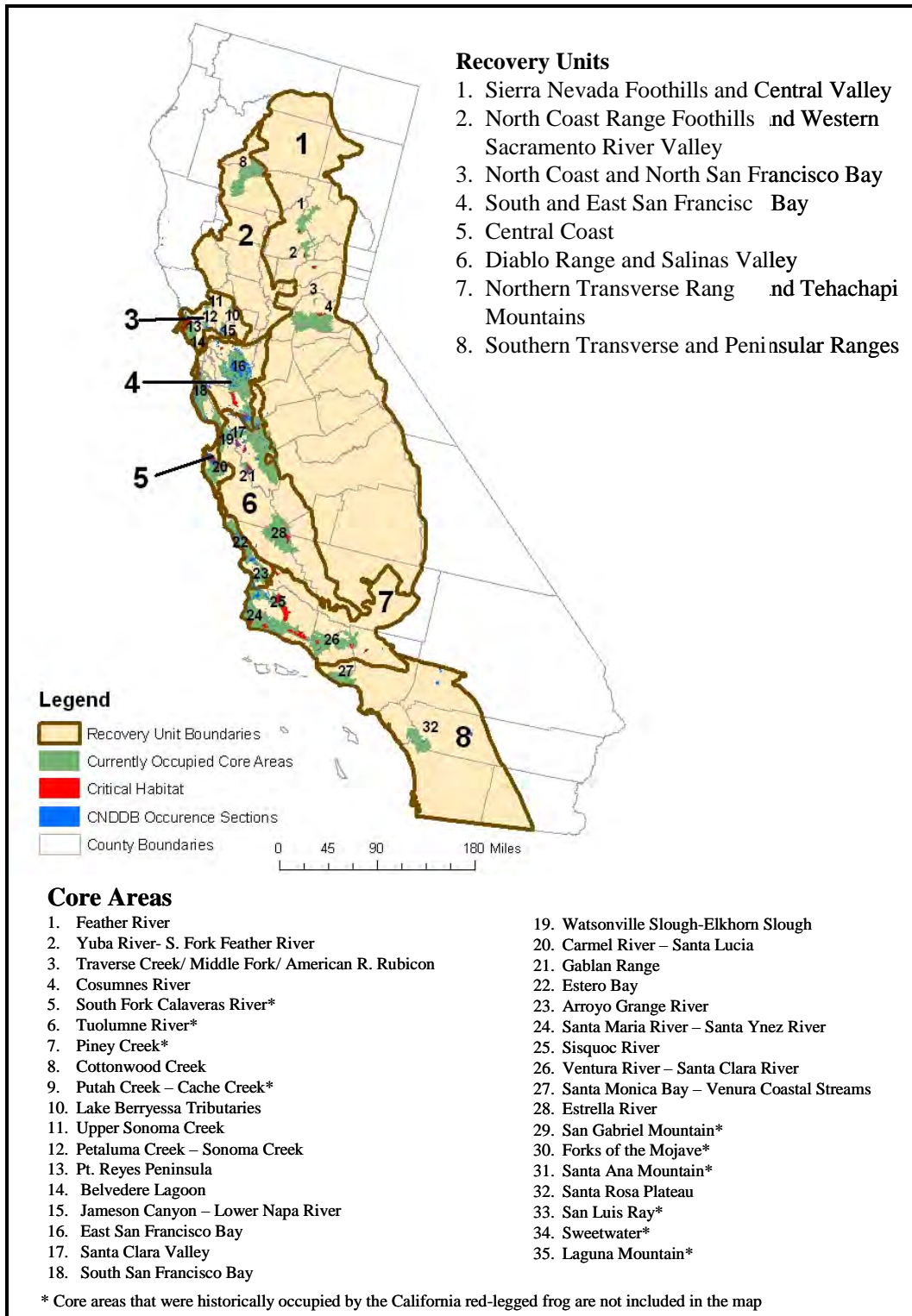


Figure 2.3. Recovery Unit, Core Area, Critical Habitat, and Occurrence Designations for CRLF

2.6. Designated Critical Habitat

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

To be included in a critical habitat designation, the habitat must be ‘essential to the conservation of the species.’ Critical habitat designations identify, to the extent known using the best scientific and commercial data available, habitat areas that provide essential life cycle needs of the species or areas that contain certain primary constituent elements (PCEs) (as defined in 50 CFR 414.12(b)). PCEs include, but are not limited to, space for individual and population growth and for normal behavior; food, water, air, light, minerals, or other nutritional or physiological requirements; cover or shelter; sites for breeding, reproduction, rearing (or development) of offspring; and habitats that are protected from disturbance or are representative of the historic geographical and ecological distributions of a species. **Table 2.6** describes the PCEs for the critical habitats designated for the CRLF and the DS.

Table 2.6. Designated Critical Habitat PCEs for the CRLF and DS ^{1,2}.

Species	PCEs	Reference
CRLF	Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond.	50 CFR 414.12(b), 2006
	Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	
	Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	
	Reduction and/or modification of aquatic-based food sources for pre-metamorphs (<i>e.g.</i> , algae)	
	Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance	
	Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal	
	Reduction and/or modification of food sources for terrestrial phase juveniles and adults	
	Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	
DS	Spawning Habitat—shallow, fresh or slightly brackish backwater sloughs and edgewaters to ensure egg hatching and larval viability. Spawning areas also must provide suitable water quality (<i>i.e.</i> , low “concentrations of pollutants”) and substrates for egg attachment (<i>e.g.</i> , submerged tree roots and branches and emergent vegetation).	59 FR 65256 65279, 1994
	Larval and Juvenile Transport—Sacramento and San Joaquin Rivers and their tributary channels must be protected from physical disturbance and flow disruption. Adequate river flow is necessary to transport larvae from upstream spawning areas to rearing habitat in Suisun Bay. Suitable water quality must be provided so that maturation is not impaired by pollutant concentrations.	
	Rearing Habitat—Maintenance of the 2 ppt isohaline and suitable water quality (low concentrations of pollutants) within the Estuary is necessary to provide delta smelt larvae and juveniles a shallow protective, food-rich environment in which to mature to adulthood.	
	Adult Migration— Unrestricted access to suitable spawning habitat in a period that may extend from December to July. Adequate flow and suitable water quality may need to be maintained to attract migrating adults in the Sacramento and San Joaquin River channels and their associated tributaries. These areas also should be protected from physical disturbance and flow disruption during migratory periods.	

¹ These PCEs are in addition to more general requirements for habitat areas that provide essential life cycle needs of the species such as, space for individual and population growth and for normal behavior; food, water, air, light, minerals, or other nutritional or physiological requirements; cover or shelter; sites for breeding, reproduction, rearing (or development) of offspring; and habitats that are protected from disturbance or are representative of the historic geographical and ecological distributions of a species.

² PCEs that are abiotic, including, physico-chemical water quality parameters such as salinity, pH, and hardness are not evaluated because these processes are not biologically mediated and, therefore, are not relevant to the endpoints included in this assessment.

More detail on the designated critical habitat applicable to this assessment can be found in Attachment 1 (for the CRLF) and Attachment 3 (for the DS). Activities that may

destroy or modify critical habitat are those that alter the PCEs and jeopardize the continued existence of the species. Evaluation of actions related to use of atrazine that may alter the PCEs of the designated critical habitat for the CRLF and DS form the basis of the critical habitat impact analysis.

As previously discussed, 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 atrazine is expected to directly impact living organisms within the action area, critical habitat analysis for atrazine is limited in a practical sense to those PCEs of critical habitat that are biological or that can be reasonably linked to biologically mediated processes.

2.7 Action Area

For listed species assessment purposes, the action area is considered to be the area affected directly or indirectly by the federal action and not merely the immediate area involved in the action (50 CFR 402.02). It is recognized that the overall action area for the national registration of atrazine uses is likely to encompass considerable portions of the United States based on the large array of both agricultural and non-agricultural uses. Based on the available atrazine monitoring data (discussed further in Section 3.2.6) and the toxicity data for the most sensitive non-vascular aquatic plant, the Agency's LOCs are likely to be exceeded in many watersheds that are in proximity to or downstream of atrazine use sites. Therefore, the overall action area for atrazine is likely to include many watersheds of the United States that co-occur and/or are in proximity to agricultural and non-agricultural atrazine use sites. However, in order to focus this assessment, the scope limits consideration of the overall action area to those geographic portions that may be applicable to the protection of the CRLF and DS included in this assessment.

In this case, the listed species being assessed reside solely within the state of California and thus this assessment is focused geographically within the state. The registered agricultural and non-agricultural uses relative to potential land cover classes from the NLCD, which represent the current and possible future extent of the use sites, represent the initial area of concern and are illustrated in Figure 2.4.

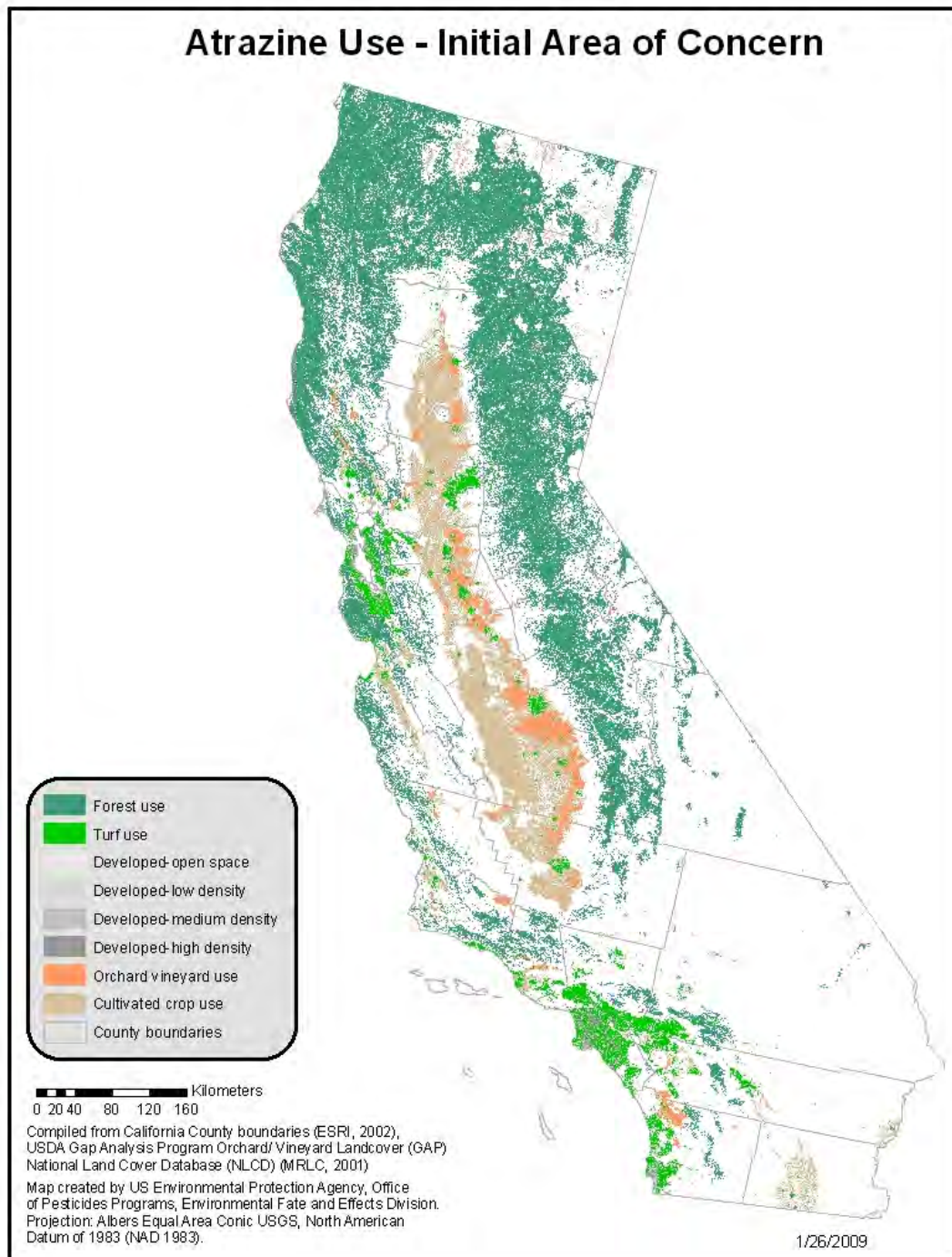


Figure 2.4. Potential Atrazine Use Sites in California Representing the Initial Area of Concern

In addition, a number of studies have been conducted that have identified some type of biological effect (see Appendix A and E) at the tested exposure levels without a corresponding no effect level. Therefore, thresholds for some types of environmental effect have not been identified, and it is not possible to identify an atrazine exposure level that is definitively associated with no environmental effects regardless of the ecological significance of the effect. For this reason, the action area (area where an effect may occur) has been conservatively defined as the entire state of California.

2.8 Assessment Endpoints and Measures of Ecological Effect

Assessment endpoints are defined as “explicit expressions of the actual environmental value that is to be protected.”⁵ Selection of the assessment endpoints is based on valued entities (*e.g.*, CRLF and DS), organisms important in the life cycle of the assessed species, 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 atrazine (*e.g.*, runoff, spray drift, *etc.*), and the routes by which ecological receptors are exposed to atrazine (*e.g.*, direct contact, *etc.*).

2.8.1. Assessment Endpoints

Assessment endpoints for the CRLF and the DS include direct toxic effects on the survival, reproduction, and growth of individuals, as well as indirect effects, such as reduction of the prey base or modification of its habitat. In addition, potential modification of critical habitat is assessed by evaluating potential effects to PCEs, which are components of the habitat areas that provide essential life cycle needs of the assessed species. Each assessment endpoint requires one or more “measures of ecological effect,” defined as changes in the attributes of an assessment endpoint or changes in a surrogate entity or attribute in response to exposure to a pesticide. Specific measures of ecological effect are generally evaluated based on acute and chronic toxicity information from registrant-submitted guideline tests that are performed on a limited number of organisms. Additional ecological effects data from the open literature are also considered. It should be noted that assessment endpoints are limited to direct and indirect effects associated with survival, growth, and fecundity, and do not include the full suite of sublethal effects used to define the action area. According the Overview Document (USEPA 2004), the Agency relies on acute and chronic effects endpoints that are either direct measures of impairment of survival, growth, or fecundity or endpoints for which there is a scientifically robust, peer reviewed relationship that can quantify the impact of the measured effect endpoint on the assessment endpoints of survival, growth, and fecundity.

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 risks for each of the assessed species associated with exposure to atrazine is provided in **Table 2.8**.

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

As described in the Agency's Overview Document (U.S. EPA, 2004), the most sensitive endpoint for each taxonomic group is used for risk estimation. For this assessment, evaluated taxa include aquatic-phase amphibians, freshwater and saltwater fish, freshwater and saltwater invertebrates, aquatic plants, birds (surrogate for terrestrial-phase amphibians), mammals, terrestrial invertebrates, and terrestrial plants. Acute (short-term) and chronic (long-term) toxicity information is characterized based on registrant-submitted studies and a comprehensive review of the open literature on atrazine.

Table 2.7 identifies the taxa used to assess the potential for direct and indirect effects from the uses of atrazine for each listed species assessed. The specific assessment endpoints used to assess the potential for direct and indirect effects to each listed species are provided in **Table 2.8**.

Table 2.7. Taxa Used in the Analyses of Direct and Indirect Effects for the Assessed Listed Species.

Listed Species	Birds / Terr. Amphibian	Mammals	Terr. Plants	Terr. Inverts.	FW Fish / Amphibian	FW Inverts.	Estuarine /Marine Fish	Estuarine /Marine Inverts.	Aquatic Plants
California red-legged frog	Direct Indirect (prey)	Indirect (prey)	Indirect (habitat)	Indirect (prey)	Direct Indirect (prey)	Indirect (prey)	N/A	N/A	Indirect (food/habitat)
Delta smelt	N/A	N/A	Indirect (habitat)	N/A	Direct ^a	Indirect (prey)	Direct ^a	Indirect (prey) ^b	Indirect (food/habitat)

N/A = Not applicable; Terr. = Terrestrial; Invert. = Invertebrate; FW = Freshwater

^a The most sensitive species across freshwater and saltwater environments was used for the DS.

^b The marine copepod was primarily used for the indirect effects assessment for the DS because it is an important food source for this species.

Table 2.8. Assessment Endpoints Used to Evaluate the Potential for the Use of Atrazine to Result in Direct and Indirect Effects to the CRLF and the DS

Taxa Used to Assess Direct and/or Indirect Effects to Assessed Species	Assessed Listed Species	Assessment Endpoints	Measures of Ecological Effects
1. Freshwater Fish and Aquatic-phase Amphibians	<u>Direct Effect</u> – -Aquatic-phase CRLF -DS	Survival, growth, and reproduction of individuals via direct effects	1a. Amphibian acute LC ₅₀ (ECOTOX) or most sensitive fish acute LC ₅₀ (guideline or ECOTOX) if no suitable amphibian data are available
	<u>Indirect Effect (prey)</u> -Aquatic-phase CRLF	Survival, growth, and reproduction of individuals via indirect effects on aquatic prey food supply (<i>i.e.</i> , fish and aquatic-phase amphibians)	1b. Amphibian chronic NOAEC (ECOTOX) or most sensitive fish chronic NOAEC (guideline or ECOTOX) 1c. Amphibian early-life stage data (ECOTOX) or most sensitive fish early-life stage NOAEC (guideline or ECOTOX)
2. Freshwater Invertebrates	<u>Indirect Effect (prey)</u> -Aquatic-phase CRLF	Survival, growth, and reproduction of individuals	2a. Most sensitive freshwater invertebrate EC ₅₀ (guideline or ECOTOX)

	-DS	via indirect effects on aquatic prey food supply (<i>i.e.</i> , freshwater invertebrates)	2b. Most sensitive freshwater invertebrate chronic NOAEC (guideline or ECOTOX)
3. Estuarine/Marine Fish	<u>Direct Effect</u> -DS	Survival, growth, and reproduction of individuals via direct effects on the DS	3a. Most sensitive estuarine/marine fish LC ₅₀ (guideline or ECOTOX) 3b. Most sensitive estuarine/marine fish chronic NOAEC (guideline or ECOTOX)
4. Estuarine/Marine Invertebrates	<u>Indirect Effect (prey)</u> -DS	Survival, growth, and reproduction of individuals via indirect effects on aquatic prey food supply (<i>i.e.</i> , estuarine/marine invertebrates)	4a. Most sensitive estuarine/marine invertebrate EC ₅₀ (guideline or ECOTOX) 4b. Most sensitive estuarine/marine invertebrate chronic NOAEC (guideline or ECOTOX)
5. Aquatic Plants (freshwater/marine)	<u>Indirect Effect (food/habitat)</u> -Aquatic-phase CRLF -DS	Survival, growth, and reproduction of individuals via indirect effects on habitat, cover, food supply, and/or primary productivity (<i>i.e.</i> , aquatic plant community)	5a. Vascular plant acute EC ₅₀ (duckweed guideline test or ECOTOX vascular plant) 5b. Non-vascular plant acute EC ₅₀ (freshwater algae or diatom, or ECOTOX non-vascular)
6. Birds	<u>Direct Effect</u> -Terrestrial-phase CRLF	Survival, growth, and reproduction of individuals via direct effects	6a. Most sensitive bird ^b or terrestrial-phase amphibian acute LC ₅₀ or LD ₅₀ (guideline or ECOTOX)
	<u>Indirect Effect (prey)</u> -CRLFs	Survival, growth, and reproduction of individuals via indirect effects on terrestrial prey (surrogate for amphibians)	6b. Most sensitive bird ^b or terrestrial-phase amphibian chronic NOAEC (guideline or ECOTOX)
7. Mammals	<u>Indirect Effect</u> -Terrestrial-phase CRLF	Survival, growth, and reproduction of individuals via indirect effects on terrestrial prey (mammals)	7a. Most sensitive laboratory rat acute LC ₅₀ or LD ₅₀ (guideline or ECOTOX) 7b. Most sensitive laboratory rat chronic NOAEC (guideline or ECOTOX)
8. Terrestrial Invertebrates	<u>Indirect Effect (prey)</u> -Terrestrial-phase CRLF	Survival, growth, and reproduction of individuals via indirect effects on terrestrial prey (terrestrial invertebrates)	8a. Most sensitive terrestrial invertebrate acute EC ₅₀ or LC ₅₀ (guideline or ECOTOX) ^c 8b. Most sensitive terrestrial invertebrate chronic NOAEC (guideline or ECOTOX)
9. Terrestrial Plants	<u>Indirect Effect (food/habitat) (non-obligate relationship)</u> -Terrestrial- and aquatic-phase CRLF -DS	Survival, growth, and reproduction of individuals via indirect effects on food and habitat (<i>i.e.</i> , riparian and upland vegetation)	9a. Distribution of EC ₂₅ for monocots (seedling emergence, vegetative vigor, or ECOTOX) 9b. Distribution of EC ₂₅ for dicots (seedling emergence, vegetative vigor, or ECOTOX)

2.8.2 Assessment Endpoints for Designated Critical Habitat

As previously discussed, designated critical habitat is assessed to evaluate actions related to the use of atrazine that may alter the PCEs of the assessed species' designated critical habitat. PCEs for the assessed species were previously described in Section 2.6. Actions

that may modify critical habitat are those that alter the PCEs and jeopardize the continued existence of the assessed species. Therefore, these actions are identified as assessment endpoints. Evaluation of PCEs as assessment endpoints is limited to those of a biological nature (*i.e.*, the biological resource requirements for the listed species associated with the critical habitat) and those for which atrazine effects data are available.

Assessment endpoints used to evaluate potential for direct and indirect effects are equivalent to the assessment endpoints used to evaluate potential modification of designated critical habitat. If a potential for direct or indirect effects is found, then there is also a potential for modification of critical habitat. Some components of 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.

2.9 Conceptual Model

2.9.1 Risk Hypotheses

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

The labeled use of atrazine within the action area may:

- directly affect the CRLF and/or the DS by causing mortality or by adversely affecting growth or fecundity;
- indirectly affect the CRLF and/or the DS and/or modify their designated critical habitat by reducing or changing the composition of food supply;
- indirectly affect the CRLF and/or the DS and/or modify their designated critical habitat by reducing or changing the composition of the aquatic plant community in the species' current range, thus affecting primary productivity and/or cover;
- indirectly affect the CRLF and/or the DS and/or modify their designated critical habitat by reducing or changing the composition of the terrestrial plant community in the species' current range;
- indirectly affect the CRLF and/or the DS and/or modify their designated critical habitat by reducing or changing aquatic habitat in their current range (via modification of water quality parameters, habitat morphology, and/or sedimentation);

2.9.2 Diagram

The conceptual model is a graphic representation of the structure of the risk assessment. It specifies the atrazine release mechanisms, biological receptor types, and effects endpoints of potential concern. The conceptual models for aquatic and terrestrial phases of the CRLF and the DS and the conceptual models for the aquatic and terrestrial PCE

components of critical habitat are shown in **Figures 2.4 and 2.5**. Although the conceptual models for direct/indirect effects and modification of designated critical habitat PCEs are shown on the same diagrams, the potential for direct/indirect effects and modification of PCEs will be evaluated separately in this assessment. Exposure routes shown in dashed lines are not quantitatively considered because the contribution of those potential exposure routes to potential risks to the CRLF and the DS and modification to designated critical habitat is expected to be negligible.

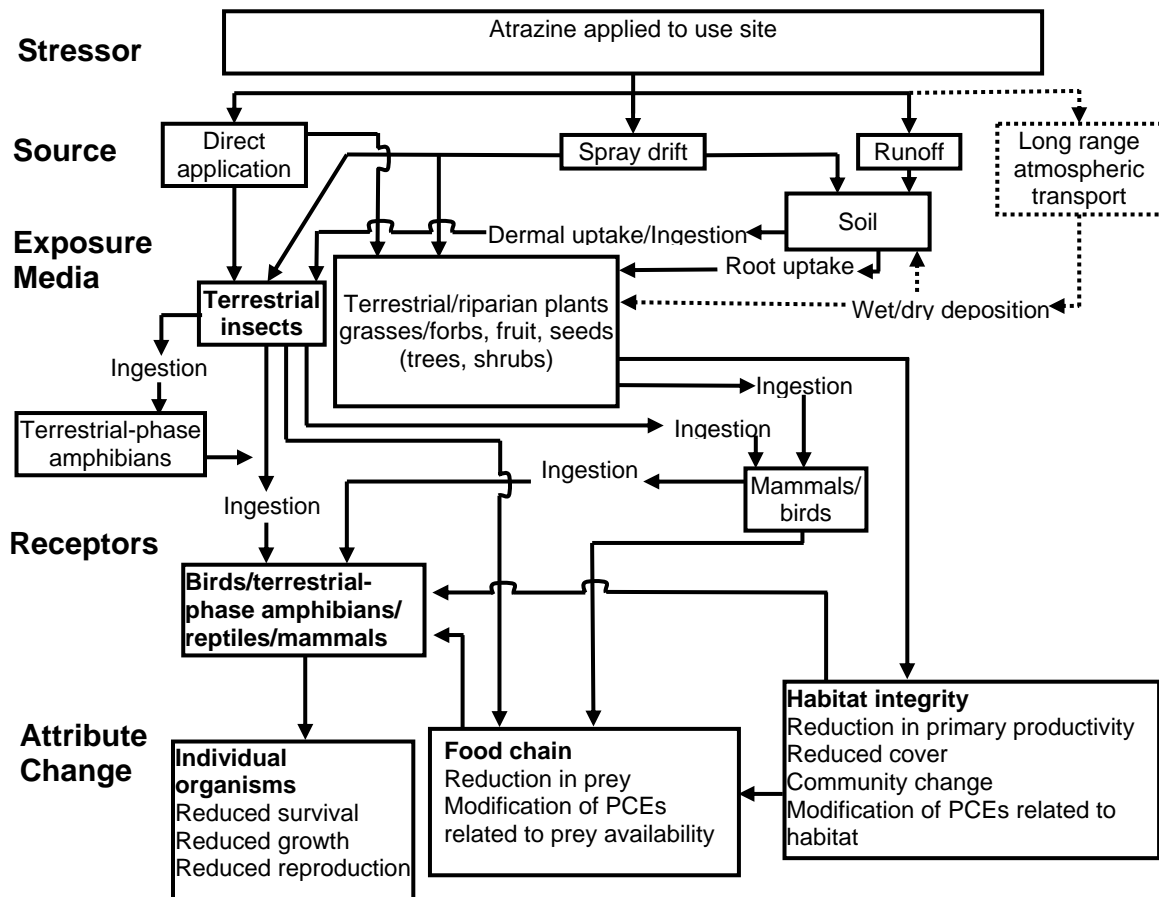


Figure 2.4. Conceptual Model for Terrestrial-Phase CRLF

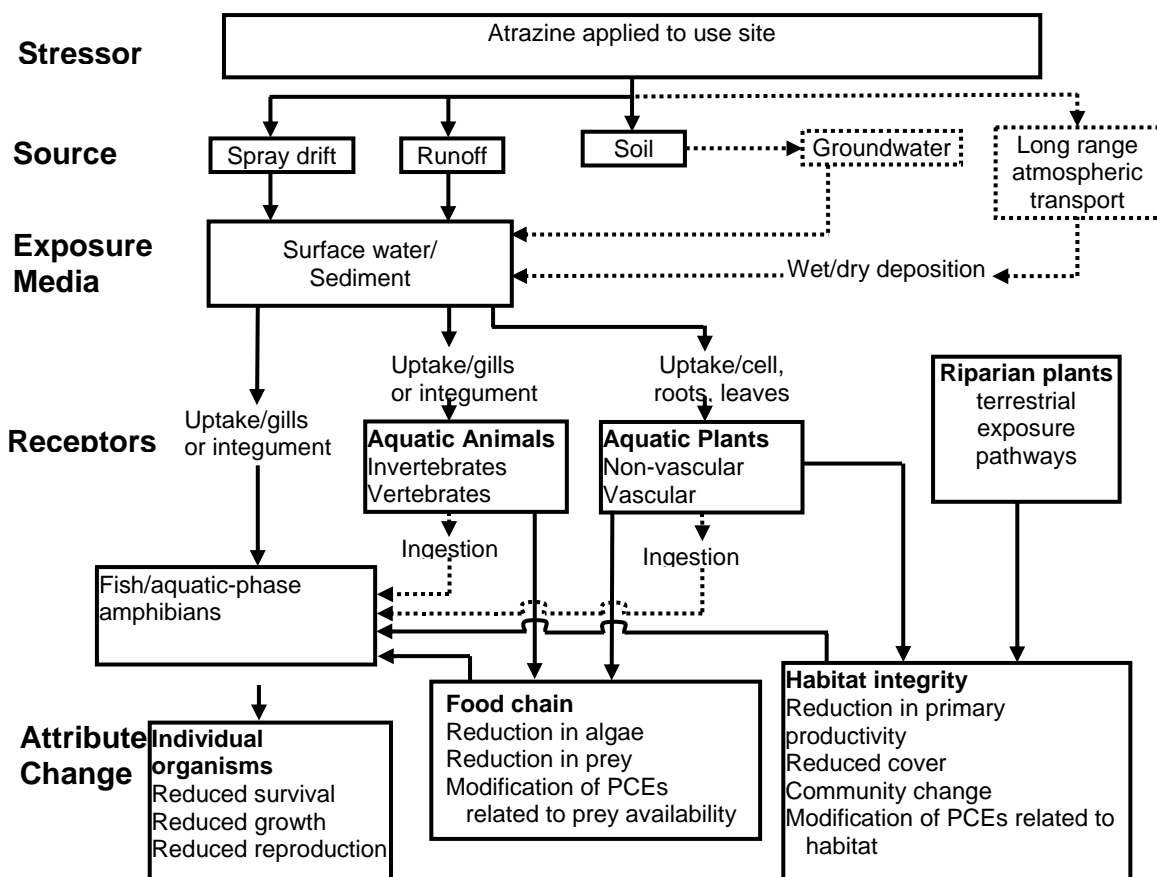


Figure 2.5. Conceptual Model for Aquatic-Phase CRLF and the DS

2.10 Analysis Plan

In order to address the risk hypothesis, the potential for direct and indirect effects to the CRLF and the DS, prey items, and habitat is estimated based on a taxon-level approach. In the following sections, the use, environmental fate, and ecological effects of atrazine are characterized and integrated to assess the risks. This is accomplished using a risk quotient (ratio of exposure concentration to effects concentration) approach. Although risk is often defined as the likelihood and magnitude of adverse ecological effects, the risk quotient-based approach does not provide a quantitative estimate of likelihood and/or magnitude of an adverse effect. However, as outlined in the Overview Document (U.S. EPA, 2004), the likelihood of effects to individual organisms from particular uses of atrazine is estimated using the probit dose-response slope and either the level of concern (discussed below) or actual calculated risk quotient value.

2.10.1 Measures of Exposure

Measures of exposure are based on aquatic and terrestrial models that predict estimated environmental concentrations (EECs) of atrazine using maximum labeled application rates and methods of application. The models used to predict aquatic EECs are the Pesticide Root Zone Model coupled with the Exposure Analysis Model System

(PRZM/EXAMS). The model used to predict terrestrial EECs on food items is T-REX. The model used to derive EECs relevant to terrestrial and wetland plants is TerrPlant. These models are parameterized using relevant reviewed registrant-submitted environmental fate data.

PRZM (v3.12.2, May 2005) and EXAMS (v2.98.4.6, April 2005) are screening simulation models coupled with the input shell pe5.pl (Aug 2007) to generate daily exposures and 1-in-10 year EECs of atrazine that may occur in surface water bodies adjacent to application sites receiving atrazine through runoff and spray drift. PRZM simulates pesticide application, movement and transformation on an agricultural field and the resultant pesticide loadings to a receiving water body via runoff, erosion and spray drift. EXAMS simulates the fate of the pesticide and resulting concentrations in the water body. The standard scenario used for ecological pesticide assessments assumes application to a 10-hectare agricultural field that drains into an adjacent 1-hectare water body, 2-meters deep (20,000 m³ volume) with no outlet. PRZM/EXAMS was used to estimate screening-level exposure of aquatic organisms to atrazine. The measure of exposure for aquatic species is the 1-in-10 year return peak or rolling mean concentration. The 1-in-10-year 60-day mean is used for assessing chronic exposure to fish; the 1-in-10-year 21-day mean is used for assessing chronic exposure for aquatic invertebrates.

Exposure estimates for the terrestrial animals assumed to be in the target area or in an area exposed to spray drift are derived using the T-REX model (version 1.4.1, 10/2008). This model incorporates the Kenaga nomograph, as modified by Fletcher *et al.* (1994), which is based on a large set of actual field residue data. The upper limit values from the nomograph represented the 95th percentile of residue values from actual field measurements (Hoerger and Kenega, 1972).

For modeling purposes, direct exposures of the CRLF to atrazine through contaminated food are estimated using the EECs for a small bird (20 g) that consumes small insects. Dietary-based and dose-based exposures of potential prey (small mammals) are assessed using the small mammal (15 g) which consumes short grass. The small bird (20g) consuming small insects and the small mammal (15g) consuming short grass are used because these categories represent the largest RQs of the size and dietary categories in T-REX that are appropriate surrogates for the CRLF and one of its prey items. Estimated exposures of terrestrial insects to atrazine are bound by using the dietary based EECs for small insects and large insects.

Birds are currently used as surrogates for terrestrial-phase amphibians and reptiles. However, amphibians and reptiles are poikilotherms (body temperature varies with environmental temperature) while birds are homeotherms (temperature is regulated, constant, and largely independent of environmental temperatures). Therefore, amphibians and reptiles tend to have much lower metabolic rates and lower caloric intake requirements than birds or mammals. As a consequence, birds are likely to consume more food than amphibians and reptiles on a daily dietary intake basis, assuming similar caloric content of the food items. Therefore, the use of avian food intake allometric equation as a surrogate to amphibians and reptiles is likely to result in an over-estimation

of exposure and risk for reptiles and terrestrial-phase amphibians. For this reason, food intake equations more specific to terrestrial phase amphibians were used to refine the potential dietary exposures to terrestrial phase CRLF. These food intake equations were incorporated into T-REX to form an exposure model called T-HERPS (v. 1.0), which allows for an estimation of food intake for poikilotherms using the same basic procedure as T-REX uses to estimate avian food intake.

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

The spray drift model, AgDRIFT, was used to assess exposures of terrestrial animals to atrazine deposited on terrestrial habitats by spray drift. In addition to the buffered area from the spray drift analysis, the downstream extent of atrazine that exceeds the LOC for the effects determination is also considered.

2.10.2 Measures of Effect

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

The assessment of risk for direct effects to the terrestrial-phase CRLF makes the assumption that toxicity of atrazine to birds is similar to or less than the toxicity to terrestrial-phase amphibians and reptiles (this also applies to potential prey items). The same assumption is made for fish and aquatic-phase CRLF.

The acute measures of effect used for animals in this screening level assessment are the LD₅₀, LC₅₀ and EC₅₀. LD stands for "Lethal Dose", and LD₅₀ is the amount of a material, given all at once, that is estimated to cause the death of 50% of the test organisms. LC stands for "Lethal Concentration" and LC₅₀ is the concentration of a chemical that is estimated to kill 50% of the test organisms. EC stands for "Effective Concentration" and the EC₅₀ is the concentration of a chemical that is estimated to produce a specific effect in 50% of the test organisms. Endpoints for chronic measures of exposure for listed and non-listed animals are the NOAEL/NOAEC and NOEC. NOAEL stands for "No Observed-Adverse-Effect-Level" and refers to the highest tested dose of a substance that has been reported to have no harmful (adverse) effects on test organisms. The NOAEC (*i.e.*, "No-Observed-Adverse-Effect-Concentration") is the highest test concentration at which none of the observed effects were statistically different from the control. The

NOEC is the No-Observed-Effects-Concentration. For non-listed plants, only acute exposures are assessed (*i.e.*, EC₂₅ for terrestrial plants and EC₅₀ for aquatic plants).

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

2.10.3 Measures of Risk

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

For this endangered species assessment, listed species LOCs are used for comparing RQ values for acute and chronic exposures of atrazine directly to the CRLF and the DS. If estimated exposures directly to the assessed species of atrazine resulting from a particular use are sufficient to exceed the listed species LOC, then the effects determination for that use is "may affect". When considering indirect effects to the assessed species due to effects to prey, the listed species LOCs are also used. If estimated exposures to the prey of the assessed species of atrazine resulting from a particular use are sufficient to exceed the listed species LOC, then the effects determination for that use is a "may affect." If the RQ being considered also exceeds the non-listed species acute risk LOC, then the effects determination is a LAA. If the acute RQ is between the listed species LOC and the non-listed acute risk species LOC, then further lines of evidence (*i.e.* probability of individual effects, species sensitivity distributions) are considered in distinguishing between a determination of NLAA and a LAA. If the RQ being considered for a particular use exceeds the non-listed species LOC for plants, the effects determination is "may affect". Further information on LOCs is provided in **Appendix D**.

2.10.4 Differences in Analysis Plan from Previous Assessments

In its February 11, 2008 non-concurrence letter for the atrazine Alabama sturgeon and dwarf wedgemussel (DWM) effects determinations, the U.S. Fish and Wildlife Service (USFWS) commented on the Agency's use of the Comprehensive Aquatic Systems Model (CASM), stating that the model may not be conservative enough in its estimation

of adverse effects thresholds to adequately predict effects to listed species. Additional concerns expressed by USFWS related to CASM include issues with false negatives, omission of mesocosm data with Brock scores of “2”, and parameterization of the model with input variables not specific to the geographic areas being assessed.

A Scientific Advisory Panel (SAP) was convened in December 2007 to discuss the use of CASM to interpret targeted atrazine monitoring data relative to the robust set of micro/mesocosm data available for atrazine. In its 2007 report on the proceedings of the SAP meeting, the SAP commended the Agency on its use of community-level modeling; however, the Panel expressed concern with issues of model validation, transparency, and the analysis of sensitivity. In order to address the recommendations of the SAP relative to CASM, the Agency is currently working on a number of tasks, including a Quality Assurance (QA) evaluation, an expanded sensitivity analysis, and validation of the model as a tool for interpreting monitoring data.

Given the ongoing work to address the recommendations of the SAP regarding CASM, the Agency acknowledges that the threshold concentrations used to evaluate potential indirect effects to listed species via community-level effects to aquatic plants may change. The threshold concentrations used in the atrazine effects determinations were based on exposures relative to the atrazine micro/mesocosm studies that were unlikely to exceed a CASM-based index. Pending further work to address concerns related to QA evaluation, sensitivity analysis, and validation of CASM, the Agency will not rely on a CASM-based index for this assessment, but will instead use results from the laboratory aquatic plant studies and the thresholds for community level effects that were described in the atrazine IRED (U.S. EPA, 2003). However, once the further work on CASM and any necessary subsequent modification of threshold concentrations is completed, the Agency may revisit the effects determinations presented in this assessment.

3. Exposure Assessment

3.1 Label Application Rates and Intervals

Atrazine labels may be categorized into two types: labels for manufacturing uses (including technical grade atrazine and its formulated products) and end-use products. Technical products, which contain atrazine of high purity, are not used directly in the environment, but instead are used to make formulated products, which can be applied in specific areas to control weeds. The formulated product labels legally limit atrazine’s potential use to only those sites that are specified on the labels and under the conditions of use (rate, timing, etc.) specified on the label.

In the January and October 2003 IREDs (U.S. EPA, 2003a and b), EPA stipulated numerous changes to the use of atrazine including label restrictions and other mitigation measures designed to reduce risk to human health and the environment. Specifically pertinent to this assessment are provisions of a Memorandum of Agreement (MOA) between the Agency and atrazine registrants. In the MOA, the Agency stipulated that certain label changes must be implemented on all manufacturing-use product labels for

atrazine and on all end-use product labels for atrazine prior to the 2005 growing season. These label changes included cancellation of certain uses, reduction in application rates, and requirements for harmonization across labels including setbacks from waterways. Specifically, the label changes prohibit atrazine use within 50 feet of sinkholes, 66 feet of intermittent and perennial streams, and 200 feet of lakes and reservoirs.

While these setbacks were required to reduce atrazine deposition to water bodies as a result of spray drift, it is expected that they will also result in a reduction in loading due to runoff across the setback zone; however, current models do not address this reduction relative to runoff quantitatively. Therefore, these restrictions as they relate to runoff are not quantitatively evaluated in this assessment. Table 3.1 provides a summary of label application rates for atrazine uses evaluated in this assessment.

Currently registered non-agricultural uses of atrazine within the action area include residential areas such as playgrounds and home lawns, turf (golf courses and recreational fields), and forestry. Agricultural uses within the action area include corn, sorghum, macadamia nuts, and guava. Other agricultural and non-agricultural uses not present in the action area (due to label restrictions or lack of crops present) include rights of way, fallow/idle⁶, and Conservation Reserve Program (CRP).

Atrazine is formulated as liquid, wettable powder, dry flowable, and granular formulations typically applied pre-emergent. Application methods for the agricultural uses include ground application (the most common application method), aerial application, band treatment, and incorporated treatment; and application using various sprayers (low-volume, hand held, directed) for liquids, and spreaders for granulars. Risks from ground boom and aerial applications are considered in this assessment because they are expected to result in the highest off-target levels of atrazine due to generally higher spray drift levels. Ground boom and aerial modes of application tend to use lower volumes applied in finer sprays than applications coincident with sprayers and spreaders, and thus have a higher potential for off-target movement via spray drift. Although sugarcane is a labeled use that is included in Table 3.1, it is only grown in a small area in Imperial County. EECs from this use were not used to calculate RQs because the CRLF and DS habitat do not overlap with sugarcane growing areas in California.

⁶ Fallow or idle land is defined by the Agency as arable land not under rotation that is set at rest for a period of time ranging from one to five years before it is cultivated again, or land usually under permanent crops, meadows or pastures, which is not being used for that purpose for a period of at least one year. Arable land, which is normally used for the cultivation of temporary crops, but which is temporarily used for grazing, is also included.

Table 3.1 Atrazine Label Application Information for the CRLF & DS Assessment^a

Scenario	Maximum Application Rate (lbs a.i./acre)	Maximum Number of Annual Applications	Formulation	Method of Application	Interval Between Applications
Forestry	4.0	1	Liquid	Aerial and Ground	NA
Residential	2.0	2	Granular	Ground	30 days
	1.0	2	Liquid	Ground	30 days
Corn	2.5	2	Liquid	Ground and Aerial	30 days
Sorghum	2.0	1	Liquid	Ground and Aerial	NA
Macadamia Nuts	4.0	2	Liquid	Ground	30 days
Guava	4.0	2	Liquid	Ground and Aerial	120 days
Turf	2.0	2	Granular	Ground	30 days
	1.0	2	Liquid	Ground	30 days
Sugarcane ^b	4.0	3	Liquid	Ground	30 days
<p>a - Based on 2003 IRED and Label Change Summary Table memorandum dated June 12, 2006 (U.S. EPA, 2006b). b – small acreage of sugarcane reported in Imperial county. Not expected to currently overlap CRLF or DS habitat but included for completeness for statewide assessment</p>					

3.2 Aquatic Exposure Assessment

3.2.1 Introduction

The assessment of exposure within the action area is dependent upon a combination of modeling and monitoring data. In accordance with the Overview Document (U.S. EPA, 2004), screening-level exposures were based on modeling which assumes a static water body. Aquatic exposures are quantitatively estimated for all of assessed uses using scenarios that represent high exposure sites for Atrazine use. Each of these sites represents a 10 hectare field that drains into a 1-hectare pond that is 2 meters deep and has no outlet. Exposure estimates generated using the standard pond are intended to

represent a wide variety of vulnerable water bodies that occur at the top of watersheds including prairie pot holes, playa lakes, wetlands, vernal pools, man-made and natural ponds, and intermittent and first-order streams. As a group, there are factors that make these water bodies more or less vulnerable than the standard surrogate pond. Static water bodies that have larger ratios of drainage area to water body volume would be expected to have higher peak EECs than the standard pond. These water bodies will be either shallower or have large drainage areas (or both). Shallow water bodies tend to have limited additional storage capacity, and thus, tend to overflow and carry pesticide in the discharge whereas the standard pond has no discharge. As watershed size increases beyond 10 hectares, at some point, it becomes unlikely that the entire watershed is planted to a single crop, which is all treated with the pesticide. Headwater streams can also have peak concentrations higher than the standard pond, but they tend to persist for only short periods of time and are then carried downstream. More details on the uncertainties associated with the various exposure assessments and modeling scenarios specifically may be found in the Uncertainty Section (Section 6).

Use-specific management practices for all of the assessed uses of Atrazine were used for modeling, including application rates, number of applications per year, application intervals, buffer widths and resulting spray drift values modeled from AgDRIFT, and the first application date for each use. The general conceptual model of exposure for this assessment is that the highest exposures are expected to occur in headwater streams adjacent to agricultural fields and other non-agricultural use sites (residential, right-of-way, turf, and forestry). Many of the streams and rivers within the action area defined for this assessment are in close proximity to both agricultural and non-agricultural uses sites (for this assessment the action area represents the entire state of California).

Available usage data (Kaul, et al., 2005) suggest that the heaviest usage of atrazine relative to the action area is likely to be in the Central Valley although the data may under-represent the non-agricultural uses (e.g. forestry). All existing PRZM scenarios were evaluated, and a subset was selected for use in this assessment. The scenarios were selected to provide a spatial context to predicted exposures.

Currently a suite of 28 PRZM California scenarios are available for use in ecological risk assessments representing predominantly agricultural uses. Of these, 16 were developed specifically for the CRLF assessments, 3 were developed for the OP cumulative assessment, and 9 are standard scenarios. Each scenario is intended to represent a high-end exposure setting for a particular use site. Scenario locations are selected based on various factors including crop acreage, runoff and erosion potential, climate, and agronomic practices. Once a location is selected, a scenario is developed using locally specific soil, climatic, and agronomic data. Each PRZM scenario is assigned a specific climatic weather station providing 30 years of daily weather values.

Specific scenarios were selected for use in this assessment using two criteria. First, an evaluation of all available PRZM scenarios was conducted, and those scenarios that represent atrazine uses (e.g. CA corn) were selected for modeling. Weather information was assigned to these scenarios at development. Second, an additional scenario (LA

sugarcane) was identified to represent the limited use of atrazine on sugarcane in Imperial County for which a scenario within the action area is not available. This scenario was used in the assessment as a surrogate for atrazine use on sugarcane but relied on climatic data from San Diego. The LA sugarcane scenario represents a highly vulnerable site a portion of the country where sugarcane use is high and likely provides an over-estimate of vulnerability to atrazine runoff associated with this very limited (< 1,000 acres) of sugarcane in Imperial County.

The residential scenario is unique in that it was developed to represent a range of homeowner use sites ranging from highly urbanized landscapes to more suburban communities. The approach for modeling residential uses was originally developed for assessment of risk from atrazine use in the Chesapeake Bay ESA and Barton Springs ESA assessments (USEPA 2006a, USEPA 2006b). In these assessments the approach accounted for the presence of impervious surfaces and unintentional overspray onto these surfaces by a paired scenario approach. The residential scenario was developed to represent a typical ¼ acre residential setting and used a curve number developed by USDA that accounted for both the pervious (i.e. turf, ornamental beds, gardens) and impervious surfaces (i.e. sidewalk, driveway, rooftop) contained within that lot. A second, impervious surface scenario, was developed to represent the other impervious surfaces within a watershed (i.e. streets, parking lots). What is clear from previous evaluations is that modeling paired scenarios with limited overspray to the impervious scenario (e.g. < 10% of the application rate) yielded lower overall exposure due to the increased runoff volume derived from the impervious scenario. This assessment includes modeling with the residential scenario alone representing more suburbanized settings and the paired residential/impervious approach representing more urbanized settings. In general, the paired approach (assuming 1% overspray to impervious surfaces) yields exposure concentrations roughly 30% of those for the residential scenario alone. More detail on the approach can be found in the previous assessments (USEPA 2006a, USEPA 2006b).

Further description (metadata) and copies of the existing PRZM scenarios may be found at the following websites.

<http://www.epa.gov/oppefed1/models/water/index.htm#przmexamsshell>

<http://www.epa.gov/oppefed1/models/water/przmenvironmentdisclaim.htm>

A summary of all the modeled scenarios along with associated weather information is included in Table 3.2. Both the agricultural and non-agricultural scenarios were used within the standard framework of PRZM/EXAMS modeling using the standard graphical user interface (GUI) shell, PE5.pl. The models and GUI used in this assessment may be found at the following website:

<http://www.epa.gov/oppefed1/models/water/index.htm>

Table 3.2 Summary of PRZM Scenarios

Use	Scenario	First Application	Weather Station (WBAN #)
Corn	CAcornOP	March 1	Sacramento (23232)
Sorghum	CAwheatRLF	June 1	Fresno (93193)
Residential	CAresidentialRLF	February 1	San Francisco (23234)
Forestry	CAforestryRLF	December 1	Eureka (23483)
Turf	CAturfRLF	February 1	San Francisco (23234)
Macadamia Nuts	CAalmond_wirrigSTD	March 1	Sacramento (23232)
Guava	CACitrus_wirrigSTD	March 1	Bakersfield (23155)
Sugarcane	LA sugarcaneSTD	March 28	San Diego (23188)

3.2.2 Model Inputs

The estimated concentrations from surface water sources were calculated using Tier II PRZM (Pesticide Root Zone Model) and EXAMS (Exposure Analysis Modeling System). PRZM is used to simulate pesticide transport as a result of runoff and erosion from a standardized watershed, and EXAMS estimates environmental fate and transport of pesticides in surface waters. The linkage program shell (PE5.pl) that incorporates the site-specific scenarios was used to run these models.

Scenarios used in this assessment consist of four California specific scenarios developed for uses being assessed (corn, residential, forestry, and turf), two California specific scenarios as surrogate for an atrazine use (almonds as surrogate for macadamia nuts and citrus as surrogate for guava), and one scenario (Louisiana sugarcane) developed from another geographic area. All scenarios were modeled using local weather data selected to represent the highest rainfall potential in a region as described above. Linked use site-specific scenarios and meteorological data were used to estimate exposure as a result of specific use for each modeling scenario. The PRZM/EXAMS model was used to calculate concentrations using the standard ecological water body scenario in EXAMS. Weather and agricultural practices were simulated over 30 years so that the 1 in 10 year exceedance probability at the site was estimated for the standard ecological water body.

One outcome of the 2003 IRED process was a modification to all existing atrazine labels that requires setback distances around intermittent/perennial streams and lakes/reservoirs. The label changes specify spray drift buffers of 66 feet and 200 feet for atrazine applications surrounding intermittent/perennial streams and lakes/reservoirs, respectively. The Agency incorporated these distances into this assessment and has modified the

standard spray drift assumptions accordingly using AgDrift to estimate the impact of a setback distance of 66 feet on the fraction of drift reaching a surface water body. The revised spray drift percentages, which are incorporated into the PRZM/EXAMS modeling, are 0.6% for ground applications and 6.5% for aerial applications. The DS uses habitat consistent with the water body types targeted for the 66 foot buffer, while in general the CRLF uses both flowing and static water as habitat. However, the 200 foot buffer is intended to protect larger water body types used as drinking water sources while the CRLF uses smaller static ponds. Thus, the 66 foot buffer has been utilized for exposure modeling because it is believed to be more representative of CRLF habitat and it yields a more protective (i.e. higher) exposure estimate. A more complete discussion of the CRLF habitats is included in Attachment 1.

Models to estimate the effect of setbacks on load reduction for runoff are not currently available. It is well documented that vegetated setbacks can result in a substantial reduction in pesticide load to surface water (USDA, NRCS, 2000). Specifically for atrazine, data reported in the USDA study indicate that well vegetated setbacks have been documented to reduce atrazine loading to surface water by as little as 11% and as much as 100% of total runoff compared to the loading without a setback. It is expected that the presence of a well-vegetated setback between the site of atrazine application and receiving water bodies would result in reduction in loading. Therefore, the aquatic EECs presented in this assessment are likely to over-estimate exposure in areas with well-vegetated setbacks.

The date of first application was developed based on several sources of information including data provided by BEAD and Crop Profiles maintained by the USDA. In general, the date of first application was selected to represent the most vulnerable window of exposure. Typical use patterns for atrazine as a pre-emergent herbicide show that the majority of first applications occur during the spring planting/emergence. For example, the distribution of Atrazine applications to corn sites from the CDPR PUR data for 2004 to 2006 was used to pick a March 1 application date is shown in **Figure 3.1** which shows a representative distribution from 2006.

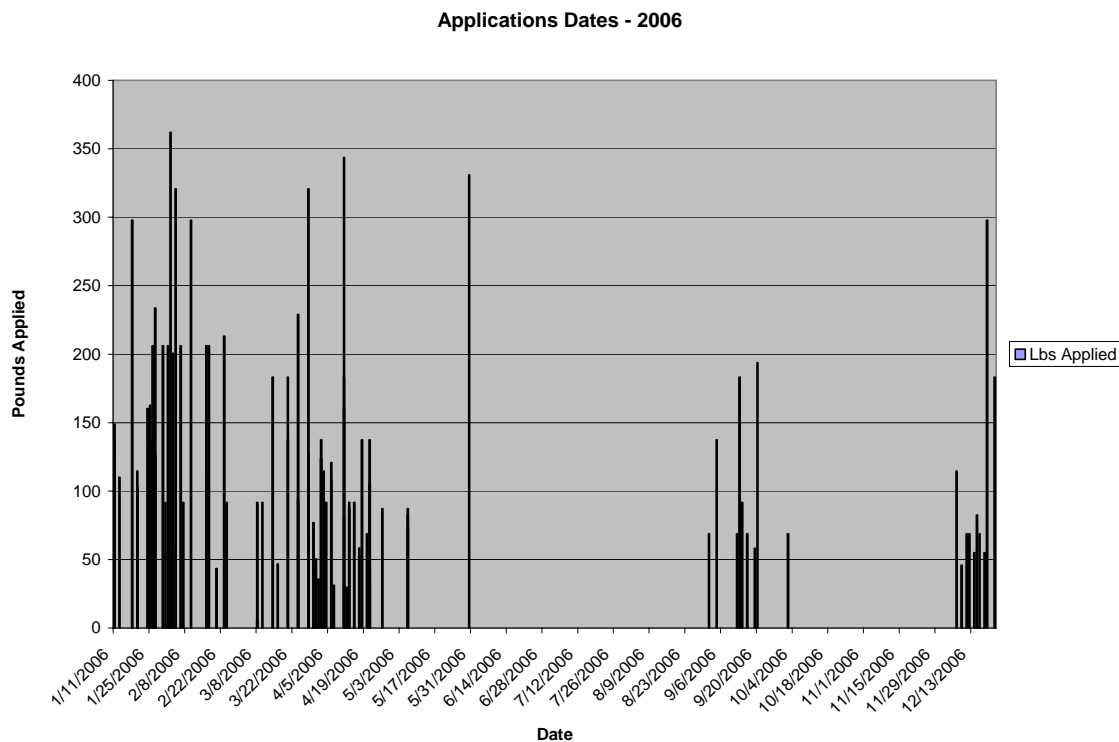


Figure 3.1 Application of Atrazine to Corn in 2006 from CDPR PUR data

More detail on the crop profiles and the previous assessments may be found at:

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

The appropriate PRZM input parameters were selected from the environmental fate data submitted by the registrant and in accordance with US EPA-OPP EFED water model parameter selection guidelines, Guidance for Selecting Input Parameters in Modeling the Environmental Fate and Transport of Pesticides, Version 2.3, February 28, 2002. These parameters are consistent with those used in both the 2003 IRED (U.S. EPA, 2003a) and the cumulative triazine risk assessment (U.S. EPA, 2006a) and are summarized in Table 3.3. More detail on these assessments may be found at:

http://www.epa.gov/oppsrrd1/REDs/atrazine_ired.pdf

http://www.epa.gov/pesticides/cumulative/common_mech_groups.htm#chloro

Table 3.3 Summary of PRZM/EZAMS Environmental Fate Data Used for Aquatic Exposure Inputs for Atrazine

Fate Property	Value	MRID ^a (or source)
Molecular Weight	215.7	MRID 41379803
Henry's constant	2.58 x10 ⁻⁹	MRID 41379803
Vapor Pressure	3 x 10 ⁻⁷	MRID 41379803
Solubility in Water	33 mg/l	MRID 41379803
Photolysis in Water	335 days	MRID 42089904
Aerobic Soil Metabolism Half-lives	152 days	MRID 40431301 MRID 40629303 MRID 42089906
Hydrolysis	stable	MRID 40431319
Aerobic Aquatic Metabolism (water column)	304 days	2x aerobic soil metabolism rate constant
Anaerobic Aquatic Metabolism (benthic)	608 days	MRID 40431323
Koc	88.78 ml/g	MRID 40431324 MRID 41257901 MRID 41257902 MRID 41257904 MRID 41257905 MRID 41257906
Application Efficiency	95 % for aerial 99 % for ground	Default value ^b
Spray Drift Fraction	6.5 % for aerial 0.6 % for ground	AgDrift adjusted values based on label restrictions
^a Master Record Identification (MRID) is record tracking system used within OPP to manage data submissions to the Agency. Each data submission is given a unique MRID number for tracking purposes. ^b Inputs determined in accordance with EFED "Guidance for Chemistry and Management Practice Input Parameters for Use in Modeling the Environmental Fate and Transport of Pesticides" dated February 28, 2002.		

3.2.3 Results

In general, these EECs show a pattern of exposure for all durations that is influenced by the persistence of the compound and the lack of flow through the static water body. Predicted atrazine concentrations, though high across durations of exposure for a single year, do not increase across the 30-year time series; therefore, accumulation is not a concern. The resulting EECs are summarized in Table 3.4.

Table 3.4 Aquatic EECs (µg/L) for Atrazine Uses in California

Use Site	Application Rate (lbs a.i./acre)	No. of Applications	Peak EEC	21-day average EEC	60-day average EEC
Corn	2.0, 0.5	2 (not to exceed 2.5 lbs a.i./year)	54.2	53.4	52.8
Sorghum	2.0	1	16.9	16.4	15.5
Residential Granular	2.0, 2.0	2 (not to exceed 4.0 lbs a.i./year)	26.6	26.4	26.2
Paired Residential/Impervious Granular ¹	2.0, 2.0	2 (not to exceed 4.0 lbs a.i./year)	8.7	8.7	8.6
Residential Liquid	1.0, 1.0	2 (not to exceed 2.0 lbs a.i./year)	17.8	17.7	17.6
Paired Residential/Impervious Liquid ¹	1.0, 1.0	2 (not to exceed 2.0 lbs a.i./year)	5.4	5.4	5.4
Forestry	4.0	1	55.7	55.0	53.8
Turf Granular	2.0, 2.0	2 (not to exceed 4.0 lbs a.i./year)	34.1	33.7	33.0
Turf Liquid	1.0, 1.0	2 (not to exceed 2.0 lbs a.i./year)	23.1	22.8	22.3
Macadamia Nut	4.0, 4.0	2	50.3	49.7	48.3
Guava	4.0, 4.0	2	63.5	62.3	60.1

Use Site	Application Rate (lbs a.i./acre)	No. of Applications	Peak EEC	21-day average EEC	60-day average EEC
Sugarcane ²	4.0, 3.0, 3.0	3	147.9	144.9	141.9

1 – Paired Residential/Impervious EEC represents a post processed blending of time series PRZM/EXAMS output assuming 50% of the residential lot is treated (turf portion only) and 50% of the 10 ha watershed is impervious with 1% overspray

2 – sugarcane use is limited to Imperial county and is considered to be outside the range of CRLF and DS habitat

3.2.4 Existing Monitoring Data

A critical step in the process of characterizing EECs is comparing the modeled estimates with available surface water monitoring data. Included in this assessment are Atrazine data from the USGS NAWQA program (<http://water.usgs.gov/nawqa>) and data from the California Department of Pesticide Regulation (CDPR). In addition, atmospheric monitoring data for Atrazine are summarized as well as a variety of atrazine monitoring data from the open literature.

3.2.4.1 USGS NAWQA Surface Water Data

Data from the USGS NAWQA website for atrazine occurrence in surface water in California were obtained on November 5, 2008. A total of 2084 surface water samples were analyzed for atrazine spanning a period from 1992 to 2007. Of these, a total of 891 samples detected atrazine (frequency of detection of 43%) though 408 of these were estimated below the limit of quantitation (LOQ). The maximum concentration detected was 0.274 ppb from Stanislaus county in 2000. The next highest detection was 0.13 ppb from 1993. All other detections were less than 0.1 ppb.

3.2.4.2 USGS NAWQA Groundwater Data

Data from the USGS NAWQA website for atrazine occurrence in groundwater in California were obtained on November 5, 2008. A total of 751 groundwater samples were analyzed for atrazine spanning a period from 1993 to 2006. Of these, a total of 219 samples detected atrazine (frequency of detection of 29%) though 76 of these were estimated below the LOQ. The maximum concentration detected was 0.472 ppb from Sacramento county in 1996. The next highest detection was 0.146 ppb from 2001. Of all samples, only 5 were greater than 0.1 ppb.

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

Data from the CDPR surface water monitoring database website for atrazine occurrence were obtained on November 5, 2008. A total of 3722 surface water samples were analyzed for atrazine spanning a period from 1991 to 2006. Of these, a total of 393 samples detected atrazine (frequency of detection of 11%). Unlike the USGS NAWQA data, the supporting information on LOQ are less clear although it appears that 266 of the

detections were below the LOQ. The maximum concentration detected was 0.74 ppb from San Joaquin county in 2005. The next highest detection was 0.44 ppb from 2005. All other detections were less than 0.1 ppb. Of all samples, only 13 were greater than 0.1 ppb.

3.2.4.4 Atmospheric Monitoring Data

Available monitoring data for atrazine in air and rainfall were evaluated to provide context to the evaluation of the extent of action area and estimated concentrations in surface water. Atrazine enters the atmosphere via volatilization and spray drift and is aerially deposited (a source of importance to some water bodies). Based on the available information (Majewski et al., 2000; Majewski and Capel, 1995; Capel et al., 1994, McConnell, et al, 2004, Kuang, et al, 2003, Foreman, et al, 1999, Dubus, et al, 2000, Ushenko, et al, 2005, CDPR, 2001), atrazine has been detected in rainwater and air samples across the United States as high as 11 ug/l (Scribner et al, 2005). In general, atrazine has been detected in some studies at variable frequency of detections but in general, detections in rainfall have been below 5 µg/L (Makjewski, et al 2002, Kuang, et al, 2003, Dubus, et al, 2000). Often these studies are non-targeted to atrazine use and there is a lack of ancillary data in these studies to determine whether these detections are due to spray drift or longer-range transport due to volatilization. However, given that most of the studies focus on major agricultural locations, that atrazine has not been detected in any of the studies conducted at higher elevations, coupled with the relatively low volatility of atrazine, it is expected that many of these detections are reflective of near field (spray drift) exposure and are not indicative of long-range transport. The concentrations detected in the reviewed studies suggest that transport of atrazine via atmospheric transport will yield exposures well below those predicted by modeling described above.

Specifically, atrazine concentrations in rainfall have been measured up to 3.5 ug/L in Germany (Braun *et al.*, 1987). In 1990-1991, the 95th and 99th percentile atrazine levels in rainfall in the mid-west were reported to be 0.42 and 1.0 ug/L, respectively (USGS Fact Sheet FS-181-97). Capel *et al.* (1994) reported the frequency of detections and pesticide levels in rainfall from 1991 to 1993 in Minnesota; in 1991, atrazine was detected in 2 % of the samples with a maximum concentration of 0.82 ug/L, in 1992 it was 18 percent and 2.2 ug/L, and in 1993 it was 71 % and 2.9 ug/L. Subsequent 1994 monitoring data from 6 Minnesota sites around the state found detections in 93% of the samples (range: 86 - 100%) and a maximum level of 2.8 ug/L (range of maximum levels: 0.74 - 2.8 ug/L). Atrazine concentrations in rainfall monitored in the Lake Michigan study ranged from ND to about 400 ng/L. At one Lake site, much higher atrazine levels were believed to be an outlier (Russell Kreis, e-mail on 11/07/2000).

The analysis in the IRED also documents the occurrence of atrazine in the atmosphere. The data indicate that atrazine can enter the atmosphere via volatilization and spray drift. The data also suggest that atrazine is frequently found in rain samples and tends to be seasonal, related to application timing. Finally, the data suggest that although frequently detected, atrazine concentrations detected in rain samples are less than those seen in the

monitoring data and modeling conducted as part of this assessment and support the contention that runoff and spray drift are the principal routes of exposure. More details on these data can be found in the 2003 IRED (U.S. EPA, 2003a).

3.2.4.5 Summary of Open Literature Sources of Monitoring Data for Atrazine

Atrazine is likely to be persistent in ground water and in surface waters with relatively long hydrologic residence times (such as in some reservoirs) where advective transport (flow) is limited. The reasons for atrazine's persistence are its resistance to abiotic hydrolysis and direct aqueous photolysis, its only moderate susceptibility to biodegradation, and its limited volatilization potential as indicated by a relatively low Henry's Law constant. Atrazine has been observed to remain at elevated concentrations longer in some reservoirs than in flowing surface water or in other reservoirs with presumably much shorter hydrologic residence times in which advective transport (flow) greatly limits its persistence.

A number of open literature studies cited in the 2003 IRED (U.S. EPA, 2003a), document the occurrence of atrazine and its degradates in both surface water and groundwater. These data support the general conclusion that higher exposures tend to occur in the most vulnerable areas in the Midwest and South and that the most vulnerable water bodies tend to be headwater streams and water bodies with little or no flow.

3.2.4.6 Miscellaneous Drinking Water Monitoring Data Derived from Surface Water

A number of national surface water data sets were evaluated as part of the 2003 IRED. Included in that analysis were data from Acetochlor Registration Partnership (ARP) Monitoring Study, the Novartis Population Linked Exposure (PLEX) Database, the USGS 1992-1993 Study of 76 Mid-Western Reservoirs (USGS Open File Report 96-393), the USGS 1989-1990 Reconnaissance Study of Mid-Western Streams (USGS Open File Report 93-457), the USGS 1994-1995 Reconnaissance Study of Mid-Western Streams (USGS Open File Report 98-181), the USGS 1990-1992 Study of 9 Mid-Western Streams (USGS Open File Report 94-396), USGS NAWQA data available in 2002, as well as numerous open literature studies. In general, these data show a pattern of atrazine exposure in various water body types (streams vs. reservoirs), collected with a variety of study objectives (human health vs. ecological health) consistent with those summarized previously in this assessment. The maximum reported concentration from the studies (excluding open literature) was 108 µg/L from the USGS study (Open File Report 93-457) for Mid-Western Streams sampled between 1989 and 1990. Atrazine exposure in rivers, streams, lakes, and reservoirs documented in the open literature cited in the 2003 IRED were consistent with these results with no concentrations above 100 µg/L (except edge of field runoff concentrations in mg/l range which were reported as diluted to µg/L ranges when reaching surface water bodies). In addition, the 2003 IRED summarized reports from the Agency's 6(a)(2) incident database and found the highest concentration at 62 µg/L.

More detail on the individual studies and analysis of the data may be found in the 2003 IRED at the following website:

http://www.epa.gov/oppsrrd1/reregistration/atrazine/efed_redchap_22apr02.pdf

Subsequent to the completion of the 2003 IRED, additional monitoring data from surface water sources used for drinking water were submitted to the Agency for review. Atrazine monitoring results from 2003 to 2005 were collected as part of the Atrazine Monitoring Program (AMP) for purposes of assessing dietary risk for human health. In this study, data were collected from over 100 community water systems (CWS) in 10 states. Monitoring was weekly through the growing season (generally April through July) with biweekly monitoring for the rest of the year. Both raw and finished water were monitored. In general, the results were consistent with those discussed above, with maximum detected concentrations of 33.1 µg/L in 2002, 39.7 µg/L in 2004, and 84.8 µg/L in 2005.

3.2.5 Impact of Typical Usage Information on Exposure Estimates

A final piece of the exposure characterization includes an evaluation of usage information. Label application information was provided by EPA's Biological and Economic Analysis Division and was previously summarized in Table 2.3. This information suggests that atrazine use on corn and forestry (the two highest uses in the CDPR PUR data) is typically 1.5 lbs a.i./acre and 3.2 lbs a.i./acre in California based on CDPR PUR data. This suggests that if typical application rates were used, atrazine exposures would be reduced below those modeled with the label maximum application rate by 25% for corn and 20% for forestry. The other uses exhibited similar reductions when considering typical application rates. Typically usage information is not incorporated into these assessments, but does provide context to the exposures predicted. Caution is used when evaluating "typical" application rate information because this represents the average of all reported applications and thus roughly 50% of the time higher application rates are being applied.

3.3. Terrestrial Animal Exposure Assessment

T-REX (Version 1.4.1) is used to calculate dietary and dose-based EECs of atrazine for birds (surrogate for reptiles and terrestrial phase amphibians), mammals, and terrestrial invertebrates. T-REX simulates a 1-year time period. For this assessment, spray and granular applications of atrazine are considered as discussed below. Terrestrial EECs were derived for the uses previously summarized in **Table 3.4**.

Terrestrial estimated environmental concentrations (EECs) of atrazine to which birds and mammals may be exposed were estimated based on the highest value measured for the foliar dissipation half-life from application of atrazine to turf in several locations throughout the southeastern United States as described in the IRED for atrazine (U.S.

EPA, 2003). These foliar dissipation half-lives are most representative of atrazine used as a postemergent herbicide applied directly to foliage of target plants. Atrazine is, however, used predominantly during crop pre-planting and pre-emergence and is under these circumstances applied directly to soil rather than to foliage. As a result, EECs based on foliar dissipation half-life data, although indicative of post-emergent applications, may not be truly representative of pre-plant and pre-emergence applications. Acute risks to mammals and birds were assessed from EECs that were based on the maximum foliar dissipation half-life of 17 days obtained from foliar dissipation studies conducted in the southeastern United States as described in U.S. EPA (2003). Upper-bound Kenaga nomogram values reported by T-REX are used for derivation of dietary EECs for the terrestrial phase CRLF and their potential prey. Potential direct acute and chronic effects of atrazine to the terrestrial-phase CRLF are derived by considering oral exposures modeled in T-REX for a small bird (20g) consuming small invertebrates. Potential impacts to mammalian prey base was evaluated in T-REX for a small mammal (15 g) consuming short grass. Resulting dietary-based EECs (mg/kg-food) and dose-adjusted EECs (mg/kg-bw) are summarized in **Table 3.5**.

Table 3.5. Upper-bound Kenaga Nomogram EECs for Dietary- and Dose-based Exposures of the CRLF and its Prey to Atrazine

Use	EECs for CRLF (small birds used as surrogate)		EECs for Prey (small mammals)	
	Dietary-based EEC (ppm)	Dose-based EEC (mg/kg-bw)	Dietary-based EEC (ppm)	Dose-based EEC (mg/kg-bw)
Residential and Turf 1 lb a.i./Acre, 2 applications, 30-day interval (spray)	170	200	310	300
Sorghum and Corn ^a , 2 lbs a.i./Acre, 1 application	270	310	480	458
Forestry 4 lbs a.i./Acre, 1 application	540	620	960	920
Macadamia nuts 4 lbs a.i./Acre, 2 applications, 30-day interval	700	795	1240	1180
Guava 4 lbs a.i./Acre, 2 applications, 120-day interval	540	620	970	920

^a Corn application was modeled using a single application of 2 lbs a.i./Acre. Although a second application of 0.5 lbs a.i./Acre could be applied 30 days later, the peak EEC would occur immediately after the first application of 2 lbs a.i./Acre.

For granular formulations (residential and turf, 2 lbs a.i./Acre), an LD50/sq. ft analysis was performed to evaluate potential risks to birds and mammals. The exposure used in this analysis is the mass of atrazine applied to a square foot area (mg/sq. ft). Based on an application rate of 2 lbs a.i./Acre (maximum granular application rate), the exposure value used in the LD50/sq. ft. analysis is 21 mg/sq. ft.

3.3.1. Potential Exposure to Terrestrial Invertebrates

T-REX is also used to calculate EECs for terrestrial insects exposed to atrazine. Dietary-based EECs calculated by T-REX for small and large insects (units of a.i./g) are used to bound an estimate of exposure to bees (used as a surrogate for terrestrial invertebrates) (Table 3.6). Available acute contact toxicity data for bees exposed to atrazine (in units of μg a.i./bee), are converted to μg a.i./g (of bee) by multiplying by 1 bee/0.128 g. The EECs are compared to the acute contact toxicity data for bees in order to derive RQs.

Table 3.6. EECs (ppm) for Indirect Effects to the Terrestrial-Phase CRLF via Effects to Terrestrial Invertebrate Prey Items

Use	Small Insect	Large Insect
Residential and Turf (spray) 1 lb a.i./Acre, 2 applications, 30-day interval	170	19
Sorghum and Corn ^a 2 lbs a.i./Acre, 1 application	270	30
Forestry 4 lbs a.i./Acre, 1 application	540	60
Macadamia nuts 4 lbs a.i./Acre, 2 applications, 30-day interval	700	78
Guava 4 lbs a.i./Acre, 2 applications, 120-day interval	540	60

^a Corn application was modeled using a single application of 2 lbs a.i./Acre. Although a second application of 0.5 lbs a.i./Acre could be applied 30 days later, the peak EEC would occur immediately after the first application of 2 lbs a.i./Acre.

In addition, a number of toxicity studies are available that evaluated effects to terrestrial invertebrates. These studies vary in their application methods. However, the available studies typically report EECs either in terms of application rate (e.g., lbs a.i./Acre) or concentration in soil, which can be converted to application rate by making assumptions regarding soil depth and soil density. For example, assuming a soil depth of 3 cm and a soil density of 1.3 g/cm³, application rates of 1 lb to 4 lbs a.i./Acre would be associated with soil concentrations of 2.9 ppm to 11 ppm. Therefore, the currently approved application rates serve as the EEC for use in risk estimation and range from 1 lb a.i./Acre to 4 lbs a.i./Acre.

3.4 Terrestrial Plant Exposure Assessment

Terrestrial plants in riparian areas may be exposed to atrazine residues carried from application sites via surface water runoff or spray drift. Exposures can occur directly to seedlings breaking through the soil surface and through root uptake or direct deposition onto foliage to more mature plants. Riparian vegetation is important to the water and stream quality of the assessed species because it serves as a buffer and filters out sediment, nutrients, and contaminants before they enter the watersheds associated with the assessed species' habitat. Riparian vegetation has been shown to be essential in the maintenance of a stable stream (Rosgen, 1996). Destabilization of the stream can have an adverse effect on habitat quality by increasing sedimentation within the watershed.

Concentrations of atrazine on the riparian vegetation were estimated using OPP's TerrPlant model (U.S. EPA, 2007d; Version 1.2.2), considering use conditions likely to occur in the watersheds associated with the action area. The TerrPlant model evaluates exposure to plants via runoff and spray drift and is EFED's standard tool for estimating exposure to non-target plants. The runoff loading of TerrPlant is estimated based on the solubility of the chemical and assumptions about the drainage and receiving areas. As previously discussed in Section 3.2.2 (model inputs), the standard spray drift assumptions were modified using AgDrift to estimate the impact of a setback distance of 66 feet on the fraction of drift reaching a surface water body. These revised spray drift percentages were also incorporated into the TerrPlant model, assuming that non-target terrestrial plants adjacent to atrazine use sites would receive the same percentage of spray drift as an adjacent surface water body. The revised spray drift percentages are 0.6% for ground applications and 6.5% for aerial applications.

TerrPlant calculates exposure values for terrestrial plants inhabiting two environments (i.e., dry adjacent areas and semi-aquatic areas). The 'dry, adjacent area' is considered to be representative of a slightly sloped area that receives relatively high runoff and spray drift levels from upgradient treated fields. In this assessment, the 'dry, adjacent area' scenario is used to estimate screening-level exposure values for terrestrial plants in riparian areas. The 'semi-aquatic area' is considered to be representative of depressed areas that are ephemerally flooded, such as marshes.

The following input values were used to estimate terrestrial plant exposure to atrazine from all uses: solubility = 33 ppm; minimum incorporation depth = 1 (TerrPlant default for incorporation depths \leq 1 inch; from product labels); application methods: ground boom, aerial, and granular (from product labels). Application rates ranging from 1 lbs. a.i./Acre to 4 lbs a.i./Acre were modeled.

Terrestrial plant EECs for non-granular and granular formulations are summarized in **Table 3.7**. EECs resulting from spray drift are derived for non-granular applications only.

Table 3.7. Screening-Level Exposure Estimates for Terrestrial Plants to Atrazine

Use/ App. Rate (lbs a.i./acre)	Application Method	Total Loading to Dry Adjacent Areas (lbs a.i./acre)	Total Loading to Semi-Aquatic Areas (lbs a.i./acre)	Drift EEC (lbs a.i./acre)
Forestry, guava, macadamia nuts / 4.0	Aerial	0.34	1.1	0.26
	Ground	0.10	0.82	0.02
Corn and Sorghum / 2.0	Aerial	0.17	0.53	0.13
	Ground	0.05	0.41	0.01
Turf and Residential (granular) / 2.0	Granular	0.04	0.4	NA
Turf and Residential (spray) / 1.0	Ground	0.026	0.21	0.006

4. Effects Assessment

This assessment evaluates the potential for atrazine to directly or indirectly affect the CRLF and the DS or modify designated critical habitat. As previously discussed in Section 2, assessment endpoints for the assessed species include direct toxic effects on survival, reproduction, and growth, as well as indirect effects, such as reduction of the prey base and/or modification of its habitat that in turn affect survival, reproduction or growth. In addition, potential destruction and/or modification of critical habitat are assessed by evaluating potential effects to the PCEs, which are components of the critical habitat areas that provide essential needs to the species, such as water quality and food base (see Section 2.4).

Acute (short-term) and chronic (long-term) toxicity information is characterized based on registrant-submitted studies and a comprehensive review of the open literature on atrazine, consistent with the Overview Document (U.S. EPA, 2004). Potential direct and indirect effects to the CRLF and the DS and potential modification to critical habitat are evaluated in accordance with the methods (both screening and species-specific refinements) described in the Agency's Overview Document (U.S. EPA, 2004).

Other sources of information, including use of the acute probit dose response relationship to establish the probability of an individual effect and reviews of the Ecological Incident Information System (EIIS), are conducted to further refine the characterization of potential ecological effects associated with exposure to atrazine.

A summary of the available aquatic and terrestrial organism ecotoxicity information, use of the probit dose-response relationship, and the incident information for atrazine are provided in the following sections. A summary of the available data directly used in this assessment is presented. A more comprehensive discussion of the available toxicity data is included in Appendix A of this assessment, and additional open literature studies are included in Appendices E and F.

4.1. Ecotoxicity Study Data Sources

Toxicity endpoints are established based on data generated from guideline studies submitted by the registrant and from open literature studies that meet the criteria for inclusion into the ECOTOX database maintained by EPA/Office of Research and Development (ORD) (U.S. EPA, 2004). Open literature data presented in this assessment were obtained from the 2003 atrazine IRED as well as ECOTOX information obtained in queries in September, 2007 and October, 2008. The ECOTOX search included all open literature data for atrazine (i.e., pre- and post-IRED). In order to be included in the ECOTOX database, papers must meet the following minimum criteria:

- the toxic effects are related to single chemical exposure;
- the toxic effects are on an aquatic or terrestrial plant or animal species;
- there is a biological effect on live, whole organisms;

- a concurrent environmental chemical concentration/dose or application rate is reported; and
- there is an explicit duration of exposure.

Meeting the minimum criteria for inclusion in ECOTOX does not necessarily mean that the data are suitable for use in risk estimation. Data that pass the ECOTOX screen are evaluated along with the registrant-submitted data, and may be incorporated qualitatively or quantitatively into this endangered species risk assessment. In general, only effects data in the open literature that are more conservative than the registrant-submitted data are considered. The degree to which open literature data are quantitatively or qualitatively characterized is dependent on whether the information is relevant to the assessment endpoints (i.e., maintenance of survival, reproduction, and growth; alteration of PCEs in the critical habitat impact analysis) identified in the problem formulation. For example, endpoints such as biochemical modifications are not likely to be used to calculate risk quotients unless it is possible to quantitatively link these endpoints with reduction in survival, reproduction, or growth (e.g., the magnitude of effect on the biochemical endpoint needed to result in effects on survival, growth, or reproduction is known). A summary of all accepted open literature is included in Appendix E, and a bibliography of all open literature considered as part of this assessment regardless of whether the data were accepted or rejected by ECOTOX is included in Appendix F. A bibliography of all registrant-submitted data is included in Appendix G.

As described in the Agency's Overview Document (U.S. EPA, 2004), the most sensitive endpoint for each taxa is used for RQ calculation. Tables 4.3 (aquatic organisms) and 4.4 (terrestrial organisms) summarizes the most sensitive ecological toxicity endpoints for the CRLF and the DS and their designated critical habitat based on an evaluation of both the submitted studies and the open literature. Toxicity information used in this assessment is further described in the following sections. Additional information on the available submitted and open literature toxicity studies is provided in Appendix A.

4.2. Toxicity Categories

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

Table 4.1 Categories of Acute Toxicity for Aquatic Organisms

LC/EC ₅₀ (mg/L)	Toxicity Category
< 0.1	Very highly toxic
> 0.1 - 1	Highly toxic
> 1 - 10	Moderately toxic
> 10 - 100	Slightly toxic
> 100	Practically nontoxic

4.3. Toxicity of Chemical Mixtures

As previously discussed in the problem formulation, the available toxicity data show that other pesticides may combine with atrazine to produce synergistic, additive, and/or

antagonistic toxic interactions. The results of available toxicity data for mixtures of atrazine with other pesticides are presented in Appendix A. Interactive effects between atrazine and several organophosphate insecticides including diazinon, chlorpyrifos, and methyl parathion, as well as herbicides including alachlor have been described as more than additive. If atrazine is present in the environment in combination with other chemicals, the toxicity of the mixture may be increased relative to the toxicity of each individual chemical, offset by other environmental factors, or even reduced by the presence of antagonistic contaminants if they are also present in the mixture. The variety of chemical interactions presented in the available data set suggest that the toxic effect of atrazine, in combination with other pesticides used in the environment, can be a function of many factors including but not necessarily limited to (1) the exposed species, (2) the co-contaminants in the mixture, (3) the ratio of atrazine and co-contaminant concentrations, (4) differences in the pattern and duration of exposure among contaminants, and (5) the differential effects of other physical/chemical characteristics of the receiving waters (e.g. organic matter present in sediment and suspended water). Quantitatively predicting the combined effects of all these variables on mixture toxicity to any given taxa with confidence is beyond the capabilities of the available data.

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

Atrazine has registered products that contain multiple active ingredients. Analysis of the available open literature and acute oral mammalian LD₅₀ data for multiple active ingredient products relative to the single active ingredient is provided in Appendix C. The results of this analysis show that an assessment based on the toxicity of the single active ingredient of atrazine is appropriate.

4.4 Toxicity of Atrazine to Aquatic Organisms

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

Table 4.2 Aquatic Toxicity Profile for Atrazine

Assessment Endpoint	Acute/ Chronic	Species	Toxicity Value Used in Risk Assessment (ug/L)	MRID	Comment
Freshwater fish (surrogate for aquatic-phase amphibians)	Acute	Rainbow Trout (<i>O. mykiss</i>)	96 hr LC50: 5300	00024716 (Beliles and Scott, 1965)	Acceptable study Probit slope: 2.7 (95% confidence interval could not be derived)
	Chronic	Brook Trout (<i>Salvelinus fontinalis</i>)	NOAEC = 65 LOAEC = 120	00024377 (Macek et al., 1976)	Acceptable 44-week life-cycle study; 7.2% reduction in length; 16% reduction in weight occurred at the LOAEC
Freshwater invertebrates	Acute	Midge (<i>Chironomus tentans</i>)	LC50: 720	00024377 Macek et al. 1976	Supplemental; 48-hr study, no probit slope could be derived because raw data were not included.
	Chronic	Scud (<i>Gammarus fasciatus</i>)	NOAEC = 60 LOAEC = 120	00024377 (Macek et al., 1976)	Acceptable: 30-day study; 25 % reduction in development of F ₁ to seventh instar at the LOAEC
Estuarine/ marine fish	Acute	Sheepshead Minnow (<i>Cyprinodon variegatus</i>)	LC50: 2000	Hall et al . 1994 (MRID 45208303)	Acceptable study; Probit slope was 4.4 ^a (95% CI: 2.8 – 5.9)
	Chronic	Sheepshead Minnow (<i>Cyprinodon variegatus</i>)	NOAEC: 1900 LOAEC: 3400	Ward & Ballantine 1985 (MRID 45202920)	89% reduction in juvenile survival was observed at the LOAEC of 3400 µg/L.
Estuarine/ marine invertebrates	Acute	Saltwater invertebrate, Copepod (<i>Acartia tonsa</i>)	LC50: 88 Probit Slope: 0.95	Thursby et al., 1990 (MRID 45202918)	Data used as initial screen to assess indirect effects to listed species from reduction of animal food supply.
	Chronic	Saltwater invertebrate, Mysid shrimp (<i>Americamysis bahia</i>)	NOAEC: 80	Ward & Ballantine, 1985 (MRID 45202920)	37% Reduction in survival occurred at the LOAEC of 190 µg/L. Data used as initial screen to assess indirect effects to listed species from reduction of animal food supply.
	Chronic	Saltwater invertebrates, copepod (<i>A. tonsa</i>)	Estimated NOAEC: 7	None	Estimated using an acute to chronic ratio of 12.5 for mysid shrimp: 1000 ug/L (MRID 45202920) / 80 ug/L (MRID 45202920) = 12.5. Acute LC50 / ACR = Estimated NOAEC; 88 / 12.5 = 7
Aquatic plants	--	Freshwater algae <i>Chlorella vulgaris</i>	7-day EC ₅₀ = 1	00023544 (Torres & O'Flaherty, 1976)	Supplemental study
	--	Duckweed	14-day EC ₅₀ =	43074804	Supplemental study

Assessment Endpoint	Acute/ Chronic	Species	Toxicity Value Used in Risk Assessment (ug/L)	MRID	Comment
		(<i>Lemna gibba</i>)	37 µg/L	(Hoberg, 1993)	

4.4.1 Toxicity to Fish

Fish toxicity data were used to evaluate potential direct effects to aquatic phase CRLF and the DS and indirect effects to the CRLF. A summary of acute and chronic fish and aquatic-phase amphibian data, including data from the open literature, is provided in the following sections. Additional information is included in Appendix A.

4.4.1.1 Acute Exposure (Mortality) Studies

Atrazine toxicity has been evaluated in numerous fish species, and the results of these studies demonstrate a wide range of sensitivities to atrazine. LC50 values range from 2000 to 60,000 µg/L (2 mg/L to 60 mg/L) (see Appendix A for additional details on these studies). Therefore, atrazine is classified as moderately toxic to fish on an acute basis.

Atrazine has been tested in both saltwater and freshwater species. The most sensitive species was used to calculate risk quotients regardless of the salinity environment because the DS enters both freshwater and saltwater environments. Therefore, the lowest LC50 of 2,000 µg/L reported in sheepshead minnows (MRID 45208303), was used for risk quotient calculations for the DS. The most sensitive freshwater LC50 of 5300 ug/L (rainbow trout) was used for RQ calculations for the CRLF.

More sensitive reliable acute LC50 values were not located in the open literature. A 96-hr LC50 was reported in Harlequinfish of 500 ug/L (Ecotox No. 122076). However, it is unclear if atrazine was the test substance in this study. Insufficient information was included in the publication to allow for identification of the test material. Therefore, this value was not used to calculate risk quotients.

4.4.1.2 Chronic Exposure (Growth/Reproduction) Studies

Chronic freshwater fish toxicity studies were used to assess potential direct effects to the DS and aquatic phase CRLFs via potential effects to growth and reproduction. Freshwater fish life-cycle studies for atrazine are available and summarized in Appendix A. Following 44 weeks of exposure to atrazine in a flow-through system, statistically significant reductions in brook trout mean length (7.2%) and body weight (16%) were observed at a concentration of 120 µg/L, as compared to the control (MRID 00024377). The corresponding NOAEC for this study is 65 µg/L. Although the acute toxicity data for atrazine show that rainbow trout are the most sensitive freshwater fish and the sheepshead minnow is the most sensitive saltwater fish species, available chronic studies on these species produced NOAEC values that were above 65 ug/L. Therefore, the

lowest NOAEC was used to estimate potential chronic risks. Further information on chronic fish toxicity data for atrazine is provided in Appendix A.

4.4.1.3 Fish: Sublethal Effects and Additional Open Literature Information

In addition to registrant submitted studies, data were located in the open literature that report sublethal effect levels to freshwater fish at levels lower than the NOAEC of 65 ug/L from the life cycle study described above.

Reported sublethal effects in rainbow trout show increased plasma vitellogenin levels in both female and male fish and decreased plasma testosterone levels in male fish at atrazine concentrations of approximately 50 µg/L (Wieser and Gross, 2002 [MRID 45622304]). Vitellogenin (Vtg) is an egg yolk precursor protein expressed normally in female fish and dormant in male fish. The presence of Vtg in male fish is used as a molecular marker of exposure to estrogenic chemicals. There is a high degree of variability with the Vtg effects in these studies, which confounds the ability to resolve the effects of atrazine on plasma steroids and vitellogenesis.

Effects of atrazine on freshwater fish behavior, including a preference for the dark part of the aquarium following one week of exposure (Steinberg et al., 1995 [MRID 45204910]) and a reduction in grouping behavior following 24-hours of exposure (Saglio and Trijase, 1998 [MRID 45202914]), have been observed at atrazine concentrations of 5 µg/L. In addition, alterations in rainbow trout kidney histology have also been observed at atrazine concentrations of 5 µg/L and higher (Fischer-Scherl et al., 1991 [MRID 45202907]).

In salmon, potentially sensitive endpoints that have been reported included effects on gill physiology and endocrine-mediated olfactory functions. Data from Waring and Moore (2004; ECOTOX No. 72625) suggest that salmon smolt gill physiology, represented by changes in Na-K-ATPase activity, was altered at 2 µg/L atrazine and higher, and Moore et al. (2007) observed similar effects at 0.5 ug/L. Survival was evaluated after transfer to full salinity sea water (33 ‰) in Waring and Moore (2004). Atrazine exposure for 5 to 7 days in freshwater followed by transfer to full salinity sea water (Waring and Moore, 2004) resulted in higher mortality rates at 14 ug/L (14% mortality). In a separate study presented in the same publication, 15% mortality was observed at 1 ug/L. No controls died, and statistical significance was not indicated. As noted in Attachment 3, the DS is euryhaline species (species adapted to living in fresh and brackish water) that occupies estuarine areas with salinities below 2 ‰. It rarely occurs in estuarine waters with more than 10-12 ppt salinity, about one-third salinity of sea water (USFWS, 1993, 1994, and 1995). The salinity used in by Waring and Moore (2004) simulated full strength seawater (33 ‰). Therefore the relevance of findings from this study to the DS is uncertain.

Tierney et al. (2007) studied the effect of 30 minute exposure to atrazine on behavioral and neurophysiological responses of juvenile rainbow trout to an amino acid odorant (L-histidine at 10^{-7} M). L-histidine was chosen because it has been shown to elicit an avoidance response in salmonids; however, control fish exposed to L-histidine at 10^{-7} M

showed a slight preference (1.2 response ratio). Although the study authors conclude that L-histidine preference behavior was altered by atrazine at exposures ≥ 1 ug/L, no significant decreases in preference behavior were observed at 1 ug/L. Furthermore, no dose response relationship was observed in the behavioral response following pesticide exposure. At 1 and 100 ug/L, non-significant decreases in L-histidine preference were observed; however a statistically significant avoidance of L-histidine was observed at 10 ug/L, but not 100 ug/L. Hyperactivity (measured as the number of times fish crossed the centerline of the tank) was observed in trout exposed to 1 and 10 ug/L atrazine. In the study measuring neurophysiological responses following atrazine exposure, electro-olfactogram (EOG) response was significantly reduced (EOG measures changes in nasal epithelial voltage due to response of olfactory sensory neurons). Although this study produced a more sensitive effects endpoint for freshwater fish, the data were not used quantitatively in the risk assessment because of the following reasons: 1) A negative control was not used; therefore, potential solvent effects cannot be evaluated; 2) The study did not determine whether the decreased response of olfactory epithelium to specific chemical stimuli would likely impair similar responses in intact fish; and 3) A quantitative relationship between the magnitude of reduced olfactory response to an amino acid odorant in the laboratory and reduction in trout imprinting and homing, alarm response, and reproduction (i.e., the ability of trout to detect, respond to, and mate with ovulating females) in the wild is not established.

Although these studies raise questions about the effects of atrazine on plasma steroid levels, behavior modifications, gill physiology, and endocrine-mediated functions in freshwater and anadromous fish, it is not possible to quantitatively link these sublethal effects to the selected assessment endpoints for the assessed species (i.e., survival, growth, and reproduction of individuals). Also, effects on survival, growth, or reproduction were not observed in the available life-cycle studies at concentrations that induced these reported sublethal effects. Therefore, these potential sublethal effects on fish are not used as part of the quantitative risk characterization. Further detail on sublethal effects data is provided in Appendix A.

4.4.2. Toxicity to Amphibians

4.4.2.1. Summary of SAP White Papers and Recommendations Regarding Sublethal Effects to Amphibians

As discussed in the October 2003 IRED, the Agency conducted an evaluation of the submitted studies regarding the potential effects of atrazine on amphibian gonadal development and presented its assessment in the form of a white paper for external peer review to a FIFRA Scientific Advisory Panel (SAP) in June 2003⁷. In the white paper dated May 29, 2003, the Agency summarized seventeen studies consisting of both open literature and registrant-submitted laboratory and field studies involving both native and non-native species of frogs (U.S. EPA, 2003d). Based on this assessment, the Agency

⁷ The Agency's May 2003 White Paper on Potential Developmental Effects of Atrazine on Amphibians is available at <http://www.epa.gov/oscpmont/sap/2003/june/finaljune2002telconfreport.pdf>.

concluded and the SAP concurred that there was sufficient evidence to formulate a hypothesis that atrazine exposure may impact gonadal development in amphibians, but there were insufficient data to confirm or refute the hypothesis (<http://www.epa.gov/oscpmont/sap/2003/June/junemeetingreport.pdf>). Because of the inconsistency and lack of reproducibility across studies and an absence of a dose-response relationship in the currently available data, the Agency determined that the data did not alter the conclusions reached in the January 2003 IRED regarding uncertainties related to atrazine's potential effects on amphibians. The SAP supported EPA in seeking additional data to reduce uncertainties regarding potential risk to amphibians. Subsequent data collection occurred following a multi-tiered process outlined in the Agency's white paper to the SAP (U.S. EPA, 2003d).

Because of the uncertainties associated with all the laboratory and field studies conducted prior to 2003, the 2003 SAP recommended that additional studies be conducted to determine if exposure to atrazine affects amphibian gonadal development. Since the 2003 SAP meeting, the Agency reviewed 36 open literature and registrant-submitted studies related to the potential effects of atrazine on gonadal development in amphibians. These studies were reviewed and were summarized in a second white paper to the SAP (U.S. EPA, 2007). The Agency concluded and the SAP concurred that the weight-of-evidence based on these studies does not show that atrazine produces consistent, reproducible effects across the range of exposure concentrations and amphibian species tested. In laboratory studies where environmental and animal husbandry factors were controlled, atrazine exposures (0.01 – 100 µg/L) did not affect time to or size at metamorphosis, sex ratio, or gonadal development. While there were several effects on secondary gross and histological endpoints that were statistically significant, their relationship to apical endpoints of intersex and/or gonadal development effects is not considered relevant.

In their report, the SAP recommended using *X. laevis* as the test species as well as indigenous species. Because the SAP report did not identify what benefits the indigenous species would provide, the Agency concluded that testing with *X. laevis* would be sufficient for a Tier 1 study. Based on the recent nineteen studies reviewed, including the recently submitted DCI studies showing no effects of atrazine on amphibian gonadal development, the Agency has further concluded that the higher tiers of testing proposed in the 2003 White Paper (USEPA 2003) are not needed at this time.

Nonetheless, the SAP also acknowledged the difficulty in testing a sufficient number of species to conclude that an entire taxa may be at risk or conversely not at risk; however, the Panel believed that *X. laevis* has aquatic animal characteristics that differentiate it from North American species. In support of their concern, the Panel provided several specific examples of stressor sensitivity differences in northern leopard frogs (*Rana pipiens*) versus *X. laevis*.

Because the SAP suggested that Ranids (such as the CRLF) could be more sensitive than *X. laevis* and that a sufficient number of amphibian species has not been tested on which to draw definitive conclusions, this assessment used the most sensitive toxicity values across fish and amphibian species to calculate risk quotients for the CRLF. Additional discussion on available studies in amphibians is included in Appendix A.

4.4.2.2. Additional Open Literature Data in Amphibians

Available acute data for amphibians, including the leopard frog (*Rana pipiens*), wood frog (*R. sylvaticas*), and American toad (*Bufo americanus*), indicate that they are relatively insensitive to atrazine with acute LC₅₀ values > 20,000 µg/L (Allran and Karasov, 2001).

The embryo and larval stages of several amphibian species were exposed to atrazine (Birge et al. 1980). Results varied among species tested. Early life stage LC50 values (mortality + teratogenic effects) for continuous exposure of embryos (eggs exposed for several hours after fertilization) and larvae (through 4 days post-hatch) were 410 µg/L for the bullfrog (*Rana catesbeiana*), 7,680 µg/L for the leopard frog (*Rana pipiens*), 17,960 µg/L for the pickerel frog (*Rana palustris*), and >48,000 µg/L for the American toad (*Bufo americanus*). In most of these species, concentrations of atrazine in excess of 5,000 µg/L were required to cause an incidence of teratic larvae in excess of 7 percent. These data do not suggest that the reproduction studies in fish that were used to calculate risk quotients underestimated toxicity and risk to the CRLF.

Chronic mortality data for aquatic-phase amphibians confirm that exposure to atrazine does not cause direct mortality to frogs and salamanders at concentrations ranging from approximately 200 to 2,000 µg/L; these concentrations represent the highest tested atrazine treatment levels within each of the studies. The available salamander data show no effect to mortality at the highest treatment concentrations of atrazine in each of the respective studies, ranging from approximately 200 to 400 µg/L (Boone and James, 2003; Rohr et al., 2003; Forson and Storfer, 2006). Rohr et al. (2004) reported decreased embryo survival through Day 16 in streamside salamanders following exposure to 400 µg/L atrazine (NOAEC = 40 µg/L). However, most embryo mortality was associated with a white film covering the embryo, suggesting the presence of a fungal pathogen, which may have decreased survival. According to the study authors, it is unknown whether the fungi caused or simply followed mortality. In addition, reduced survival was reported in only one of the two years tested; therefore, there is a high degree of uncertainty associated with the reported results.

A number of open literature studies were identified that evaluated potential effects of atrazine on amphibians in the most recent search of the Ecotox database (October, 2008). Relevant data are summarized in Appendix A. All accepted literature located in the October, 2008 Ecotox search is included in Appendix E, and a bibliography of accepted and rejected articles is included in Appendix F. No studies were located that reported more sensitive endpoints in fish that are quantitatively linked to the assessment endpoints of survival, growth, and reproduction.

4.4.3 Toxicity to Aquatic Invertebrates

Aquatic invertebrate toxicity studies were used to assess potential indirect effects to the DS and the CRLF. A summary of acute and chronic freshwater invertebrate data, including data published in the open literature, is provided below in the following Sections.

4.4.3.1. Acute Studies

Aquatic invertebrate toxicity data were used to evaluate potential indirect effects to the CRLF and the DS because each assessed species depends on aquatic invertebrates for sustenance. For the indirect effects assessment, the most sensitive aquatic invertebrate species was initially used for risk estimation, which is consistent with U.S. EPA (2004). The most sensitive organism tested was the marine copepod. The lowest LC50 in this species was 88 µg/L; however, a wide range of LC50 values have been reported in copepods from studies that tested technical grade atrazine (LC50 values of 88, 94, 140, 500, 4300, and 7900 µg/L have been reported, see Appendix A). Reasons for the disparity across the reported acute toxicity values in the copepod are unknown. However, similar variability has been observed in other species that have been tested by multiple laboratories. For example, studies conducted in the midge produced LC50s that spanned 2 orders of magnitude (values ranged from 720 to >33,000 µg/L). Other than the copepod, all reported acute toxicity values for the other 12 aquatic invertebrate species tested are 720 µg/L and higher.

The most important food organism for all sizes of the DS has been reported to be *Eurytemora affinis* (USFWS, 1995 and 2004 b), which is a marine copepod. Two studies were located that evaluated the toxicity of atrazine to *E. affinis* (Table 4.3).

Table 4.3. Data on the toxicity of atrazine to *Eurytemora affinis*.

Species	Results	Data Source	Comment
Copepod nauplii < 24 hours old (<i>Eurytemora affinis</i>)	LC50s (ug/L): Sal. 5 g/L: 500 Sal. 15 g/L: 2,600 Sal. 25 g/L : 13,300	MRIDs 45208303 & 45227711	Supplemental Study

Copepods (<i>Eurytemora affinis</i>) from the Seine river estuary (France).	An acute 96-hour LC50 was estimated for the copepod <i>E. affinis</i> nauplii of 125 ug/L for atrazine. A 10-day study was conducted using <i>E. affinis</i> (nauplius stage) that produced a NOAEC for survival of 25 ug/L and a LOAEC of 49 ug/L. Delayed maturity was also observed at 25 ug/L in the 30-day exposure study.	Forget-Leray et al., 2004 (Ecotox No. 80951)	Open literature study suitable for use qualitatively in risk characterization because reporting limitations and use of DMSO as a solvent preclude its use to calculate RQs. Reporting limitations included number and identification of test concentrations, % mortality at the LOAEC, and control responses.
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4.4.3.2. Chronic Exposure Studies

The most sensitive chronic endpoint for freshwater invertebrates was based on a 30-day flow-through study on the scud, which showed a 25% reduction in the development of F₁ to the seventh instar at atrazine concentrations of 140 µg/L; the corresponding NOAEC was 60 µg/L (MRID 00024377).

The most sensitive chronic bioassay in saltwater species was a 28-day study in mysid shrimp (*Americamysis bahia*) that reported a NOAEC of 80 µg/L; a 37% reduction in juvenile survival occurred at the LOAEC of 190 µg/L. Additional details on this study (MRID 45202920) and other chronic bioassays are described in Appendix A.

An uncertainty in the chronic bioassay data is that chronic toxicity data suitable for risk quotient derivation are not available on the most acutely sensitive marine invertebrate (copepod). An estimated acute to chronic ratio of 12.5 was derived for mysid shrimp based on an acute LC50 of 1000 ug/L (MRID 45202920) and a chronic NOAEC of 80 ug/L (MRID 45202920). Applying this ACR to the acute LC50 in copepods of 88 ug/L results in an estimated NOAEC of 7 ug/L.

Acute and chronic studies used to calculate risk quotients for aquatic invertebrates are summarized in Table 4.4.

Table 4.4. Acute and Chronic Aquatic Invertebrate Toxicity Values Used in Risk Estimation of Atrazine

Species Tested	Species Assessed / Effect	Study Type / Endpoints	Toxicity Value (µg/L)	Reference (MRID)	Comment
Saltwater invertebrate, acute Copepod (<i>Acartia tonsa</i>)	Delta Smelt / indirect effects from reduction in animal food supply	Acute toxicity / mortality	LC50: 88 Probit Slope: 0.95	Thursby <i>et al.</i> , 1990 (MRID 45202918)	Data used as initial screen to assess indirect effects to listed species from reduction of animal food supply.
Freshwater invertebrate, acute , Midge	CRLF / indirect effects from reduction in animal food supply	Acute toxicity / mortality	LC50: 720	00024377 Macek <i>et al.</i> 1976	Supplemental
Saltwater invertebrate, chronic , Mysid shrimp (<i>Americamysis bahia</i>)	Delta Smelt / indirect effects from reduction in animal food supply	Chronic exposure / growth and survival	NOAEC: 80	Ward & Ballantine, 1985 (MRID 45202920)	37% Reduction in survival occurred at the LOAEC of 190 µg/L. Data used as initial screen to assess indirect effects to listed species from reduction of animal food supply.
Saltwater invertebrate, acute Copepod (<i>Acartia tonsa</i>)	Delta Smelt / indirect effects from reduction in animal food supply	Chronic exposure / growth and survival	Estimated NOAEC: 7 µg/L	None	Estimated value based on an acute to chronic ratio of 12.5 from mysid shrimp acute and chronic studies.
Freshwater invertebrate, chronic , Scud	CRLF / indirect effects from reduction in animal food supply	Chronic exposure / 25 % red. in development of F ₁ to seventh instar.	NOAEC: 60	Macek <i>et al.</i> 1976 (MRID 00024377)	This study, as the most sensitive aquatic invertebrate chronic study, was used to characterize potential chronic toxicity of atrazine to the CRLF.

a Slope information on the toxicity study that was used to derive the RQ for freshwater invertebrates is not available. Therefore, the probability of an individual effect was calculated using a probit slope of 4.4, which is the only technical grade atrazine value reported in the available freshwater invertebrate acute studies; 95% confidence intervals could not be calculated based on the available data (Table A-18).

4.4.4 Toxicity to Aquatic Plants

Aquatic plant toxicity studies were used as one of the measures of effect to evaluate whether atrazine may affect primary production. Aquatic plants may also serve as dietary items of aquatic phase CRLFs. In addition, freshwater vascular and non-vascular plant data are used to evaluate a number of the PCEs associated with the critical habitat impact analysis.

Laboratory studies were used to evaluate the potential of atrazine to affect primary productivity. A summary of these data is provided the following sections.

4.4.4.1. Aquatic Plants: Laboratory Data

Numerous aquatic plant toxicity studies have been submitted to the Agency. A summary of the data for freshwater vascular and non-vascular plants is provided below. Appendix A includes a more comprehensive description of these data.

The Tier II results for freshwater aquatic plants produced EC₅₀ values for four different species of freshwater algae at concentrations as low as 1 µg/L, based on data from a 7-day acute study (MRID 00023544). Vascular plants are less sensitive to atrazine than freshwater non-vascular plants with an EC₅₀ value of 37 µg/L, based on reduction in duckweed growth (MRID 43074804).

Comparison of atrazine toxicity levels for three different endpoints in algae suggests that the endpoints in decreasing order of sensitivity are cell count, growth rate and oxygen production (Stratton, 1984). Walsh (1983) exposed *Skeletonema costatum* to atrazine and concluded that atrazine is only slightly algicidal at relatively high concentrations (i.e., 500 and 1,000 µg/L). Caux et al. (1996) compared the cell count IC₅₀ and fluorescence LC₅₀ and concluded that atrazine is algicidal at concentrations affecting cell counts. Abou-Waly et al. (1991) measured growth rates on days 3, 5, and 7 for two algal species. The pattern of atrazine effects on growth rates differs sharply between the two species. Atrazine had a strong early effect on *Anabaena flos-aquae* followed by rapid recovery in clean water (i.e., EC₅₀ values for days 3, 5, and 7 are 58, 469, and 766 µg/L, respectively). The EC₅₀ values for *Selenastrum capricornutum* continued to decline from day 3 through 7 (i.e., 283, 218, and 214 µg/L, respectively). Based on these results, it appears that the timing of peak effects for atrazine may differ depending on the test species.

Recovery from the effects of atrazine and the development of resistance to the effects of atrazine in some vascular and non-vascular aquatic plants have been reported and may add uncertainty to these findings. However, reports of recovery are often based on differing interpretations of recovery. Thus, before recovery can be considered as an uncertainty, an agreed upon interpretation is needed. For the purposes of this assessment, recovery is defined as a return to pre-exposure levels for the *affected population*, not for a replacement population of more tolerant species. Further research would be necessary in order to quantify the impact that recovery and resistance would have on aquatic plants.

4.4.4.2. Freshwater Field/Mesocosm Studies

Microcosm and mesocosm studies with atrazine provide measurements of primary productivity that incorporate the aggregate responses of multiple species in aquatic plant communities. Because plant species vary widely in their sensitivity to atrazine, the overall response of the plant community may be different from the responses of the individual species measured in laboratory toxicity tests. Mesocosm and microcosm studies allow observation of population and community recovery from atrazine effects and of indirect effects on higher trophic levels. In addition, mesocosm and microcosm studies, especially those conducted in outdoor systems, incorporate partitioning,

degradation, and dissipation, factors that are not usually accounted for in laboratory toxicity studies, but that may influence the magnitude of ecological effects.

Atrazine has been the subject of many mesocosm and microcosm studies in ponds, streams, lakes, and wetlands. The durations of these studies have ranged from a few weeks to several years at exposure concentrations ranging from 0.1 µg/L to 10,000 µg/L. Most of the studies have focused on atrazine effects on phytoplankton, periphyton, and macrophytes; however, some have also included measurements on animals. A summary of the available freshwater and saltwater aquatic microcosm, mesocosm, and field studies that were reviewed as part of the 2003 IRED is included in Appendix A. The 2003 IRED (U.S. EPA, 2003a) suggested that, potential adverse effects on sensitive aquatic plants and non-target aquatic organisms including their populations and communities are likely to be greatest when atrazine concentrations in water equal or exceed approximately 10 to 20 µg/L on a recurrent basis or over a prolonged period of time. However, the duration that constitutes an extended period of time was not defined.

4.5 Toxicity of Atrazine to Terrestrial Organisms

4.5.1 Toxicity to Birds

As specified in the Overview Document, the Agency uses birds as a surrogate for terrestrial-phase amphibians when sufficient toxicity data for each specific taxonomic group are not available (U.S. EPA, 2004). A summary of acute and chronic bird data is provided in Table 4.5 and is summarized in the following Sections. Additional studies and details on the studies summarized below are included in Appendix A.

Table 4.5. Summary of available acute and subacute toxicity studies in birds

Reference (MRID)	Species Tested	Study type/Endpoints	Toxicity value	Comment
Fink 1976 (MRID 00024721)	Northern bobwhite quail (<i>Colinus virginianus</i>)	Acute oral gavage toxicity / mortality	940 mg/kg-bw Slope = 3.8	Acceptable Study. The range of acute oral gavage LD50s in birds is 940 mg/kg-bw to 4200 mg/kg-bw.
Degradate Desethyl Atrazine (DEA) 96% Stafford, 2005c (MRID 46500009)	Northern bobwhite quail (<i>Colinus virginianus</i>)	Acute oral gavage toxicity / mortality	768 mg/kg-bw slope = 6.2	These data suggest that the degradate DEA is approximately as toxic to birds on an acute oral basis as atrazine.
Hill <i>et al.</i> 1975 (MRID 00022923)	Mallard duck (<i>Anas platyrhynchos</i>) 10-days old ducklings	Subacute dietary / mortality	> 5,000 (30 % mortality at 5,000 ppm)	All submitted subacute dietary studies in birds report LC50s that are higher than 5,000 ppm.

4.5.1.1 Birds: Acute Exposure (Mortality) Studies

The available data in birds suggest that atrazine is slightly toxic to avian species on an acute oral exposure basis. The lowest reported LD50 is 940 mg/kg-bw. Signs of intoxication in mallards first appeared 1 hour after treatment and persisted up to 11 days (U.S. EPA, 2003a). In pheasants, remission of signs of intoxication occurred by 5 days after treatment. Signs of intoxication included weakness, hyper-excitability, ataxia, and tremors; weight loss occurred in mallards.

One degradate (desethyl atrazine, DEA) has been shown to be roughly as toxic as atrazine to birds on an acute oral basis. Other degradates evaluated, including deisopropyl atrazine (DIA) and hydroxyatrazine (HA) are considerably less toxic than atrazine to birds on an acute oral basis (Appendix A). However, DACT, which has been shown to be of equivalent toxicity compared with atrazine in mammals, has not been tested in birds.

Because all subacute avian LC₅₀ values are greater than 5,000 ppm, atrazine is categorized as practically non-toxic to avian species on a subacute dietary basis. In the subacute dietary study in mallard ducks, 30% mortality was observed at the highest test concentration of 5,000 ppm (MRID 00022923). The time to death was Day 3 for the one Japanese quail and Day 5 for three mallard ducks (U.S. EPA, 2003).

4.5.1.2 Birds: Chronic Exposure (Growth, Reproduction) Studies

Reproduction studies in birds have reported reproductive effects at atrazine concentrations of 675 ppm and higher. In bobwhite quail, the following endpoints were affected at 675 ppm atrazine: egg production, embryo viability, hatchling and 14-day weight, and number of defective eggs (MRID 42547102). Bobwhite and mallard tests show similar toxic effects on reduced egg production and embryo viability/hatchability with LOAEC and NOAEC values of 675 and 225 ppm, respectively, for both species. These data are summarized in Table 4.6.

Table 4.6. Summary of Avian Reproduction Studies

Surrogate Species/ Study Duration	% ai	NOAEC/ LOAEC (ppm ai)	Statistically sign. ($p < 0.05$) LOAEC Endpoints	MRID No. Author/Year	Study Classification
Northern bobwhite (<i>Colinus virginianus</i>) 20 weeks	97.1	NOAEC 225 LOAEC 675	29 % red. in egg production 67 % incr. in defective eggs 27 % red. in embryo viability 6-13 % red. in hatchling body wt. 10-16 % red. in 14-day old body wt. 8.2 % red. in 14-day old body wt. (after recovery period)	425471-02 Pedersen & DuCharme 1992	Acceptable
Mallard duck (<i>Anas platyrhynchos</i>) 20 weeks	97.1	NOAEC 225 LOAEC 675	49 % red. in egg production 61 % red. in egg hatchability 12-17 % red. in food consumption	425471-01 Pedersen & DuCharme 1992	Acceptable

4.5.1.3. Sublethal Effects in Birds

No treatment-related sublethal effects were observed in the available studies described in Appendix A at levels lower than those used to calculate risk quotients.

4.5.1.4. Toxicity to Reptiles and Terrestrial Phase Amphibians

Limited data are available in terrestrial phase amphibians and in reptiles.

Atrazine was tested on eggs of the turtle, red-eared slider (*Pseudemys elegans*) and the American alligator (*Alligator mississippiensis*) to determine if atrazine produced endocrine effects on the sex of the young (Gross, 2001). The turtle and alligator eggs were placed in nests constructed of sphagnum moss treated with 0, 10, 50 100 and 500 µg/L for 10 days shortly after being laid. No adverse effects were found. Analysis of the embryonic fluids indicated that no atrazine was present in the eggs at the detection limit (0.5 µg/L). Under these conditions, atrazine does not appear to have permeated the leathery shell of reptiles (MRID 455453-03 and 455453-02).

Two additional open literature studies on snapping turtle and alligator egg exposures to atrazine suggest that exposure of reptilian eggs to atrazine does not cause significant alteration in gonadal development and aromatase activity at environmentally relevant concentrations (De Solla et al., 2005 and Crain et al., 1999). These studies are described further in Appendix A.

4.5.2 Toxicity to Mammals

A summary of acute and chronic mammalian data, including data published in the open literature, is provided below.

4.5.2.1 Mammals: Acute Exposure (Mortality) Studies

Atrazine is slightly toxic to mammals on an acute oral basis. LD50s are also available for two degradates (DEA and DIA). These degradates are also slightly toxic to rats on an acute oral basis; however, LD50s for both degradates were lower than the LD50 for atrazine. Acute oral toxicity data are summarized in Table 4.7.

Table 4.7. Summary of Acute Toxicity of Atrazine and its Degradates of Concern

Chemical	Acute Mammal LD50 (mg/kg-bw)
Atrazine	1900 (MRID 00024709)
HA	Not available
DEA	1240 (MRID 43013201)
DIA	1100 (MRID 43013202)
DACT	Not available

4.5.2.2 Reproduction Toxicity in Mammals

In a 2-generation reproduction study (MRID 40431303) technical grade atrazine was administered to Charles River (CRCD, VAF/PLUS) rats 30/sex/dose in the diet at concentrations of 0, 10, 50, and 500 ppm. Parental body weights, body weight gain, and food consumption were statistically significantly reduced at the 500 ppm dose in both sexes and both generations throughout the study. Compared to controls, body weights for F₀ high dose males and females at 70 days into the study were decreased by 12% and 15%, respectively while F₁ body weight for the same time period was decreased by 15% and 13% for males and females, respectively. The only other parental effect which may have been treatment related was a slight, but statistically significant, increase in relative testes weight which occurred in both generations of the high dose. There did not appear to be any reproductive effects from compound exposure. Measured reproductive parameters from both generations did not appear to be altered in a dose-related manner.

The LOAEL was 500 ppm (39 mg/kg/day in males, 43 mg/kg/day in females) based on decreased body weights, body weight gains, and food consumption. The NOAEL was 50 ppm (3.8 mg/kg/day in males, 3.7 mg/kg/day in females). The NOAEL of 50 ppm diet (3.7 mg/kg-bw) was used to calculate risk quotients.

Reproduction studies are not available on the degradates of concern. However, the degradates have been tested in prenatal developmental studies in rodents. These studies are summarized in Table 4.8. Results for atrazine from the same guideline study are also presented for comparison. The data suggest that the degradates are approximately as toxic as atrazine in these studies. All NOAELs of the degradates are within a factor of 5 of the atrazine NOAEL; the NOAEL for DEA and DIA are 2-fold lower, and the NOAEL for DACT is approximately 4-fold lower than the NOAEL for atrazine in the prenatal developmental study.

Table 4.8. Comparison of Toxicity Reference Values for Atrazine and Degradates of Concern in Guideline Prenatal Developmental Toxicity Studies

Chemical	Study type/Data Source	Study Summary
Atrazine	Prenatal developmental in rodents / 40566302	Maternal NOAEL = 10 mg/kg/day. Maternal LOAEL = 70 mg/kg/day, based on reduced body weight gain Developmental NOAEL = 10 mg/kg/day Developmental LOAEL = 70 mg/kg/day based on delayed or no ossification at several sites
	MRID 41065201	Maternal NOAEL = 25 mg/kg/day. Maternal LOAEL = 100 mg/kg/day based on reduced body weight gain and food consumption.

		Developmental NOAEL = 25 mg/kg/day. Developmental LOAEL = 100 mg/kg/day based on increased incidence of delayed ossification of skull bones.
HA	Prenatal developmental in rodents / MRID 41065202	Maternal NOAEL = 25 mg/kg/day Maternal LOAEL = 125 mg/kg/day based on decreased food consumption during the dosing period and enlarged and mottled kidneys. Developmental NOAEL = 25 mg/kg/day. Developmental LOAEL = 125 mg/kg/day based on increased incidence of partially ossified interparietal and hyoid bones and decreased fetal body weight.
DIA	Prenatal developmental in rodents / MRID 43013209	Maternal NOAEL = 5 mg/kg/day Maternal LOAEL = 25 mg/kg/day based on decreased food consumption, body weight/weight gain and food consumption Developmental NOAEL = 25 mg/kg/day Developmental LOAEL = 100 mg/kg/day based on increased fetal and litter incidences of fused sternebrae 1 and 2 and increased fetal incidence of poor ossification of the proximal phalanx of posterior digit 5 (a skeletal variation)
DEA	Prenatal developmental in rodents / MRID 43013208	Maternal NOAEL= 5 mg/kg/day Maternal LOAEL= 25 mg/kg/day based on decreased body weight gain and food consumption Developmental NOAEL= 5 mg/kg/day Developmental LOAEL 25 mg/kg/day based on . increased fetal and litter incidences of fused sternebrae 1 and 2
DACT	Prenatal developmental toxicity in rodents / MRID 41392402	Maternal NOAEL = 25 mg/kg/day Maternal LOAEL = 75 mg/kg/day, based on decreased body weight gain during dosing. Developmental NOAEL is 2.5 mg/kg/day. Developmental LOAEL = 25 mg/kg/day, based on increases in incidences of incompletely ossified parietals, interparietals and unossified hyoids.

4.5.2.3. Metabolism in Mammals

One potential exposure route for the CRLF is secondary exposure that may occur via consumption of contaminated mammals. Therefore, the potential for atrazine to remain in a small mammal for a duration that is sufficient to result in exposure to CRLFs that may consume small mammals was evaluated using available metabolism and

pharmacokinetic data. These data were reviewed by the Health Effects Division (HED), are summarized in Table 4.19, and indicate that atrazine is excreted relatively rapidly. However, small mammals may retain atrazine for a sufficient duration such that secondary exposure may occur. In addition, metabolism may not be important in determining whether secondary exposure may occur because it is assumed that atrazine may be retained in the gut for a sufficient amount of time to allow for secondary exposure to occur.

Table 4.9. Summary of Available Metabolism Data for Atrazine in Rodents

Guideline No./ Study Type	MRID No. (year) /Doses	Results
870.7485 Metabolism and pharmacokinetics	40431304 (1987) 0, 1, and 100 mg/kg for a single dose given through oral gavage. 1.0 mg/kg/day for 15 days by oral gavage.	<i>Distribution, accumulation</i> Distribution was dose-dependent and independent of sex. Distribution appeared to follow first-order kinetics and the half-life in the tissues was 31.3 hours. <i>Excretion</i> Approximately 95% of the atrazine excreted within 7 days of dosing. Urinary route accounted for about 75% of the excretion feces accounted for 20%. Route of excretion did not seem to vary among sexes or with dose.
870.7485 Metabolism and pharmacokinetics	MRID 40431305 (1987) The animals were dosed daily for 10 days through a stomach tube with dose levels of 0, 1, 3, 7, 10, 50 or 100 mg/kg/day.	<i>Distribution, accumulation</i> Distribution was highest in the red blood cell, followed by the liver, ovary and kidney. When the dose increased the amount distributed in the tissues increased. The distribution appeared to follow first-order kinetics and the tissue half-life was 38.6 hours. This indicates that atrazine, with possible exception of the red blood cell, does not bioaccumulate.
870.7485 Metabolism and pharmacokinetics	MRID 40431306 (1987) Rats were given test 100 mg/kg article was given through the stomach tube in a single oral dose. Other rats were given 16.18 and 19.64 mg/kg and urine was collected over a 24 hr period. The urine was analyzed for metabolites.	<i>Excretion</i> In the rats given 100 mg/kg greater than 100% of the administered radioactivity was recovered within 3 days of dosing. Urine was found to contain 47.3% of the radioactivity and the feces 49.3%. The tissues contained 5.75% and 1.4% was found in the blood. <i>Metabolism</i> Metabolites indicate that dechlorination of the triazine ring and N-dealkylation are the major metabolic pathways for atrazine in rats. Oxidation of the alkyl substituents of atrazine appears to be of minor metabolic importance.
870.7485 Metabolism and pharmacokinetics	MRID 42165503 (1993) Fecal and urinary samples from rats exposed in a	<i>Metabolic profile</i> No sex differences in metabolic profiles were evident.

Guideline No./ Study Type	MRID No. (year) /Doses	Results
	separate metabolism study (MRID 40431304) were obtained and analyzed to determine metabolism profiles.	The major fecal metabolite was DACT which accounted for 40% of the total fecal radioactivity.
870.7485 Metabolism and pharmacokinetics	MRID 44713802 (1993) single oral dose of 1 or 100 mg/kg through oral gavage	<p><i>Distribution, accumulation</i> Time to maximum blood concentration (t_{cmax}) was 2 hours and 24 hours for the low and high dose groups, respectively. With exception of red blood cells, whole blood, and skeletal muscle, tissue burden for any specific tissue or organ represented less than 1% of the administered dose by 14 days post dosing</p> <p><i>Excretion</i> Urinary excretion was 64.72% of the total administered low dose over a 48-hour period and 66.16% of the total administered high dose over a 168-hour period. Within 48 hours urinary excretion was 100% and 94% complete for the low-dose and high-dose group, respectively. Fecal elimination accounted for 10.80% and 19.69% of the total dose for the low and high dose groups, respectively.</p>

4.5.3 Toxicity to Terrestrial Invertebrates

Terrestrial invertebrate toxicity data are used to evaluate potential indirect effects to the CRLF and to adversely modify designated critical habitat. A summary of the available terrestrial insect data is summarized in Table 4.10 below. Additional details on the data are included in Appendix A.

Atrazine is practically non-toxic to honey bees (LD50: 97 ug/bee). It also did not cause adverse effects in fruit flies exposed to 15 ug/fly. LC50 values in earthworms ranged from 273 to 926 ppm soil (Mosleh et al., 2003; Haque and Ebing, 1983). Atrazine did not produce statistically significant ($p < 0.05$) adverse effects in studies on several beetle species at any level tested, which ranged from application rates of approximately 1 lb a.i./Acre to 8 lbs a.i./Acre (Kegel, 1989; Brust, 1990; Samsøe-Petersen, 1995).

The most sensitive terrestrial invertebrate species tested was the springtail (*Onychiurus apuanicus* and *O. armatus*). Exposure to *O. apuanicus* at 2.5 ppm resulted in 18% mortality, and exposure to *O. armatus* at 20 ppm resulted in 51% mortality (Mola et al., 1987); lower levels were not tested. These soil concentrations are associated with an application rate of approximately 1 lb a.i./Acre and 7 lbs a.i./Acre, respectively, assuming

a soil density of 1.3 grams/cm³ and a soil depth of 3 cm. Additional details on these studies may be found in Appendix A.

Available terrestrial insect toxicity data are summarized in Table 4.10.

Table 4.10. Summary of Available Terrestrial Invertebrate Toxicity Studies

Species	Toxicity Summary	Comment	Citation
Beetles	NOAECs ranged from 0.8 lbs a.i./Acre to 8 lbs a.i./Acre	Soil sprayed with atrazine at levels that ranged from 0.8 to 8 lbs a.i./Acre did not result in statistically significant (p<0.05) reductions in survival. LOAEC: Not achieved	Kegel, 1989 Ecotox No. 64007 Brust, 1990 Ecotox No. 70406 Samsøe-Petersen, 1995 Ecotox No. 63490
Earthworms	28-day LC50: 381 mg/kg soil 14-Day LC50: 273- 926 mg/kg soil	Spiked soil studies; endpoints included mortality and body mass	Mosleh et al., 2003 Ecotox No. 77549 Haque and Ebing, 1983 Ecotox No. 40493
Micro arthropods	NOAEC: 0.9 – 1.8 lbs a.i./Acre LOAEC: 5.4 lbs a.i./Acre	The LOAEC was based on reduced abundance from a field study (Fretello et al., 1985); it could not be determined if reduced abundance was caused by migration (repellency), by toxic effects, or both.	Cortet et al., 2002 Ecotox No. 75784 Fratello et. al., 1985 Ecotox No. 59428
Springtails	30-Day LD50: 17 ppm to 20 ppm (approximately 7 lbs a.i./Acre) ^a LOAEC: 2.5 - 20 ppm soil (approx. 1 – 7 lbs a.i./Acre) ^a	Exposure occurred via treated soil; mortality rate at 2.5 and 20 was 18% and 51%, respectively, compared with 0% in controls.	Mola et al., 1987. Ecotox No. 71417
Fruit flies <i>Drosophila</i>	NOAEC: 15 ug/fly	No increased mortality occurred in groups exposed to atrazine alone relative to controls.	Lichtenstein et al., 1973 Ecotox No. 2939
Honey bees	LD50: >97 ug/bee	5% mortality occurred at the highest dose tested (97 ug/bee)	MRID 00036935
Earthworm	LOAEC: 8 lb/acre	Field study examining the impacts of several herbicides on soil invertebrate populations. The endpoint measured was abundance of several species. Study authors suggested that reduced abundance was likely caused by repellency and not direct toxicity.	Fox, 1964 Ecotox No. 36668
Wire worm	NOAEC: Not achieved		
Springtail			

^a Application rate was estimated from soil concentration by assuming a soil density of 1.3 grams/cm³ and a soil depth of 3 cm.

4.5.4 Toxicity to Terrestrial Plants

Terrestrial plant toxicity data are used to evaluate the potential for atrazine to affect the riparian zone of occupied water bodies and critical habitat. Riparian zone effects could impact habitat and stream water quality as discussed in detail in Section 5.2.

Plant toxicity data from both registrant-submitted studies and studies in the scientific literature were reviewed for this assessment. Registrant-submitted studies are conducted under conditions and with species defined in EPA toxicity test guidelines. Sub-lethal endpoints such as plant growth, dry weight, and biomass are evaluated for both monocots and dicots, and evaluate effects at both seedling emergence and vegetative life stages. A guideline study generally evaluates toxicity to ten crop species. A drawback to these tests is that they are conducted on herbaceous agricultural crop species only, and extrapolation of effects to other species, such as woody shrubs and trees and wild herbaceous species, contributes uncertainty to risk conclusions. However, atrazine is labeled for use in forestry production; therefore effects to these types of trees are not anticipated at concentration anticipated in the environment. In addition, preliminary data (discussed below) suggests that sensitive woody plant species exist; however, damage to most woody species at labeled application rates is not expected.

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

Based on the results of the submitted terrestrial plant toxicity tests, it appears that emerged seedlings are more sensitive to atrazine via soil/root uptake exposure than emerged plants via foliar routes of exposure. However, all tested plants, with the exception of corn in the seedling emergence and vegetative vigor tests and ryegrass in the vegetative vigor test, exhibited adverse effects following exposure to atrazine.

For Tier II seedling emergence, the most sensitive dicot is the carrot and the most sensitive monocots are oats. EC₂₅ values, on an equivalent application rate basis, for oats and carrots, which are based on a reduction in dry weight, are 0.003 and 0.004 lb ai/A, respectively; NOAEC values for both species are 0.0025 lb ai/A. Table 4.11 summarizes the Tier II terrestrial plant seedling emergence toxicity data.

For Tier II vegetative vigor studies, the most sensitive dicot is cucumber and the most sensitive monocot is onion. In general, dicots appear to be more sensitive than monocots via foliar routes of exposure with all tested monocot species showing a significant reduction in dry weight at EC₂₅ values ranging from 0.008 to 0.72 lb ai/A. In contrast, two of the four tested monocots showed no effects from atrazine (corn and ryegrass), while EC₂₅ values for oats and onion were 0.61 and 2.4 lb ai/A, respectively. Table 4.12 summarizes the terrestrial plant vegetative vigor toxicity data used to derive risk quotients in this assessment.

Table 4.11. Nontarget Terrestrial Plant Seedling Emergence Toxicity (Tier II).

Surrogate Species	% ai	EC ₂₅ / NOAEC (lbs ai/A)	Endpoint Affected	MRID No. Author/Year	Study Classification
Monocot - Corn (<i>Zea mays</i>)	97.7	> 4.0 / > 4.0	No effect	420414-03 Chetram 1989	Acceptable
Monocot - Oat (<i>Avena sativa</i>)	97.7	0.004 / 0.0025	red. in dry weight	420414-03 Chetram 1989	Acceptable
Monocot - Onion (<i>Allium cepa</i>)	97.7	0.009 / 0.005	red. in dry weight	420414-03 Chetram 1989	Acceptable
Monocot - Ryegrass (<i>Lolium perenne</i>)	97.7	0.004 / 0.005	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Root Crop - Carrot (<i>Daucus carota</i>)	97.7	0.003 / 0.0025	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Soybean (<i>Glycine max</i>)	97.7	0.19 / 0.025	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Lettuce (<i>Lactuca sativa</i>)	97.7	0.005 / 0.005	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Cabbage (<i>Brassica oleracea alba</i>)	97.7	0.014 / 0.01	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Tomato (<i>Lycopersicon esculentum</i>)	97.7	0.034 / 0.01	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Cucumber (<i>Cucumis sativus</i>)	97.7	0.013 / 0.005	red. in dry weight	420414-03 Chetram 1989	Acceptable

Table 4.12. Nontarget Terrestrial Plant Vegetative Vigor Toxicity (Tier II).

Surrogate Species	% ai	EC ₂₅ / NOAEC (lbs ai/A)	Endpoint Affected	MRID No. Author/Year	Study Classification
Monocot - Corn (<i>Zea mays</i>)	97.7	> 4.0 / > 4.0	No effect	420414-03 Chetram 1989	Acceptable
Monocot - Oat (<i>Avena sativa</i>)	97.7	2.4 / 2.0	red. in dry weight	420414-03 Chetram 1989	Acceptable
Monocot - Onion (<i>Allium cepa</i>)	97.7	0.61 / 0.5	red. in dry weight	420414-03 Chetram 1989	Acceptable
Monocot - Ryegrass (<i>Lolium perenne</i>)	97.7	> 4.0 / > 4.0	No effect	420414-03 Chetram 1989	Acceptable
Dicot - Root Crop - Carrot (<i>Daucus carota</i>)	97.7	1.7 / 2.0	red. in plant height	420414-03 Chetram 1989	Acceptable
Dicot - Soybean (<i>Glycine max</i>)	97.7	0.026 / 0.02	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Lettuce (<i>Lactuca sativa</i>)	97.7	0.33 / 0.25	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Cabbage (<i>Brassica oleracea alba</i>)	97.7	0.014 / 0.005	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Tomato (<i>Lycopersicon esculentum</i>)	97.7	0.72 / 0.5	red. in plant height	420414-03 Chetram 1989	Acceptable
Dicot - Cucumber (<i>Cucumis sativus</i>)	97.7	0.008 / 0.005	red. in dry weight	420414-03 Chetram 1989	Acceptable

In addition, a report on the toxicity of atrazine to woody plants (Wall *et al.*, 2006; MRID 4687040001) was reviewed by the Agency. A total of 35 species were tested at application rates ranging from 1.5 to 4.0 lbs ai/A. Twenty-eight species exhibited either no or negligible phytotoxicity. Seven of 35 species exhibited >10% phytotoxicity. However, further examination of the data indicate that atrazine application was clearly associated with severe phytotoxicity in only one species (Shrubby Althea). These data suggest that, although sensitive woody plants exist, atrazine exposure to most woody plant species at application rates of 1.5 to 4.0 lbs ai/A is not expected to cause adverse effects. A summary of the available woody plant data is provided in Appendix A.

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

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

Individual effect probabilities are calculated based on an Excel spreadsheet tool IECV1.1 (Individual Effect Chance Model Version 1.1) developed by the U.S. EPA, OPP, Environmental Fate and Effects Division (June 22, 2004). The model allows for such calculations by entering the mean slope estimate (and the 95% confidence bounds of that estimate) as the slope parameter for the spreadsheet. In addition, the acute RQ is entered as the desired threshold.

The following probit slopes were used (probability of individual mortality calculations are presented in Section 5.2):

Saltwater Fish: Probit slope = 4.4 (95% C.I. of 2.8 – 5.9), sheepshead minnow – MRID 43344901

Freshwater Fish: Probit Slope = 2.7 (95% CI not determined), rainbow trout – MRID 00024716

Freshwater Aquatic Invertebrate: Probit slope = 4.4, scud – MRID 45202917

Slope information on the most sensitive aquatic invertebrate (midge) is not available. Therefore, the probability of an individual effect was calculated using the probit slope of 4.4, which is the only technical grade atrazine value reported across invertebrate studies; 95% confidence intervals

could not be calculated based on the available data (MRID 45202917); therefore, an upper and lower bound of 2 to 8 was used.

Saltwater Aquatic Invertebrate: Probit slope = 0.94 (95% C.I.: 0.64 to 1.3), copepod – MRID 45202918

Mammals: Probit slope = 3.3 (95% C.I.: 1.8 – 4.8), Laboratory rats (MRID 006202307)

Birds: Probit slope = 3.8 (95% C.I.: not statistically determined, lower and upper bound of 2 – 8 was used; MRID 00024721).

4.7 Incident Database Review

A review of the EIIS database for ecological incidents involving atrazine was completed on November 15, 2008. The results of this review for terrestrial, plant, and aquatic incidents are discussed below. A more complete list of the incidents including associated uncertainties is included as **Appendix H**.

A number of incidents have been reported in which atrazine has been associated with some type of environmental effect. Incidents are maintained and catalogued by EFED in the Ecological Incident Information System (EIIS). Each incident is assigned a level of certainty from 0 (unrelated) to 4 (highly probable) that atrazine was a causal factor in the incident. As of the writing of this assessment, 489 incidents are in EIIS for atrazine spanning the years 1970 to 2005. Of these incidents, 448 were assigned a certainty index of 2 or higher (possible, N=323; probable, N=121; or highly probable, N=4). The remaining 41 incidents were assigned a certainty index of unlikely or unrelated. Most (440/489, 90%) of the incidents involved damage to terrestrial plants, and most of the terrestrial plant incidences involved damage to crops treated directly with atrazine or that were damaged from atrazine application to crops that were planted on the agricultural field in a previous crop rotation. Of the remaining 49 incidents, 47 involved aquatic animals and 2 involved birds. These incidents are summarized in Appendix H. There were 23 incidents associated with aquatic or terrestrial animal kills assigned a certainty index of 2 or higher. These incidents were further evaluated and were grouped into three categories:

1. Incidents in which atrazine concentrations were confirmed to be sufficient to either cause or contribute to the incident, including directly via toxic effects to aquatic organisms or indirectly via effects to aquatic plants, resulting in depleted oxygen levels;
2. Incidents in which insufficient information is available to conclude whether atrazine may have been a contributing factor – these may include incidents where there was a correlation between atrazine use and a fish kill, but the presence of atrazine in the affected water body was not confirmed; and
3. Incidents in which causes other than atrazine exposure are more plausible (e.g., presence of substance other than atrazine confirmed at toxic levels).

The presence of atrazine at levels thought to be sufficient to cause either direct or indirect effects was confirmed in 3 aquatic incidents evaluated. Atrazine use was also correlated with 14 incidents where its presence in the affected water was not confirmed, but the timing of atrazine application was correlated with the incident. Therefore, a definitive causal relationship between atrazine use and the incident could not be established; however, atrazine may or may not have contributed to or caused the associated incident. The remaining incidents were likely caused by some factor other than atrazine. Other causes primarily included the presence of other pesticides at levels known to be toxic to affected animals. Further information on the atrazine incidents and a summary of uncertainties associated with all reported incidents are provided in Appendix H.

5. Risk Characterization

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

5.1 Risk Estimation

Risk was estimated by calculating the ratio of the PRZM/EXAMS estimated environmental concentration (EEC) (Section 3) and the appropriate toxicity endpoint (Section 4). This ratio is the risk quotient (RQ), which is then compared to pre-established acute and chronic levels of concern (LOCs) for each category evaluated (Appendix D). Screening-level RQs are based on the most sensitive effects endpoints and the PRZM/EXAMS EECs that were summarized in Table 3.4.

In cases where the baseline RQ exceeds one or more LOC (*i.e.*, “may affect”), additional factors, including the life history characteristics of the assessed species, refinement of the baseline EECs using site-specific information, and available monitoring data are considered and used to characterize the potential for atrazine to adversely affect the assessed species and their designated critical habitat. Risk quotients used to evaluate potential direct and indirect effects to the CRLF and DS and to designated critical habitat are in Sections 5.1.1 (direct effects) and 5.1.2 (indirect effects). RQs are described and interpreted in the context of an effects determination in Section 5.2 (risk description).

5.1.1 Direct Effects RQs

The species considered in this risk assessment include a frog and a fish species. Direct effects to the DS were evaluated using the lowest acute and chronic toxicity values across freshwater and saltwater fish species. Direct effects to the aquatic phase CRLF were evaluated using the lowest freshwater acute and chronic toxicity values across fish and amphibian toxicity studies. However, the available data in fish provide more sensitive toxicity endpoints than the available data in aquatic phase amphibians. Therefore, fish acute and chronic toxicity values were used to calculate RQs for aquatic phase amphibians. Direct effects to terrestrial phase CRLFs were evaluated using the lowest acute and chronic toxicity values across bird and terrestrial phase amphibian species that have been tested in adequate studies. Toxicity values used to calculate RQs are discussed in Section 4, and exposure values are discussed in Section 3. RQs used to estimate acute and chronic direct effects are in Tables 5.1 and 5.2.

Table 5.1 Summary of Aquatic RQs Used to Estimate Direct Effects to aquatic phase CRLF and the DS

Use Site	Exposure Type	Surrogate Species	Toxicity Value (ug/L)	EEC	RQ ^a	Probability of Individual Effect ^b
Guava (highest EEC across all uses)	Acute	Sheepshead Minnow	LC ₅₀ : 2000	Peak = 64 ug/L	0.03	1 in 9E+10 (1 in 9E+4 to 1 in 8E+18) ^a
		Rainbow trout	LC ₅₀ : 5300		0.01	1 in 3E7 (no confidence limit available from study)
	Chronic	Brook Trout	NOAEC: 65 ug/L	60-day = 60 ug/L	0.92	--
^a RQ < acute endangered species LOC of 0.05 and the chronic LOC of 1.0 Lowest LC ₅₀ across fish species was 2000 ug/L from sheepshead minnow study (MRID 43344901); lowest chronic NOAEC was from a brook trout study of 65 ug/L (MRID 00024377). ^b Based on a probit slope value of 4.4 for the sheepshead minnow from a different study with 95% confidence intervals of 2.8 to 5.9 (MRID 433449-01)						

Table 5.2 Summary of RQs Used to Estimate Direct Effects to Terrestrial-Phase CRLFs Based on an LD50 of 940 mg/kg-bw and a NOAEC of 225 mg/kg-diet in Bobwhite Quail

Use Site	Exposure Type	EEC	RQ	Probability of Individual Effect ^a	Distance from Treated Field LOC is Exceeded ^b
Macadamia Nuts	Acute	800 mg/kg-bw	1.2	>1 in 2	88 feet
	Chronic	700 mg/kg-diet	3.1	--	6 feet
Guava, Forestry	Acute	620 mg/kg-bw	0.9	1 in 2 (1 in 2 to 1 in 3)	88 feet
	Chronic	540 mg/kg-diet	2.4	--	6 feet
Sorghum, Corn, Residential	Acute	310 mg/kg-bw	0.45	1 in 11 (1 in 40 to 1 in 360)	26 feet
	Chronic	270 mg/kg-diet	1.2	--	Adjacent
Residential, Turf (Spray)	Acute	200 mg/kg-bw	0.29	1 in 49 (1 in 7 to 1 in 1E5)	Adjacent
	Chronic	170 mg/kg-diet	0.78	--	--
Residential, Turf, Granular	Acute	21 mg/sq. ft.	1.5 (LD50/sq. ft)	--	--

^a Based on a probit slope of 3.8 from MRID 00024721. 95% confidence interval could not be calculated; therefore, a lower and upper bound slope of 2 to 8 was used (U.S. EPA, 2004).

^b AgDrift was run using the default settings in Tier 1 and assuming a single application. Aerial application was assumed except for macadamia nuts and turf (ground spray).

Based on the highest modeled EEC for atrazine use patterns within the action area, acute or chronic direct effects RQs do not exceed the endangered species LOC of 0.05 for aquatic phase CRLFs or for the DS. However, avian RQs exceeded the endangered species LOC of 0.1 for acute effects and 1.0 for chronic effects. Birds are used as surrogate species for terrestrial phase CRLFs. These RQs are further characterized in the context of the effects determination in Section 5.2.

5.1.2 Indirect Effects

This section presents RQs used to evaluate the potential for atrazine to induce indirect effects. Pesticides have the potential to exert indirect effects upon listed species by inducing changes in structural or functional characteristics of affected communities. Perturbation of forage or prey availability and alteration of the extent and nature of habitat are examples of indirect effects. A number of these indirect effects are also considered as part of the critical habitat modification evaluation. In conducting a screen for indirect effects, direct effects LOCs for each taxonomic group (i.e., freshwater fish, invertebrates, aquatic plants, and terrestrial plants) are employed to make inferences concerning the potential for indirect effects upon listed species that rely upon non-listed organisms in these taxonomic groups as resources critical to its life cycle (U.S. EPA, 2004). This approach used to evaluate indirect effects to listed species is endorsed by the Services (USFWS/NMFS, 2004b). If no direct effect listed species LOCs are exceeded for organisms on which the assessed species depends for survival or reproduction, indirect effects are not expected to occur.

If LOCs are exceeded for organisms on which the assessed species depend for survival or reproduction, dose-response analysis is used to estimate the potential magnitude of effect associated with an exposure equivalent to the EEC. The greater the probability that exposures will produce effects on a taxa, the greater the concern for potential indirect effects for listed species dependant upon that taxa (U.S. EPA, 2004).

As an herbicide, indirect effects to the assessed species from potential effects on primary productivity of aquatic plants are a principle concern. If plant RQs fall between the endangered species and non-endangered species LOCs, a no effect determination for listed species that rely on multiple plant species to successfully complete their life cycle (termed plant dependent species) is determined. If plant RQs are above non-endangered species LOCs, this could be indicative of a potential for adverse effects to those listed species that rely either on a specific plant species (plant species obligate) or multiple plant species (plant dependant) for some important aspect of their life cycle (U.S. EPA, 2004). Based on the information provided in Section 2.3, the assessed species do not have any known obligate relationship with a specific species of aquatic plant.

Direct effects to riparian zone vegetation could also indirectly affect the assessed species by reducing water quality and available spawning habitat via increased sedimentation. Direct impacts to the terrestrial plant community (i.e., riparian habitat) are evaluated using submitted terrestrial plant toxicity data. If terrestrial plant RQs exceed the Agency's LOC for direct effects to non-endangered plant species, based on EECs derived using EFED's Terrplant model (Version 1.2.1) and submitted guideline terrestrial plant toxicity data, a conclusion that atrazine may affect the CRLF and DS via potential indirect effects to the riparian habitat (and resulting impacts to habitat due to increased sedimentation) is made. Further analysis of the potential for atrazine to affect the CRLF and the DS via reduction in riparian habitat includes a description of the importance of riparian vegetation to the assessed species and types of riparian vegetation that may potentially be impacted by atrazine use within the action area.

RQs used to evaluate the potential for atrazine to induce indirect effects to the assessed species are presented in Sections 5.1.2.1 to 5.1.2.4. These RQs suggest that potential indirect effects could occur by potentially impacting food availability and primary productivity as indicated by LOC exceedances. These RQs were based on the most sensitive surrogate species tested across aquatic invertebrate, fish, and aquatic plant species tested. Discussion of these RQs in the context of this effects determination is presented in Section 5.2.

5.1.2.1 Aquatic Invertebrates

Aquatic invertebrate RQs are summarized in Table 5.3 and are used to evaluate the potential for atrazine to affect the CRLF and the DS by potentially impacting the food supply. Both the CRLF and the DS consume aquatic invertebrates as part of their diet. Acute risk quotients for invertebrates were based on peak EECs in the standard pond and the lowest acute toxicity value for freshwater and saltwater invertebrates. Chronic risk

was based on 21-day EECs and the lowest chronic toxicity value for freshwater and saltwater invertebrates.

Table 5.3. Summary of Acute and Chronic RQs for Aquatic Invertebrates Used to Evaluate Potential Indirect Effects to the CRLF and the DS Resulting from Potential Impacts to Food Supply

Taxonomic Group	Acute RQ Range Across All Uses				Chronic RQ Range Across All Uses		
	Peak EEC (µg/L)	Acute LC50	Range of Acute RQ ^a	Probability of Individual Effect	21-day EEC (µg/L)	Chronic NOAEC (µg/L)	Chronic RQ ^a
Saltwater Invertebrate (DS food item)	17 to 64 (range across all uses)	88µg/L (Copepod) MRID 45202918	0.19 to 0.73	1 in 4 to 1 in 2	16 to 62	7 (estimated) ^b	2 to 9
Freshwater Invertebrate (CRLF food item)	17 to 64 (range across all uses)	720 µg/L (Midge) MRID 00024377	0.02 to 0.09	1 in 3E13 to 1 in 5E5	16 to 62	60 (scud) MRID 00024377	0.27 to 1.0

^a RQs for some uses exceed the endangered species LOC of 0.05, the restricted use LOC of 0.2, the acute LOC of 0.5, and the chronic LOC of 1.0. Discussion of these RQs in the context of this effects determination is presented in Section 5.2.

^b Chronic NOAEC was estimated using the following equation: Estimated NOAEC = LC50 / ACR; Estimated NOAEC = 88 / 12.5 = 7.

The endangered species LOC (0.05) was exceeded for freshwater invertebrates for all uses modeled except sorghum, turf, and residential uses. The endangered species LOC was exceeded for saltwater invertebrates for all uses modeled. The acute LOC (0.5) was exceeded for turf, residential, and sorghum for saltwater invertebrates, and the chronic LOC of 1.0 for saltwater invertebrates was exceeded for all uses. These RQs were based on the most sensitive surrogate species across aquatic invertebrate species tested. Discussion of these RQs in the context of this effects determination is presented in Section 5.2.

5.1.2.2 Terrestrial Invertebrates

Terrestrial invertebrate RQs are used to evaluate the potential for atrazine to affect the CRLF by potentially impacting the food supply. Terrestrial invertebrate RQs are presented in Table 5.4.

Table 5.4. Summary of Acute and Chronic RQs for Terrestrial Invertebrates Used to Evaluate Potential Indirect Effects to the CRLF Resulting from Potential Impacts to Food Supply

EEC	Acute LC50	Range of Acute RQ	Probability of Individual Effect	RQ Interpretation
1 lb a.i./Acre to 4 lbs a.i./Acre	Approx. 7 lbs a.i./Acre ^a	0.15 - 0.6	1 in 10,000 to 1 in 6 based on a probit slope of 4.5 ^b	Interim LOC for terrestrial invertebrates of 0.05 was exceeded for all uses. The LOC was estimated to be exceeded for up to approx. 120 feet from the application site based on AgDrift analysis. Further evaluation of the RQs as they relate to an effects determination is warranted.
^a This LC50 is an empirical value that was not statistically derived; 51% mortality occurred at 20 ppm soil (Mola et al., 1987; Ecotox No. 71417). Assuming a soil depth of 3 cm and a soil density of 1.3 g/cm ³ , an application rate of 7 lbs a.i./Acre would be associated with a soil concentration of 20 ppm. This calculation assumes no foliar interception (e.g., direct spraying of bare ground). ^b Probability of an individual effect was based on a default slope of 4.5 because the available studies did not allow for an estimation of a probit slope. Reasonable lower and upper bounds of the probit slope are 2 and 8 respectively (U.S. EPA, 2004).				

The endangered species LOC was exceeded for terrestrial invertebrates. AgDrift analysis using default Tier 1 settings indicated that LOCs were estimated to be exceeded for up to approximately 120 feet from the application site. Discussion of these RQs in the context of this effects determination is presented in Section 5.2.

5.1.2.3 Mammals

Potential risks to mammals are derived using T-REX, acute and chronic rat toxicity data. RQs are typically derived for various sizes of mammals (15-, 35-, and 1000-gram); however, RQs are not presented for 1000-gram mammals because it is improbable that even the largest CRLF would consume a mammal of that size. Therefore, the evaluation for potential indirect effects to the CRLF resulting from potential reductions in mammal abundance as food is based on the 15-gram size class, which results in higher RQs than the 35-gram mammal. The California mouse is a particular species known to be consumed by the CRLF. The California mouse is omnivorous and consumes grasses, fruits, flowers, and invertebrates (USC, 2005; http://wotan.cse.sc.edu/perobase/systematics/p_calif.htm). Therefore, the short grass food item was used to determine if mammals could be impacted; however, RQs based on EECs on other food items were also derived. A range of RQs for mammals is presented in Tables 5.5 (use pattern with lowest EECs) and 5.6 (use pattern with highest EECs).

Table 5.5. Acute and Chronic Mammal RQs; 1 lb a.i./Acre, 2 applications, 30-day application interval^a

DIETARY	Acute dose-	Probability of	Chronic dose-	Chronic	Distance (feet) From
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CATEGORY	based RQ	Individual Effect ^b	based RQ	dietary based RQ	Treated Site Resulting in No LOC Exceedance ^c
Short Grass	0.07	1 in 1E5 (1 in 96 to 1 in 7E24)	36	6.2	260
Tall Grass	0.03	1 in 6E9 (1 in 390 to 1 in 2E42)	17	2.9	120
Broadleaf Plants/ Small Insects	0.04	1 in 9E18 (1 in 4E2 to 1 in 7E35)	20	3.5	150
Fruits/Pods/Seeds/ Large Insects	<0.01	1 in 9E18 (<1 in 3E4 to <1 in 1E72)	2.3	0.4	Adjacent

^a Dose-based evaluation based on a 15-gram mammal; LOC exceedances are bolded.
^b Probability of individual effect was calculated based on the default slope of 4.5 with lower and upper reasonable bounds of 2 to 9 because the data did not fit probit assumptions.
^c Distance calculated using AgDRIFT assuming single aerial application and default Tier 1 parameters

Table 5.6. Acute and Chronic Mammal RQs; 4 lb a.i./Acre, 2 applications, 30-day interval^a

Dietary Category	Acute Dose-Based RQ	Probability of Individual Effect ^b	Chronic Dose-Based RQ	Chronic dietary-Based RQ	Distance From Treated Site Resulting In No LOC Exceedance ^c
Short Grass	0.29	1 in 130 (1 in 7 to 1 in 2E6)	146	25	1700 ft
Tall Grass	0.13	1 in 3E4 (1 in 26 to <1 in 1E6)	67	11	500
Broadleaf Plants/Small Insects	0.16	1 in 6E3 (1 in 18 to <1 in 1E6)	82	14	620
Fruits/Pods/Seeds/ Large Insects	0.02	1 in 9E13 (1 in 3E3 to <1 in 1E6)	9.1	1.6	69

= LOC exceedances (RQ ≥0.1) are bolded and shaded.
^a Dose-based evaluation based on a 15-gram mammal
^b Probability of individual effect was calculated based on the default slope of 4.5 with lower and upper reasonable bounds of 2 to 9 because the data did not fit probit assumptions. Probabilities less than 1 in 1,000,000 are denoted as <1E6.
^c Distance calculated using AgDRIFT assuming a single application, aerial application and default Tier 1 parameters except that Tier III was used for short grass because the distance extended beyond the limit for Tier 1 (1000 feet).

For granular formulations, an LD50/sq. ft analysis was performed using T-REX version 1.4.1. Granular formulations are only labeled for use on residential and turf at 2 lbs a.i./Acre. The LD50/sq. ft for a small (15-gram) mammal was 0.34. RQs for granular applications are similar to those presented for the macadamia nut use (short grass) and exceed the endangered species LOC of 0.1, but not the acute risk LOC of 0.5. The probability of an individual effect at the mammalian RQ of 0.34 is 1 in 60 (reasonable lower and upper bounds of 1 in 6 to 1 in 81,000).

Based on acute and chronic LOC exceedances, there is potential for atrazine to impact mammal abundance, which could result in indirect effects to the CRLF. Discussion of these RQs in the context of this effects determination is presented in Section 5.2.

5.1.2.4 Aquatic Plants

Aquatic plants serve as food supply for both the CRLF and the DS. Therefore, RQs for vascular and non-vascular plants are used to evaluate the potential for atrazine to affect the CRLF and the DS by potentially impacting the food supply, water quality, and habitat. These RQs are presented in Table 5.7. LOCs were exceeded for all uses.

Table 5.7. RQs used to evaluate the potential for atrazine to induce indirect effects to the CRLF and the DS by affecting aquatic plants.

Endpoint	Taxonomic Group	Toxicity Value	Peak EEC	RQ	LOC Exceedances
Reduction in food supply; Primary productivity	Vascular Aquatic Plants	EC50: 37 ug/L	Residential, Sorghum, Turf: 17 – 34 ug/L	0.5 – 0.9	None
			Forestry, corn, Macadamia nuts, and guava: 50 – 64 ug/L	1.4 – 1.7	LOCs were exceeded for forestry, corn, macadamia nuts, and guava uses
	Non-Vascular Aquatic Plants	EC50: 1 ug/L	17 – 64 ug/L	17 – 64	LOCs were exceeded for all uses.

Potential indirect effects resulting from potential impacts on terrestrial plants were assessed using RQs from terrestrial plant seedling emergence and vegetative vigor EC₂₅ data as a screen. Based on the results of the submitted terrestrial plant toxicity tests, it appears that emerging seedlings are more sensitive to atrazine via soil/root uptake than emerged plants via foliar routes of exposure. However, all tested plants, with the exception of corn in the seedling emergence and vegetative vigor tests, and ryegrass in the vegetative vigor test, exhibited adverse effects following exposure to atrazine. The results of these tests indicate that a variety of terrestrial plants that may inhabit riparian zones may be sensitive to atrazine exposure. RQs used to estimate potential indirect effects to the CRLF and the DS from potential effects to seedling emergence and vegetative vigor of terrestrial plants within riparian areas are summarized in Tables 5.8 and 5.9, respectively.

Terrestrial plant RQs were highest for semi-aquatic areas. RQs exceeded LOCs for all uses except corn. Carrots were the most sensitive plant with RQs of 69 (ground spray, turf and residential) to 350 (aerial application, guava).

For dry adjacent areas, terrestrial plant RQs are above the Agency's LOC for all species except corn. For species with LOC exceedances, RQ values based on aerial application of atrazine at 4.0 lb ai/A range from 1.8 to 113; the maximum RQ value based on an equivalent ground application is 35, approximately a three-fold reduction as compared to

aerial applications. Granular application of atrazine to residential lawns at 2.0 lb ai/A may also impact terrestrial plants exposed to atrazine via runoff with RQs ranging from <1 (corn and soybeans) to 13 (carrots). Monocots and dicots show similar sensitivity to atrazine; therefore, RQs are similar across both taxa.

Table 5.8 Non-target Terrestrial Plant Seedling Emergence RQs

Surrogate Species	EC ₂₅ (lbs ai/A) ^a	EEC Dry adjacent areas ^b	RQ Dry adjacent areas ^b	EEC Semi-aquatic areas	RQ Semi-aquatic areas
Monocot - Corn	> 4.0	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	<LOC	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	<LOC
Monocot - Oat	0.004	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: 43 - 85 Ground: 6.5 - 26 Granular: 10	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	132 – 265 52 – 206 100
Monocot - Onion	0.009	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: 19 - 38 Ground: 2.9 - 12 Granular: 4.4	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	60 – 120 23 – 92 44
Monocot - Ryegrass	0.004	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: 43 - 85 Ground: 6.5 - 26 Granular: 10	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	132 – 265 52 – 206 100
Dicot - Carrot	0.003	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: 57 - 113 Ground: 8.7 - 35 Granular: 13	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	180 – 350 69 – 274 130
Dicot - Soybean	0.19	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: < LOC – 1.8 Ground: < LOC Granular: < LOC	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	2.8 - 5.6 1.1 – 4.3 2.1
Dicot - Lettuce	0.005	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: 34 - 68 Ground: 5.2 - 21 Granular: 8	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	106 – 212 41 – 165 80
Dicot - Cabbage	0.014	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: 12 - 24 Ground: 1.9 – 7.4 Granular: 2.9	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	38 – 76 15 – 59 29
Dicot - Tomato	0.034	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: 5.0 - 10 Ground: 0.75 – 3.1 Granular: 1.2	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	16 - 31 6.1 – 24 12
Dicot - Cucumber	0.008	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	Aerial: 21 - 43 Ground: 3.2 - 13 Granular: 5	Aerial: 0.53 – 1.1 Ground: 0.21 – 0.82 Granular: 0.4	66 – 132 26 – 103 50
^a From Chetram (1989); MRID 420414-03; the lowest value of the vegetative vigor and seedling emergence EC ₂₅ was used to calculate RQs.					
^b Range of EECs and RQs based on use scenarios presented in Section 3.					

Drift only EECs are included in Table 5.9. LOCs were exceeded for aerial applications and ground applications for all uses.

Table 5.9 Non-target Terrestrial Plant Drift Only RQs

Surrogate Species	EC₂₅ (lbs ai/A)^a	Drift EEC (lbs ai/A)^b	Drift RQ^b
Monocot - Corn	> 4.0	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	<LOC
Monocot - Oat	0.004	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	32 – 65 1.5 - 6
Monocot - Onion	0.009	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	14 – 29 0.7 – 2.7
Monocot - Ryegrass	0.004	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	32 – 65 1.5 – 6
Dicot - Carrot	0.003	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	43 – 87 2 - 8
Dicot - Soybean	0.19	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	0.68 – 1.4 <LOC
Dicot - Lettuce	0.005	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	26 – 52 1.2 – 4.8
Dicot - Cabbage	0.014	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	9.3 – 18.6 0.4 – 1.7
Dicot - Tomato	0.034	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	3.8 - 7.7 <LOC
Dicot - Cucumber	0.008	Aerial: 0.13 – 0.26 Ground: 0.006 – 0.02	16 – 33 1.8 - 3
^a From Chetram (1989); MRID 420414-03.			
^b Range of EECs and RQs based on use scenarios presented in Section 3			

LOCs were exceeded for both aquatic and terrestrial plants, which could result in indirect effects to the CRLF or the DS. These LOC exceedances and their impact on the effects determination are described in Section 5.2.

5.1.3 Primary Constituent Elements of Designated Critical Habitat

For atrazine use, the assessment endpoints for designated critical habitat PCEs involve the same endpoints as those being assessed relative to the potential for direct and indirect effects to the listed species assessed here. Therefore, the effects determinations for direct and indirect effects presented in Section 5.1 are used as the basis of the effects determination for potential modification to designated critical habitat.

5.1.4 Spatial Extent of Potential Effects

Since this screening level risk assessment defines taxa that are predicted to be exposed through runoff and drift to atrazine at concentrations above the Agency's Levels of Concern (LOC), analysis of the potential spatial extent of potential effects requires expansion of the area from the treated site to include all areas where risk to the CRLF and the DS exceed LOCs.

An LAA effects determination applies to those areas where it is expected that the pesticide's use will directly or indirectly affect the CRLF or its designated critical habitat. To determine this area, the footprint of atrazine's use pattern is identified, using corresponding land cover data. The spatial extent of the effects determination also includes areas beyond the initial area of concern that may be impacted by runoff and/or spray drift. The identified direct and indirect effects and modification of critical habitat are anticipated to occur only for those currently occupied core habitat areas, CNDDDB occurrence sections and designated critical habitat for the CRLF that overlap with the area where effects may occur (potential use areas + distance down stream or down wind from use sites where habitat may be affected). The determination of the buffer distance and downstream dilution for spatial extent of the effects determination is described below.

5.1.4.1 Spray Drift

In order to determine terrestrial and aquatic habitats of concern due to atrazine exposures through spray drift, it is necessary to estimate the distance that spray applications can drift from the treated area and still be present at concentrations that exceed levels of concern. A quantitative analysis of spray drift distances was completed using AgDrift (v. 2.01) using inputs included in Table 5.10.

Table 5.10. Input parameters for simulation of atrazine in spray drift using AgDrift

Parameter Description	Macadamia Nuts	Forestry, Guava	Corn and Sorghum	Residential Turf (spray only)
Application Method	Ground	Aerial	Aerial	Ground
Single Application Rate	4 lb a.i./A	4 lb a.i./A	2 lb a.i./A	1 lb a.i./A
Droplet Size Distribution (DSD)	ASAE very fine to fine	ASAE fine to medium	ASAE fine to medium	ASAE very fine to fine
Release height	High Boom	NA	NA	High Boom

The spray drift analysis is based on potential for direct and indirect effects. Distances from atrazine application sites to which potential risk extends are listed in Table 5.11. The highest RQs are the terrestrial plant RQs, which are up to 350. Therefore, potential risks for both direct and indirect effects would be below concern levels at distances that reduce exposure such that terrestrial plant LOCs are not exceeded.

Table 5.11. Distance from Atrazine Use Site Needed to Reduce Exposure from Spray Drift to Levels that do Not Exceed LOCs

Use	Max. Allowed Application Rate (lbs a.i./Acre)	AgDrift Tier, Application Method	Distance From Treated Site Where LOCs are not Exceeded		
			Indirect Effects (DS and CRLF based on RQs for Terrestrial Plants) ^{b,c}	Direct Effects (DS and aquatic phase CRLF)	Direct Effects (Terrestrial phase CRLF)
Macadamia Nuts	4	Tier 1, Ground spray	>1000 feet	No LOC exceedances	13
Forestry, Guava	4	Tier 3 ^a , Aerial spray	>5200 feet	No LOC exceedances	88
Corn and Sorghum	2	Tier 3 ^a , Aerial spray	2900	No LOC exceedances	26
Residential Turf (spray only)	1	Tier 1, Ground spray	121	No LOC exceedances	3

^a AgDrift Tier 3 was used with default inputs except that the maximum downwind distance was expanded to 5200 feet.

^b The maximum downwind distance for ground spray for AgDrift is 1000 feet for ground spray and approximately 5200 feet for aerial spray.

^c For forestry and guava, a distance of 4900 feet is needed to reduce exposures to levels that do not exceed LOCs for the most sensitive monocot (EC25 = 0.004); however, the distance needed to reduce estimated spray drift to levels that do not exceed LOCs is greater than 5200 feet for the most sensitive dicot (EC25 = 0.003). LOC exceedance for only the most sensitive terrestrial plant tested may not be sufficient to result in indirect effects to the assessed species. However, LOC exceedance for the most sensitive terrestrial plant is conservatively used to indicate the spatial extent of potential indirect effects.

5.1.4.2 Downstream Dilution Analysis

The maximum downstream extent of atrazine exposure in streams and rivers where the EEC can potentially be above levels that would exceed the most sensitive LOC was not estimated. Given the broad scope of labeled uses and corresponding large potential area that atrazine may be applied, it is likely that multiple uses for (and applications of) atrazine may occur simultaneously within the same areas. Also, given that atrazine use could be precluded from only a few landcover classes, no credible watershed dilution can be done, and a downstream dilution analysis was not conducted.

5.1.4.3 Overlap between CRLF habitat and Spatial Extent of Potential Effects

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

Figure 5.1 and **Figure 5.2** show that there is some overlap between the initial area of concern mapped according to atrazine's use pattern and the CRLF and the DS habitat, including currently occupied core areas, CNDDDB occurrence sections, and designated critical habitat. This map does not include areas beyond the initial area of concern that may be impacted by runoff and/or spray drift. It is expected that any additional areas of

CRLF habitat that are located approximately 5200 ft (to account for offsite movement via spray drift) may also be impacted and are part of the full spatial extent of the LAA/modification of critical habitat effects determination. In addition, aquatic habitat down stream from atrazine use sites may also be impacted. However, the distance downstream of atrazine use sites where potential dilution (resulting from drainage from non-use sites) is sufficient to reduce EECs to levels that do not result in LOC exceedances has not been defined because of the large spatial extent of potential use sites in California.

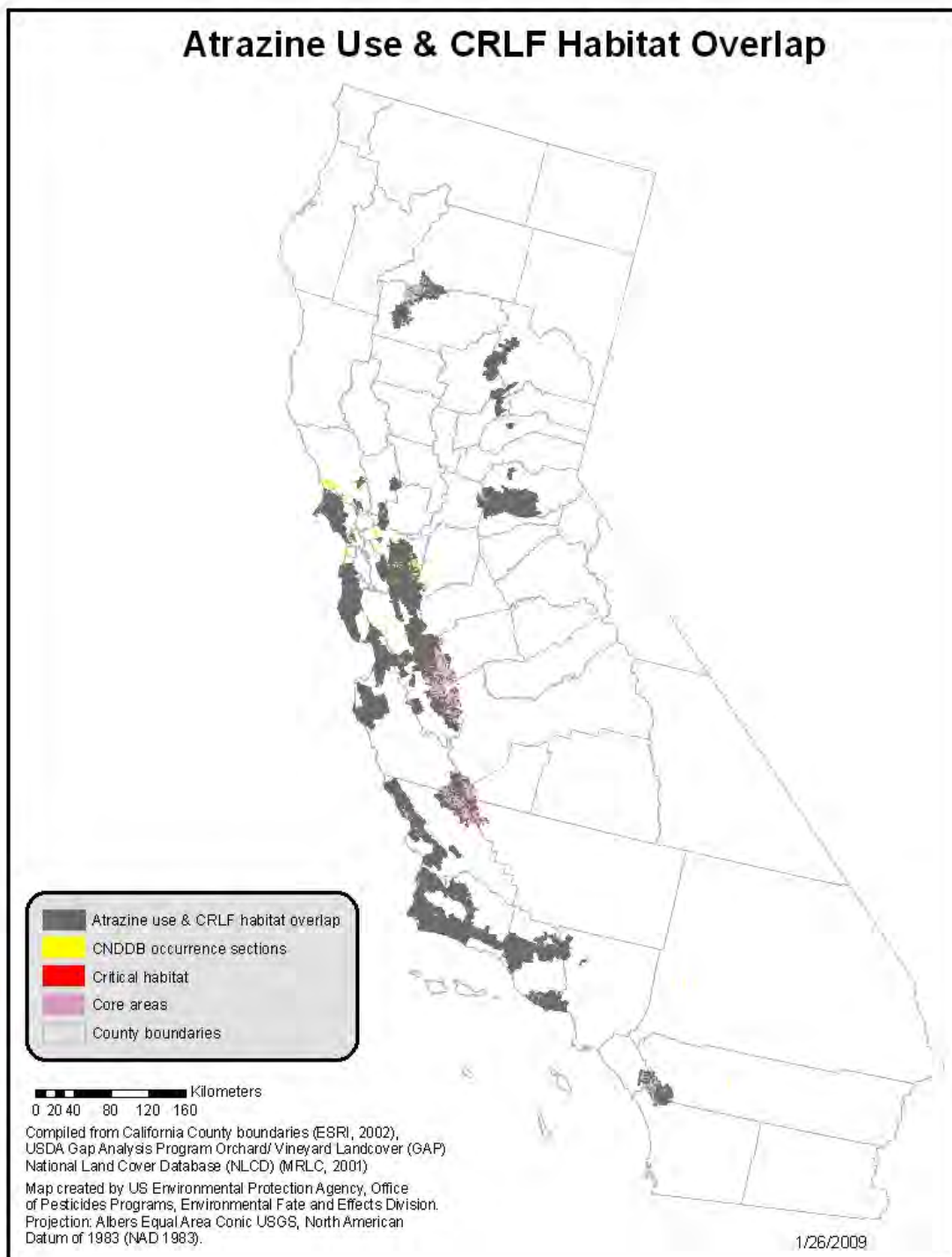


Figure 5.1. Overlap Map: CRLF Habitat and Atrazine Initial Area of Concern

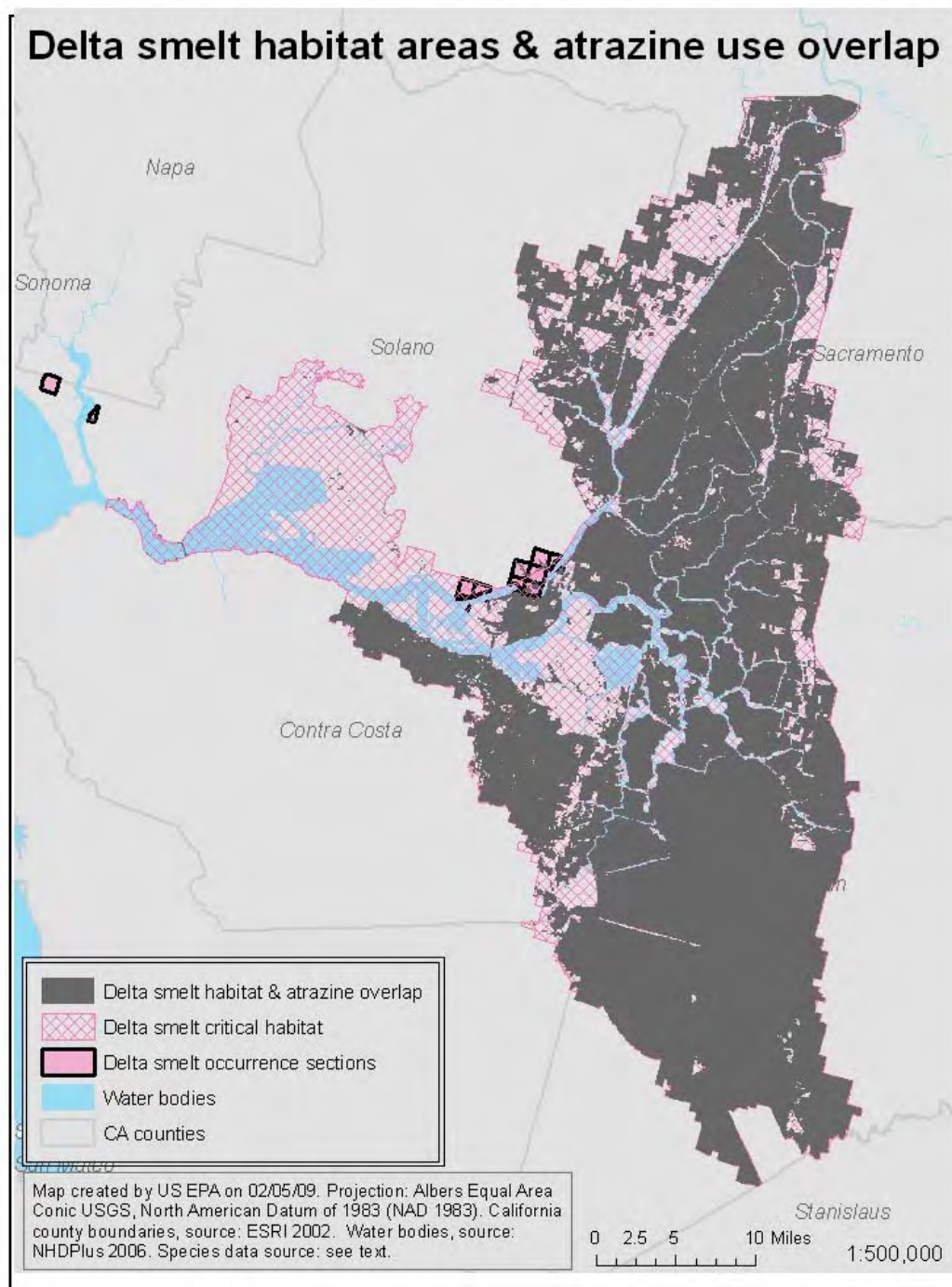


Figure 5.2. Overlap Map: DS Habitat and Atrazine Initial Area of Concern

5.2 Risk Description

The risk description synthesizes overall conclusions regarding the likelihood of adverse impacts leading to an effects determination (*i.e.*, “no effect,” “may affect, but not likely to adversely affect,” or “likely to adversely affect”) for the assessed species and the potential for modification of their designated critical habitat.

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

LOCs were not exceeded for fish. Fish are used as a surrogate for aquatic phase amphibians. Therefore, the preliminary effects determination for direct effects to the DS and aquatic phase CRLF is “no effect.”

However, acute or chronic LOCs were exceeded for birds, aquatic invertebrates, terrestrial invertebrates, aquatic plants, and terrestrial plants for all uses included in this assessment. Birds are used as a surrogate for terrestrial phase amphibians. Therefore, a preliminary “may effect” determination is made for direct effects to terrestrial phase CRLFs and for potential indirect effects to the aquatic and terrestrial phase CRLFs and the DS based on potential impacts to their habitat and food supply.

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

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

- Significance of Effect: Insignificant effects are those that cannot be meaningfully measured, detected, or evaluated in the context of a level of effect where “take” occurs for even a single individual. “Take” in this context means to harass or harm, defined as the following:
 - Harm includes significant habitat modification or degradation that results in death or injury to listed species by significantly impairing behavioral patterns such as breeding, feeding, or sheltering.
 - Harass is defined as actions that create the likelihood of injury to listed species to such an extent as to significantly disrupt normal behavior

patterns which include, but are not limited to, breeding, feeding, or sheltering.

- Likelihood of the Effect Occurring: Discountable effects are those that are extremely unlikely to occur.
- Adverse Nature of Effect: Effects that are wholly beneficial without any adverse effects are not considered adverse.

A description of the risk and effects determination for each of the established assessment endpoints for the assessed species and their designated critical habitat is provided in the following sections. The effects determination section for each listed species assessed will follow a similar pattern. Each will start with a discussion of the potential for direct effects, followed by a discussion of the potential for indirect effects. For those listed species that have designated critical habitat, the section will end with a discussion on the potential for modification to the critical habitat from the use of atrazine.

5.2.1. Direct Effects

5.2.1.1. DS and Aquatic Phase CRLFs

The potential for atrazine to directly affect the CRLF is based on the available data in amphibians and freshwater fish. The potential for atrazine to directly affect the DS is based on the available data in freshwater and saltwater fish. A substantial amount of literature is available that has evaluated effects of atrazine to amphibians and to fish. This assessment used the most sensitive acute and chronic toxicity values across fish and amphibian species tested. This is likely to result in a conservative assessment of potential risks to the CRLF because the available data suggest that amphibians are less sensitive than fish to atrazine. RQs were based on acute studies in sheepshead minnows (LC50 = 2000 ug/L) and rainbow trout (LC50 = 5300 ug/L) and a chronic NOAEC in brook trout (NOAEC = 65 ug/L).

The highest acute RQ was 0.03 based on an LC50 of 2000 ug/L in sheepshead minnows. Based on a probit slope value of 4.4 for the sheepshead minnow with 95% confidence intervals of 2.8 to 5.9 (MRID 43344901), the associated probability of an individual effects would be approximately 1 in 10^{11} (95% CI: 10^5 to 10^{19}). Acute RQs were lower for freshwater fish.

The highest chronic RQ was 0.92 based on a life cycle study in brook trout. The RQ was based on a NOAEC from an acceptable life-cycle study (MRID 00024377). A 7% reduction in length and a 16% reduction in weight was observed at LOAEC of 120 ug/L.

Also, as discussed in Section 4, data that evaluated the potential for atrazine to affect gonadal development in frogs has been discussed in two SAPs (U.S. EPA, 2003; U.S. EPA, 2007). The Agency concluded and the SAP concurred that the weight-of-evidence based on the available data does not show that atrazine produces consistent, reproducible effects across the range of exposure concentrations and amphibian species tested. In laboratory studies where environmental and animal husbandry factors were controlled,

atrazine exposures (0.01 – 100 µg/L) did not affect time to or size at metamorphosis, sex ratio, or gonadal development. While there were several effects on secondary gross and histological endpoints that were statistically significant, their relationship to apical endpoints of intersex and/or gonadal development effects is not considered relevant.

The presence of atrazine at levels thought to be sufficient to cause either direct or indirect effects was confirmed in 3 aquatic incidents. These incidents occurred in 1984, 1996, and 1998. A number of changes have been implemented to the atrazine labels since that time; therefore, it is possible that atrazine is no longer allowed to be used in a manner that is consistent with these incidents. Atrazine use was also correlated with 14 incidents where its presence in the affected water was not confirmed, but the timing of application of atrazine and other pesticides was correlated with the incident; however, atrazine levels were not confirmed or other pesticides were also implicated in the incidents. Therefore, a definitive causal relationship between atrazine use and the incident could not be established, and atrazine may or may not have contributed to the associated incident. The remaining incidents were likely caused by some factor other than atrazine such as the presence of other pesticides at levels known to be toxic to affected animals.

Two plausible scenarios exist in which atrazine applications may be responsible for the fish kills. First, atrazine concentrations in surface waters from runoff and/or spray drift may be much higher in shallow water adjacent to treated fields than estimated by models or found in monitoring studies. For example, incident I007948-012, which was associated with concentrations of atrazine >200 ppb was a small, ¼ acre pond. All other factors being equal (e.g., depth), estimated concentrations in a ¼ acre pond would be expected to be approximately 4-times higher than atrazine concentrations in the standard 1 acre ecological pond used for estimating pesticide concentrations. Second, atrazine in surface water may kill aquatic plants and the decay of dead plants may lower dissolved oxygen to levels too low for fish survival. Given the available LC50s for fish (2000 µg/L or higher, see Appendix A) are considerably higher than measured concentrations associated with the aquatic animal incidences (up to 223 µg/L), if atrazine was associated with the fish kills, the more plausible cause would be from effects on oxygen levels by reducing aquatic plant communities.

Further information on the atrazine incidents and a summary of uncertainties associated with all reported incidents are provided in Appendix H.

Therefore, the weight of evidence based on the currently available data suggest that atrazine is not expected to directly adversely affect aquatic phase CRLFs at exposure levels predicted in this assessment. The incident data suggest that atrazine could indirectly affect aquatic animals. Potential for indirect effects is evaluated in Section 5.2.2. The effects determination for direct effects to the DS and aquatic phase CRLFs is “no effect” based on the lack of LOC exceedances and the low probability of an effect occurring at the RQs presented in this assessment. The incidents of aquatic animal kills associated with atrazine use suggest that aquatic animals could be indirectly impacted as discussed further in Section 5.2.2.

Conclusion, Direct effects to the DS and aquatic phase CRLFs

Use Pattern: All labeled uses assessed

Effects Determination: No effect

Basis for Determination: No endangered species LOC exceedances; low associated probability of an individual mortality; incidents associated with fish kills are considered suggestive of potential indirect effects to aquatic animals and not of potential for direct mortality to fish.

5.2.1.2. Terrestrial Phase CRLFs

Acute and chronic LOCs were exceeded for birds. Acute RQs ranged from 0.3 (residential and turf) to 1.2 (Macadamia nuts). These RQs exceed the endangered species LOC and are associated with a probability of an individual effect of approximately 1 in 50 to 1 in 2 based on a probit slope of 3.8 (MRID 00024721).

Birds were used as surrogate species for terrestrial phase CRLFs. Terrestrial phase amphibians are poikilotherms, which means that their body temperature varies with environmental temperature, while birds are homeotherms (temperature is regulated, constant, and largely independent of environmental temperatures). As a consequence, the caloric requirements of terrestrial phase amphibians are markedly lower than birds. Therefore, on a daily dietary intake basis, birds consume more food than terrestrial phase amphibians. This can be seen when comparing the caloric requirements for free living iguanid lizards (used in this case as a surrogate for terrestrial phase amphibians) to song birds (U.S. EPA, 1993):

$$\text{iguanid FMR (kcal/day)} = 0.0535 (\text{bw g})^{0.799}$$

$$\text{passerine FMR (kcal/day)} = 2.123 (\text{bw g})^{0.749}$$

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

Because the existing risk assessment process is driven by the dietary route of exposure, a finding of safety for birds, with their much higher feeding rates and, therefore, higher potential dietary exposure, is reasoned to be protective of terrestrial phase amphibians. For this not to be the case, terrestrial phase amphibians would have to be 40 times more sensitive than birds for the differences in dietary uptake to be negated. However, existing dietary toxicity studies in amphibians are lacking. To quantify the potential differences in food intake between birds and terrestrial phase CRLFs, food intake equations for the iguanid lizard replaced the food intake equation in T-REX for birds, and additional food items of the CRLF were evaluated. These functions were encompassed in a model called T-HERPS. T-HERPS is available at: <http://www.epa.gov/oppefed1/models/terrestrial/index.htm>. Results of this analysis are presented in Table 5.12 (use pattern with lowest EECs and RQs) and 5.13 (use pattern with highest EECs and RQs).

Table 5.12. Upper Bound Kenaga, Acute Terrestrial Herpetofauna Dose-Based Risk Quotients (Turf and Residential; 1 lbs a.i./Acre, 2 applications, 30-day interval)

Size Class (grams)	LD50	EECs and RQs									
		Broadleaf Plants/ Small Insects		Fruits/Pods/ Seeds/ Large Insects		Small Herbivore Mammals		Small Insectivore Mammal		Small Amphibians	
		EEC	RQ	EEC	RQ	EEC	RQ	EEC	RQ	EEC	RQ
1.4	940	6.79	0.01	0.75	<0.01	N/A	N/A	N/A	N/A	N/A	N/A
100		5.32	0.01	0.59	<0.01	71.64	0.08	4.48	<0.01	0.18	<0.01
238		4.37	<0.01	0.49	<0.01	30.10	0.03	1.88	<0.01	0.15	<0.01

Table 5.13. Upper Bound Kenaga, Acute Terrestrial Herpetofauna Dose-Based Risk Quotients (Macadamia Nuts; 4 lbs a.i./Acre, 2 applications, 30-day interval)

Size Class (grams)	LD50	EECs and RQs									
		Broadleaf Plants/ Small Insects		Fruits/Pods/ Seeds/ Large Insects		Small Herbivore Mammals		Small Insectivore Mammal		Small Amphibians	
		EEC	RQ	EEC	RQ	EEC	RQ	EEC	RQ	EEC	RQ
1.4	940	27	0.03	3.0	<0.01	N/A	N/A	N/A	N/A	N/A	N/A
37		27	0.03	3.0	<0.01	N/A	N/A	N/A	N/A	0.93	<0.01
100		21	0.02	2.37	<0.01	287	0.30	18	0.02	0.74	<0.01
238		17	0.02	1.9	<0.01	120	0.13	7.5	0.01	0.61	<0.01

These data suggest that dietary exposures from consumption of insects by terrestrial phase CRLFs is likely to result in exposures that do not exceed LOCs. However, the endangered species LOC was exceeded for large terrestrial phase CRLFs that consume small herbivorous mammals. The California mouse is a prey item of the CRLF, and this mouse reportedly eats grasses. Also, the metabolism data in mammals suggests that atrazine elimination in rats is not exceedingly fast such that secondary exposure to the CRLF is unlikely. However, some assumptions included in the assessment of small mammals as a food item are conservative because the CRLF eats a variety of food items and LOCs were not exceeded for CRLFs that eat insectivorous mammals. The RQ for a 100-gram CRLF that consumes a 35-gram mammal that recently consumed contaminated short grass was 0.3 for macadamia nuts. RQs remain above the endangered species LOC for CRLFs that consume herbivorous mammals for all uses except turf and residential uses (spray applications, Table 5.9). However, potential risks from granular formulations for turf and residential uses could not be precluded because granular formulations are applied at higher rates. Also, the LD50/sq. ft. analysis assumes multiple exposure pathways occurs; therefore, considering only differences in food intake between birds and CRLFs would not affect risk conclusions.

Current assessment methods of potential risks from chronic exposures for birds do not consider food intake levels. Therefore, T-HERPS does not quantify potential effects of reduced food intake of terrestrial phase CRLFs relative to birds for chronic risk assessments. However, chronic RQs would be expected to be reduced by a similar magnitude seen in the acute analysis.

Therefore, potential effects to CRLFs that consume herbivorous mammals cannot be precluded, and the effects determination for direct effects to the CRLF is likely to adversely affect.

Conclusion, Direct effects to terrestrial phase CRLFs

Use Pattern: All labeled uses assessed

Effects Determination: Likely to adversely affect

Basis for Determination: LOC exceedances for birds; refinements incorporating dietary habits of CRLFs did not preclude potential risks for CRLFs that consume herbivorous mammals.

5.2.2. Indirect Effects, DS and Aquatic Phase CRLF

As discussed in Section 2, the diet of aquatic-phase CRLF tadpoles and DS larvae is composed primarily of unicellular aquatic plants (*i.e.*, algae and diatoms) and detritus. However, aquatic invertebrates are also consumed by both CRLFs and the DS, and fish are consumed by adult CRLFs. Therefore, potential impacts to each of these potential food items are evaluated.

5.2.2.1. Potential Impacts to Fish (CRLF only)

Fish are food items of the CRLF. The effects assessment for potential direct impacts to the DS and aquatic phase CRLFs was “no effect.” This conclusion was based on the most sensitive fish acute and chronic toxicity values. Because fish are not expected to be directly impacted, potential reductions in fish as a food source of the CRLF is not expected. Therefore, the effects determination to CRLFs resulting from impacts to fish as a dietary item is also “no effect.”

5.2.2.2. Potential Impacts to Aquatic Invertebrates

CRLF

The acute LOC of 0.5 was not exceeded for freshwater invertebrates based on toxicity values from the most sensitive freshwater species (scud, MRID 00024377). The highest acute RQ was 0.09. At this RQ, the probability of an effect would be approximately 1 in 500,000. Based on the low anticipated direct impacts to the most sensitive freshwater invertebrates, any potential impact to aquatic phase CRLFs would be immeasurable in the environment and would, therefore, constitute an insignificant effect. Therefore, exceedance of the endangered species LOC suggests that there could be some effect to

sensitive aquatic invertebrates; however, such an effect would be insignificant to the CRLF. Therefore, the effects determination is “not likely to adversely affect.”

DS

The DS eats small zooplankton. They primarily eat planktonic copepods, cladocerans, amphipods, and insect larvae. However, the most important food organism appears to be *Eurytemora affinis*, which is a euryhaline copepod (USFWS, 1995 and 2004 b). An LC50 of 125 ug/L (Ecotox No. 80951) and 500 ug/L (MRID 45208303 and 45227711) was reported for *E. affinis*. However, LC50s in another saltwater copepod, *A. tonsa*, ranged from 88 ug/L to 4300 ug/L. RQs were based on the most sensitive LC50 of 88 ug/L. RQs ranged from 0.2 to 0.7. The probit slope was shallow for this species (slope = 0.94), which resulted in an estimated probability of an individual effect to range from 1 in 4 to 1 in 2 at RQs of 0.2 to 0.7. These levels of effect could result in a magnitude of effect to the copepod that could result in indirect effects to the DS. The two acute LC50s for *E. affinis* fall within the range of LC50s for the most sensitive saltwater copepod species tested, *A. tonsa*.

The probit dose-response slopes for the copepod species tested have consistently produced shallow dose-response curves. Of the four available probit dose-response slopes, 3 are less than a slope of 1. Assuming a probit slope of 1.0 results in a probability of an individual effect of 1 in 10 at an RQ of 0.05. RQs for all uses modeled in this assessment exceeded 0.05 for both copepod species tested.

Based on LOC exceedances and the estimated probability of an individual effect of 10% or more for the predominant food item of the DS, it is concluded that labeled uses of atrazine could result in adverse effects to the DS resulting from potential impacts to food abundance.

Acceptable chronic studies in copepods are not currently available. Chronic RQs for saltwater invertebrates were based on an estimated NOAEC in copepods. The chronic LOC would not be exceeded based on the most sensitive NOAEC in mysid shrimp of 80 ug/L. However, the copepod was shown to be more sensitive on an acute basis than the mysid shrimp. An open literature study was located that evaluated chronic exposures to copepods (Ecotox No. 73333). In this study, percent reproductive failure occurred at 25 ug/L and higher, and total viable offspring production per female was significantly decreased at 2.5 ug/L and higher. These data suggest that the chronic LOC would be exceeded for all uses. However, the reliability of this study is questionable given the reporting deficiencies and use of DMSO as a solvent (Appendix A), which may increase its toxicity. As reported in Appendix A, the most sensitive mysid shrimp LC50 was 1000 ug/L (MRID 45202920). Based on a chronic NOAEC of 80 ug/L in mysid shrimp (MRID 45202920), an acute to chronic ratio (ACR) of 12.5 could be derived. Applying an ACR of 12.5 to the acute LC50 of 88 ug/L results in an estimated NOAEC of 7 ug/L. 21-Day EECs estimated for this assessment were approximately 2 to 9 times higher than this estimated NOAEC.

Conclusion:**CRLF**

Use Pattern: All uses

Effects Determination: Not likely to adversely affect (NLAA)

Basis for Determination: Potential impacts to freshwater invertebrates are not likely to be sufficient to result in measureable impacts to the CRLF resulting from a reduction in food. Therefore, although the endangered species LOC is exceeded for the most sensitive freshwater invertebrates, resulting potential impacts to the CRLF are considered insignificant, and a “not likely to adversely affect” determination is made for aquatic phase CRLFs resulting from potential impacts to aquatic invertebrate food base.

DS

Use Pattern: All uses

Effects Determination: Likely to adversely affect (LAA)

Basis for Determination: Copepods were reported to be the most important food source for the DS. Copepod abundance could potentially be impacted to an extent that could adversely affect the DS. Therefore, the effects determination for the DS based on potential reductions in food is “likely to adversely affect.”

5.2.2.3. Potential Impacts To Aquatic Plants

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

Algal RQs ranged from approximately 17 to 64, which means that the EECs calculated for this assessment are 17 to 64 times higher than the most sensitive algal EC50 of 1 ug/L. The IRED (U.S. EPA, 2003) indicated that community level effects are likely to occur at concentrations that exceed 10 to 20 ug/L for an extended period of time or on a recurrent basis. Although the duration that constitutes an extended period of time was not defined, the peak, 21-day, and 60-day EECs all exceed 10 ug/L for all uses included in this assessment. However, residential EECs from highly urbanized areas were considerably lower than other EECs and ranged from 5 ug/L to 9 ug/L. Therefore, potential indirect effects may not occur in more urbanized areas with more impervious surfaces.

There is no known obligate relationship between the CRLF or the DS and any particular aquatic plant species. Therefore, if less sensitive aquatic plant species are present, the CRLF may not be adversely impacted by atrazine at the EECs presented in this

assessment. Non-vascular aquatic plant EC50s ranged from approximately 1 ug/L to 300 ug/L. The median value of data in Appendix A for non-vascular plant studies is 50 ug/L with 25th and 75th percentile values of 23 ug/L and 110 ug/L, respectively. The presence of less tolerant aquatic plant species could result in less severe impacts to aquatic plant food sources. A tool (CASM) is available that may be used to further refine atrazine concentrations that are associated with community level effects to aquatic plants. This tool accounts for parameters such as species sensitivity. However, as previously described, this tool is currently being refined based on input from the SAP earlier in 2008. These refinements are going to a second SAP in May, 2009. Because CASM is currently being refined, it was not used for this assessment, but may be used to further characterize potential effects to the CRLF after it has been discussed by the SAP in May, 2009.

Conclusion, Indirect effects to DS and aquatic phase CRLFs from Impacts to aquatic plants

Use Pattern: All labeled uses assessed

Effects Determination: Likely to adversely affect

Basis for Determination: The best available information suggests that labeled atrazine use could impact the CRLF and the DS based on potential impacts to aquatic plant food items and water quality resulting from changes in the aquatic or riparian plant community.

5.2.3. Indirect Effects, Dietary Items of Terrestrial Phase CRLFs

As discussed in Section 2, the diet of terrestrial-phase CRLFs includes terrestrial invertebrates, small mammals, and amphibians. Potential impacts to each of these potential food items are evaluated.

5.2.3.1. Terrestrial Invertebrates

Studies that showed statistically significant ($p < 0.05$) effects to terrestrial invertebrates were typically at levels that were above highest labeled application rates used in this assessment. The most sensitive terrestrial insect tested was the springtail (*Onychiuridae*). Mortality rates in *Onychiurus armatus* were approximately 50% at 20 ppm soil, which is associated with an application rate of 7 lbs a.i./Acre assuming a soil depth of 3 cm and a soil density of 1.3 g/cm³. Another species of springtail, *O. armatus*, was associated with 18% mortality at soil levels associated with approximately 1 lb a.i./Acre (Mola et al., 1987), which is within the range of labeled atrazine application rates. An application rate of 5.4 lbs a.i./Acre was associated with reduced abundance of microarthropods (Fratello et. al., 1985); however, reduced abundance could have been caused by indirect effects (migration/repellency). Application rates of 0.9 and 1.8 lbs a.i./Acre did not affect abundance of microarthropods (Cortet et al., 2002; Fratello et. al., 1985).

Atrazine did not affect survival in a number of beetle species at application rates that ranged from 0.8 to 8 lbs a.i./Acre (Kegel, 1989; Brust, 1990; Samsoe-Petersen, 1995). No studies in beetles established definitive LOAEC or EC50 values. Because the studies

in beetles produced free-standing NOAECs, their utility is somewhat limited; however, they do suggest that abundance would not likely be affected to an extent that would result in indirect effects to the CRLF at atrazine applications up to 8 lbs a.i./Acre for ground beetles (*Poecilus*) and 2 lbs a.i./Acre for carabid beetles.

In addition, earthworm LC50s were 270 and 380 ppm soil (Mosleh et al., 2003; Haque and Ebing, 1983). The highest soil concentrations expected from the maximum labeled application rate (4 lbs a.i./Acre) on the treated field would be approximately 11 ppm in the top 3 cm of soil (RQ would be approximately 0.04).

Also, the acute contact LD50 in honey bees was >97 ug/bee (5% mortality occurred at the highest dose level) (MRID 00036935). A dose of 97 ug/bee corresponds to an atrazine concentration on the bee of approximately 757 ppm, assuming an adult honey bee weighs 128 mg (Mayer and Johansen, 1990). The corresponding exposure value to honey bees at an application rate of 4 lbs a.i./Acre is approximately 60 ppm. Although the resulting RQ (0.079) would be above the interim LOC for endangered terrestrial invertebrates of 0.05, the resulting probability of an individual mortality would be approximately 1 in 3,000,000 assuming a probit slope of 4.5. The default probit slope was used because insufficient mortality occurred at the highest dose tested in the honey bee study (MRID 00036935) to allow for a calculation of a probit slope.

Conclusion, Indirect effects to DS and aquatic phase CRLFs from potential impacts to terrestrial invertebrates

Use Pattern: All labeled uses assessed

Effects Determination: Not likely to adversely affect

Basis for Determination: The available data suggest that some species of terrestrial invertebrates could be directly or indirectly affected by atrazine at labeled application rates. However, the magnitude of such effects for many species of terrestrial invertebrates is not likely to result in indirect effects to the CRLF.

5.2.3.2. *Mammals*

Terrestrial phase CRLFs consume small mammals. This assessment used a 15-gram herbivorous mammal to determine if there could be a potential reduction in mammal abundance. Acute RQs for a 15-gram mammal ranged from 0.06 to 0.3 depending on the use pattern. Assuming a probit slope of 4.5, the probability of an individual effect would be approximately 1 in 170. Assuming that probability of an individual effect provides insight into the potential for reductions in a local population of small mammals, a probability of 1 in 170 (<1%) would result in an immeasurable impact to mammal abundance and would, therefore, constitute an insignificant effect.

However, reproduction RQs ranged from 28 to 150. The toxicity endpoint used in the RQs was based on a NOAEL of 3.8 mg/kg-bw. The LOAEL was 39 mg/kg/day in males, 43 mg/kg/day in females based on growth effects. The RQs indicate that potential

exposures exceed the LOAEC for all uses. Also, a similar NOAEL was observed for short-term exposures based on developmental delays in female and male adolescent rats at 6.25 mg/kg/day (U.S. EPA, 2003). It is difficult to determine if these effects would impact mammalian abundance to an extent that could result in indirect effects to the CRLF; however, such impacts cannot be precluded. Therefore, the effects determination for the potential for atrazine to affect the CRLF from potential impacts to mammalian prey is likely to adversely affect.

Conclusion, Indirect effects to DS and aquatic phase CRLFs from potential impacts to mammals

Use Pattern: All labeled uses assessed

Effects Determination: Likely to adversely affect

Basis for Determination: Exceedances of chronic LOCs.

5.2.3.3. *Amphibians*

The effects determination for potential direct effects to aquatic phase CRLFs was “no effect.” Therefore, the effects determination is also “no effect” for terrestrial phase amphibians based on potential reductions in abundance of aquatic phase amphibians as a food source.

The effects determination for terrestrial phase CRLFs was “likely to adversely affect”. This determination was based solely on frogs that consume potentially contaminated herbivore mammals. Terrestrial amphibian prey of the CRLF include small amphibians such as tree frogs that do not prey on mammals. Therefore, the mammalian food group is not relevant in the evaluation of potential reductions in amphibian prey abundance. Although bird RQs exceeded LOCs (Section 5.1), LOCs were not exceeded for insectivorous amphibians using the T-HERPS model. Therefore, reductions in amphibian prey at levels likely to affect the CRLF are not likely to occur, and the effects determination for the CRLF based on potential reductions in terrestrial amphibians is “not likely to adversely affect.”

Conclusion, Indirect effects to DS and aquatic phase CRLFs from potential impacts to terrestrial phase amphibians.

Use Pattern: All labeled uses assessed

Effects Determination: Not likely to adversely affect

Basis for Determination: Bird LOCs were exceeded; however, incorporation of food consumption data more appropriate for amphibians indicates that terrestrial phase amphibians are not likely to be impacted by exposure to atrazine to an extent that is expected to impact CRLFs.

5.2.3.4. **Overall Conclusions, Indirect effects (impacts to dietary items) to terrestrial phase CRLFs**

Use Pattern: All labeled uses assessed

Effects Determination: Likely to adversely affect

Basis for Determination: Potential effects to small, herbivorous and insectivorous mammals; other food items such as terrestrial invertebrates and amphibians are not likely to be impacted to an extent that would affect CRLFs.

5.2.4. Indirect Effects, Potential Impacts to Terrestrial Plants

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

As shown in Tables 5.8 and 5.9, seedling emergence or vegetative vigor RQs exceed LOCs for a number of the tested plant species. Based on exceedance of the seedling emergence LOCs for all species tested except corn, the following general conclusions can be made with respect to potential harm to riparian habitat via runoff exposures:

- Atrazine may enter riparian areas via runoff where it may be taken up through the root system of sensitive plants.
- Comparison of seedling emergence EC₂₅ values to EECs estimated using TERRPLANT suggests that inhibition of new growth may occur. Inhibition of new growth could result in degradation of high quality riparian habitat over time because as older growth dies from natural or anthropogenic causes, plant biomass may be prevented from being replenished in the riparian area. Inhibition of new growth may also slow the recovery of degraded riparian areas that function poorly due to sparse vegetation because atrazine deposition onto bare soil would be expected to inhibit the growth of new vegetation.
- Because LOCs were exceeded for most species tested (9/10) in the seedling emergence studies, it is likely that many species of herbaceous plants could be potentially affected by exposure to atrazine in runoff.

Because RQs for terrestrial plants are above the Agency's LOCs, atrazine use is considered to have the potential to directly impact plants in riparian areas, potentially resulting in degradation of stream water quality via sedimentation and alteration of habitat. Therefore, an analysis of the potential for habitat degradation to affect the CRLF and the DS is necessary.

Riparian plants beneficially affect water and stream quality in a number of ways in both adjacent river reaches and areas downstream of the riparian zone. Riparian vegetation provides a number of important functions in the stream/river ecosystem, including the following:

- serves as an energy source;
- provides organic matter to the watershed;
- provides streambank stability;
- provides shading, which ensures thermal stability of the stream; and
- serves as a buffer, filtering out sediment, nutrients, and contaminants before they reach the stream.

A general discussion of riparian habitat and its relevance to the CRLF and the DS is provided below. Additional details are presented in Appendix I.

It is difficult to estimate the magnitude of potential impacts of atrazine use on riparian habitat and the magnitude of potential effects on stream water quality from such impacts as they relate to survival, growth, and reproduction of the CRLF. The level of exposure and any resulting magnitude of effect on riparian vegetation are expected to be highly variable and dependent on many factors. The extent of runoff and/or drift into stream corridor areas is affected by the distance the atrazine use site is offset from the stream, local geography, weather conditions, and quality of the riparian buffer itself. The sensitivity of the riparian vegetation is dependent on the susceptibility of the plant species present to atrazine and composition of the riparian zone (e.g. vegetation density, species richness, height of vegetation, width of riparian area).

Quantification of risk to the CRLF and the DS from potential effects to riparian areas is precluded by the following factors:

- The relationship between distance of soil input into the watershed and sediment deposition in areas critical to survival, reproduction, and growth of the CRLF and the DS is not known; and
- Riparian areas within the action area are highly variable in their composition and location with respect to atrazine use; therefore, their sensitivity to potential damage is also variable.

In addition, even if plant community structure was quantifiably correlated with riparian function, it may not be possible to discern the effects of atrazine on species composition separate from other agricultural actions or determine if atrazine is a significant factor in altering community structure. Plant community composition in agricultural field margins is likely to be modified by many agricultural management practices. Vehicular impact and mowing of field margins and off-target movement of fertilizer and herbicides are all likely to cause changes in plant community structure of riparian areas adjacent to agricultural fields (Jobin et al., 1997; Kleijn and Snoeijs, 1997; Schippers and Joenje, 2002). Although herbicides are commonly identified as a contributing factor to changes in plant communities adjacent to agricultural fields, some studies identify fertilizer use as

the most important factor affecting plant community structure near agricultural fields (e.g. Schippers and Joenje, 2002) and community structure is expected to be affected by a number of other factors (de Blois et al., 2002). Thus, the effect of atrazine alone on riparian community structure is complicated by other multiple stressors likely to occur within the action area.

In summary, terrestrial plant RQs are above terrestrial plant LOCs for all uses; therefore, labeled use of atrazine has the potential to affect riparian vegetation within the CRLFs habitats. However, water quality and sedimentation / siltation in a stream may depend on numerous factors, and determining whether atrazine use is expected to result in an overall increase in sediment/silt levels in a habitat is difficult. Until further analysis is performed on specific land management practices and sensitivity of riparian vegetation in areas surrounding the CRLF habitat, potential effects to riparian vegetation as indicated by terrestrial plant RQ exceedance, it is presumed to potentially adversely affect the CRLF, the DS, and their designated critical habitat

Because woody plants are typically not sensitive to atrazine at expected exposure concentrations, riparian areas that are predominantly woody shrubs and trees are not likely to be adversely impacted by atrazine use to an extent that would be expected to result in measurable effects on the CRLF. Therefore, atrazine is not likely to adversely affect CRLFs in watersheds with predominantly forested riparian areas.

Therefore, habitats of the CRLF and the DS that are in close proximity to potential atrazine use sites and where the riparian vegetation is comprised of sensitive grasses and non-woody plants, the effects determination is “likely to adversely affect.” A graphic representation of the effects determination for this assessment endpoint, based on evaluation of the sedimentation, streambank stability, and thermal stability attributes for riparian vegetation is provided in Figure 5.2.

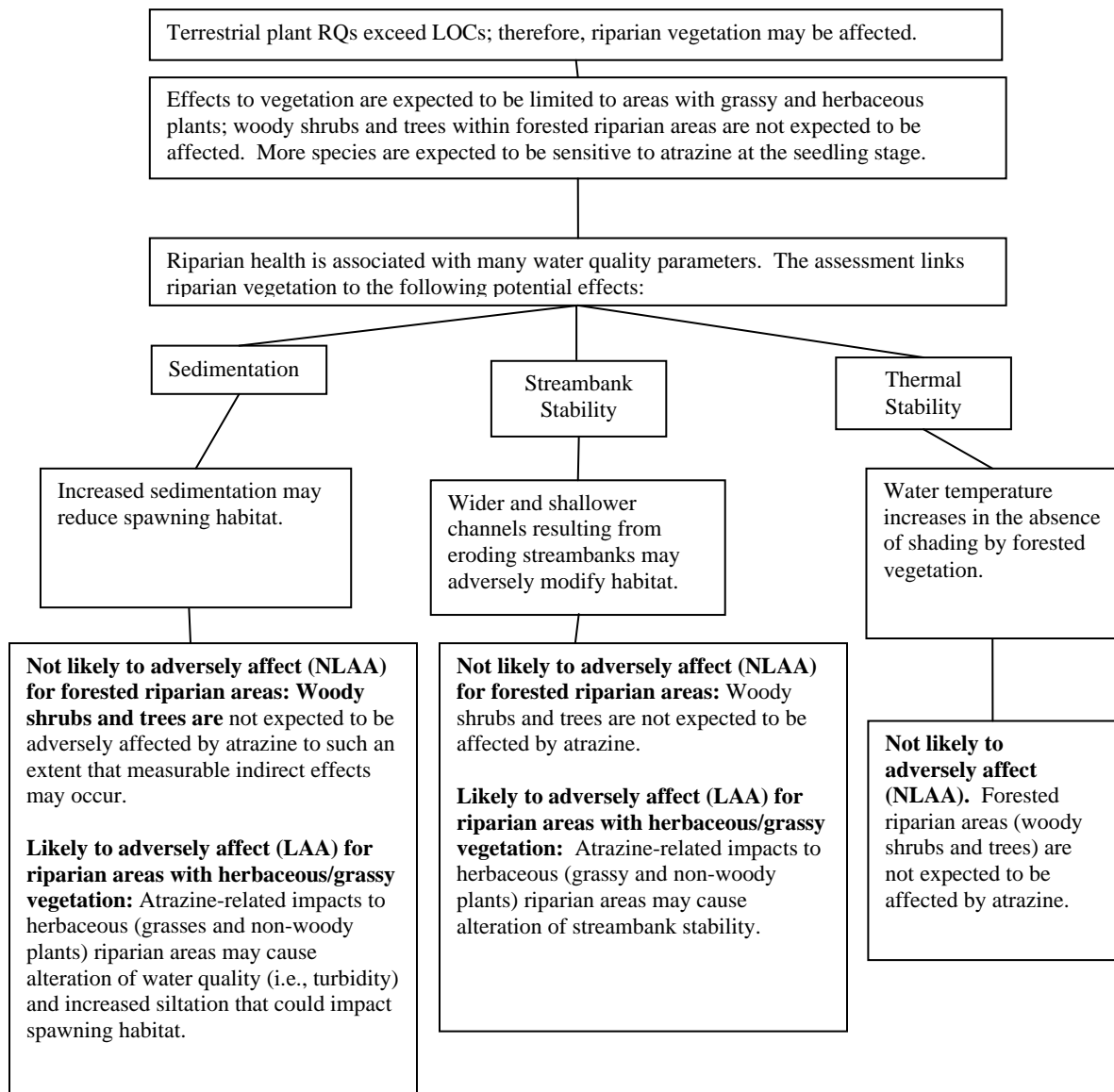


Figure 5.2 Summary of the Potential of Atrazine to Affect the CRLF and the DS via Riparian Habitat Effects

5.3. Modification of Designated Critical Habitat

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

5.3.1. CRLF

5.3.1.1. Aquatic-Phase PCEs

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

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

Conclusions for potential indirect effects to the CRLF via direct effects to aquatic and terrestrial plants are used to determine whether modification to critical habitat may occur. As previously discussed, atrazine may cause habitat modification by potentially impacting aquatic plants and terrestrial plants.

The remaining aquatic-phase PCE is “alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.” Atrazine may impact algae as food items for tadpoles. Atrazine may also impact riparian areas that are predominantly grassy or herbaceous. Therefore, there is a potential for habitat modification by potentially impacting the chemical characteristics of the habitat.

5.3.1.2. Terrestrial-Phase PCEs

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

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

As an herbicide, atrazine may affect sensitive terrestrial plants. Terrestrial plant LOCs were exceeded for all uses.

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

The fourth terrestrial-phase PCE is based on alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source. There is a potential for habitat modification based on potential direct (Section 5.2.1) and indirect effects (Sections 5.2.2) to terrestrial-phase CRLFs.

5.3.2. DS

Primary constituent elements (PCEs) of designated critical habitat for the DS include the following:

- Spawning Habitat—shallow, fresh or slightly brackish backwater sloughs and edgewaters to ensure egg hatching and larval viability. Spawning areas also must provide suitable water quality (*i.e.*, low “concentrations of pollutants”) and substrates for egg attachment (*e.g.*, submerged tree roots and branches and emergent vegetation).
- Larval and Juvenile Transport—Sacramento and San Joaquin Rivers and their tributary channels must be protected from physical disturbance and flow disruption. Adequate river flow is necessary to transport larvae from upstream spawning areas to rearing habitat in Suisun Bay. Suitable water quality must be provided so that maturation is not impaired by pollutant concentrations.
- Rearing Habitat—Maintenance of the 2 ppt isohaline and suitable water quality (low concentrations of pollutants) within the Estuary is necessary to provide DS larvae and juveniles a shallow protective, food-rich environment in which to mature to adulthood.
- Adult Migration— Unrestricted access to suitable spawning habitat in a period that may extend from December to July. Adequate flow and suitable water quality may need to be maintained to attract migrating adults in the Sacramento and San Joaquin River channels and their associated tributaries. These areas also should be protected from physical disturbance and flow disruption during migratory periods.
- PCEs also include more general requirements for habitat areas that provide essential life cycle needs of the species such as space for individual and population growth and for normal behavior; food, water, air, light, minerals, or other nutritional or physiological requirements; cover or shelter; sites for breeding, reproduction, rearing (or development) of offspring; and habitats that are protected from disturbance or are representative of the historic geographical and ecological distributions of a species.

The effects determination for direct effects to the DS was “no effect.” However, it was concluded that atrazine is likely to adversely affect the DS by potentially affecting its habitat (aquatic and terrestrial plants) and its food (copepods). Therefore, atrazine may also modify critical habitat of the DS that is located in close proximity to atrazine use sites.

6. Uncertainties

6.1 Exposure Assessment Uncertainties

6.1.1 Maximum Use Scenario

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

6.1.2 Impact of Vegetative Setbacks on Runoff

Unlike spray drift, models 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 (USDA, NRCS, 2000). 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.

6.1.3 Aquatic Exposure Modeling of Atrazine

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, some organisms may inhabit water bodies of different size and depth and/or are located adjacent to larger or smaller drainage areas than the EXAMS pond. In addition, the Services agree that the existing EXAMS pond represents the best currently available approach for estimating aquatic exposure to pesticides (USFWS/NMFS 2004).

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

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

Unlike spray drift, tools are currently not available to evaluate the effectiveness of a vegetative setback on runoff and loadings. The effectiveness of vegetative setbacks is highly dependent on the condition of the vegetative strip. For example, a well-established, healthy vegetative setback can be a very effective means of reducing runoff

and erosion from agricultural fields. Alternatively, a setback of poor vegetative quality or a setback that is channelized can be ineffective at reducing loadings. Until such time as a quantitative method to estimate the effect of vegetative setbacks on various conditions on pesticide loadings becomes available, the aquatic exposure predictions are likely to overestimate exposure where healthy vegetative setbacks exist and underestimate exposure where poorly developed, channelized, or bare setbacks exist.

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

6.1.4. Uncertainties regarding dilution and chemical transformations in estuaries

PRZM-EXAMS modeled EECs were initially calibrated to represent relatively small ponds and low-order streams. Therefore it would seem likely that results from the PRZM-EXAMS model should greatly over-estimate potential concentrations in much larger receiving water bodies such as estuaries, embayments, and coastal marine areas; chemicals in runoff water (or spray drift, etc.) should simply be diluted by a much larger volume of water than would be found in the 'typical' EXAMS pond. However, as chemical constituents in water draining from freshwater streams encounter brackishness or other near-marine-associated conditions, there is potential for important chemical transformations to occur. Many chemical compounds can undergo changes in mobility, toxicity, or persistence when changes in pH, Eh, salinity, dissolved oxygen (DO) content, or temperature are encountered. For example, desorption and re-mobilization of some chemicals from sediments can occur with changes in salinity (e.g., Means 1995; Swarzenski et al. 2003; Jordan et al. 2008), changes in pH (e.g., Wood and Baptista 1993; Parikh et al. 2004; Fernandez et al. 2005), Eh changes (Wood and Baptista 1993; Velde and Church 1999), and other factors. Thus, although chemicals in discharging rivers may be diluted by large volumes of water within receiving estuaries and embayments, the hydrochemistry of the marine-influenced water may negate some of the attenuating impact of the greater water volume; for example, the effect of dilution may be partly counteracted by increased mobility of a chemical in brackish water. In addition, freshwater contributions from discharging streams and rivers do not instantaneously mix with more saline water bodies. In these settings, water will commonly remain highly stratified, with fresh water lying atop denser, heavier saline water – meaning that exposure to concentrations found in discharging stream water may propagate some distance beyond the outflow point of the stream (especially near the water surface). Therefore, EFED does not automatically assume that discharging water will be rapidly diluted by the entire water volume within an estuary, embayment, or other coastal aquatic environment; PRZM-EXAMS model results should be considered consistent with concentrations that might be found near the head of an estuary unless there is specific information to indicate otherwise. Conditions nearer to the mouth of a bay or estuary,

however, may be closer to a marine-type system, and thus more subject to the notable buffering, mixing, and diluting capacities of an open marine environment. Conversely, tidal effects (pressure waves) can propagate much further upstream than the actual estuarine water, so discharging river water may become temporarily partially impounded near the mouth (discharge point) of a channel, and resistant to mixing until tidal forces are reversed.

6.1.4 Usage Uncertainties

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

6.1.5 Terrestrial Exposure Modeling of Atrazine

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

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

Differences in assimilative efficiency between laboratory and wild diets suggest that current screening assessment methods do not account for a potentially important aspect of

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

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

6.1.6 Spray Drift Modeling

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

6.2 Effects Assessment Uncertainties

6.2.1 Age Class and Sensitivity of Effects Thresholds

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

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

6.2.2 Impact of Multiple Stressors on the Effects Determination

The influence of length of exposure and concurrent environmental stressors to the CRLF and the DS (i.e., construction of dams and locks, fragmentation of habitat, change in flow regimes, increased sedimentation, degradation of quantity and quality of water in the watersheds of the action area, predators, etc.) will likely affect the species' response to atrazine. Additional environmental stressors may increase sensitivity to the herbicide, although there is the possibility of additive/synergistic reactions. Timing, peak concentration, and duration of exposure are critical in terms of evaluating effects, and these factors are expected to 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. However, as previously discussed, the Agency's LOCs are set to be protective given the wide range of possible uncertainties.

6.2.3 Use of Surrogate Species Effects Data

Freshwater fish are used as surrogate species for aquatic-phase amphibians. Data are available on atrazine that evaluated its toxicity to amphibians. Overall, these data do not suggest that amphibians are more sensitive than fish to atrazine. Therefore, endpoints based on freshwater fish ecotoxicity data are assumed to be protective of potential direct effects to aquatic-phase amphibians including the CRLF, and extrapolation of the risk conclusions from the most sensitive tested species to the aquatic-phase CRLF is likely to overestimate the potential risks to those species. Efforts are made to select the organisms most likely to be affected by the type of compound and usage pattern; however, there is an inherent uncertainty in extrapolating across phyla. In addition, the Agency's LOCs are intentionally set very low, and conservative estimates are made in the screening level risk assessment to account for these uncertainties.

6.2.4 Sublethal Effects

The assessment endpoints used in ecological risk assessment include potential effects on survival, growth, and reproduction of the CRLF and the DS and organisms on which these species depend for survival and reproduction such as invertebrates. A number of studies were located that evaluated potential sublethal effects to fish from exposure to atrazine. Although several studies reported toxicity values that were less sensitive than the submitted studies, they were not used for use in risk estimation. In particular, fish

studies were located in the open literature that reported effects on endpoints other than survival, growth, or reproduction at concentrations that were considerably lower than the most sensitive endpoint from submitted studies.

Upon evaluation of the available studies, however, the most sensitive NOAEC from the submitted full life-cycle studies was considered to be the most appropriate chronic endpoint for use in risk assessment. In the full life cycle study, fish are exposed to atrazine from one stage of the life cycle to at least the same stage of the next generation (e.g. egg to egg). Therefore, exposure occurs during the most sensitive life stages and during the entire reproduction cycle. Four life cycle studies have been submitted in support of atrazine registration. Species tested include brook trout, bluegill sunfish, and fathead minnows. The most sensitive NOAEC from these studies was 65 µg/L.

Reported sublethal effects including changes in hormone levels, behavioral effects, kidney pathology, gill physiology, and potential olfaction effects have been observed at concentrations lower than 65 µg/L (see Appendix A and Section 4). In accordance with the Overview Document (U.S. EPA, 2004) and the Services Evaluation Memorandum (USFWS/NMFS, 2003), these studies were not considered appropriate for risk estimation in place of the life cycle studies because quantitative relationships between these effects and the ability of fish to survive, grow, and reproduce has not been established. The magnitude of the reported sublethal effect associated with reduced survival or reproduction has not been established; therefore it is not possible to quantitatively link sublethal effects to the selected assessment endpoints for this ESA. In addition, in the fish life cycle studies, no effects were observed to survival, reproduction, and/or growth at levels associated with the sublethal effects. Also, there were limitations to the studies that reported sublethal effects that preclude their quantitative use in risk assessment (see Appendix A and Section 4). Nonetheless, if future studies establish a quantitative link between the reported sublethal effects and fish survival, growth, or reproduction, the conclusions may need to be revisited.

6.2.5. Exposure to Pesticide Mixtures

In accordance with the Overview Document and the Services Evaluation Memorandum (U.S. EPA, 2004; USFWS/NMFS, 2004), this assessment considers the single active ingredient of atrazine, as well as available information on registered products containing multiple active ingredients in addition to atrazine. However, the assessed species and its environments may be exposed to multiple pesticides simultaneously. Interactions of other toxic agents with atrazine could result in additive effects, more than additive effects, or less than additive effects. The available data suggest that pesticide mixtures involving atrazine may produce either synergistic or additive effects. Mixtures that have been studied include atrazine with insecticides such as organophosphates and carbamates or with herbicides including alachlor and metolachlor. A number of study authors have reported additive, more than additive, or less than additive effects in several taxa including fish, amphibians, invertebrates, and plants.

As previously discussed, evaluation of pesticide mixtures is beyond the scope of this assessment because of the myriad of factors that cannot be quantified based on the

available data. Those factors include identification of other possible co-contaminants where the CRLF and the DS resides and their concentrations, differences in the pattern and duration of exposure among contaminants, and the differential effects of other physical/chemical characteristics of the receiving waters (e.g. organic matter present in sediment and suspended water). Evaluation of factors that could influence additivity/synergism/antagonism is beyond the nature and quality of the available data to allow for an evaluation. However, it is acknowledged that not considering mixtures could over- or under-estimate risks depending on the type of interaction and factors discussed above.

6.3. Uncertainty in the Potential Effect to Riparian Vegetation vs. Water Quality Impacts

Effects to riparian vegetation were evaluated using submitted guideline seedling emergence and vegetative vigor studies and non-guideline woody plant effects data. LOCs were exceeded for seedling emergence and vegetative vigor endpoints with the seedling emergence endpoint being considerably more sensitive. Based on LOC exceedances and the lack of readily available information to allow for characterization of riparian areas of the CRLF and the DS, it was concluded that atrazine use is likely to adversely affect these species by potentially impacting grassy/herbaceous riparian vegetation resulting in increased sedimentation. However, soil retention/sediment loading is dependent on a number of factors including land management and tillage practices. Use of herbicides (including atrazine) may be incorporated into a soil conservation plan. Therefore, although this assessment concludes that atrazine is likely to adversely affect the assessed listed species and its designated critical habitat by potentially impacting sensitive herbaceous riparian areas, it is possible that adverse impacts on sediment loading may not occur in areas where soil retention strategies are used.

6.4 Location of Wildlife Species

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

7. Risk Conclusions

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

Based on the best available information, the Agency makes a Likely to Adversely Affect determination for the CRLF and the DS from labeled uses of atrazine. Additionally, the Agency has determined that there is the potential for modification of the designated critical habitat for the CRLF and the DS from the use of the chemical. Given the LAA determination for the CRLF and the DS and potential modification of designated critical habitat for a description of the baseline status and cumulative effects for the CRLF is provided in **Attachment 2** and the baseline status and cumulative effects for the DS is provided in **Attachment 4**.

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

Table 7.1 Effects Determination Summary for Effects of Atrazine on the CRLF and the DS

Species	Effects Determination ¹	Basis for Determination
California red-legged frog (<i>Rana aurora draytonii</i>)	LAA ¹	Potential for Direct Effects
		<i>Aquatic-phase (Eggs, Larvae, and Adults):</i> No acute or chronic LOCs were exceeded for fish or aquatic phase amphibians.
		<i>Terrestrial-phase (Juveniles and Adults):</i> Acute and chronic LOCs were exceeded for birds. The available toxicity data suggest that amphibians are less sensitive than birds to atrazine, and considering factors such as lower food intake of terrestrial phase amphibians relative to birds reduces EECs and RQs, but does not reduce RQs to levels that are below LOCs.
		Potential for Indirect Effects
		<i>Aquatic prey items, aquatic habitat, cover and/or primary productivity</i> Atrazine could potentially impact terrestrial and aquatic plants to an extent that could result in indirect effects to the CRLF or modification of critical habitat.
Delta Smelt (<i>Hypomesus transpacificus</i>)	LAA ¹	<i>Terrestrial prey items, riparian habitat</i> CRLFs or critical habitat could be affected as a result of potential impacts to grassy/herbaceous vegetation. Food item abundance such as terrestrial invertebrates and terrestrial phase amphibians are not expected to be impacted to an extent that is expected to adversely affect the CRLF. However, potential impacts to herbivorous mammal abundance to an extent that could indirectly affect terrestrial phase CRLFs could not be precluded.
		Potential for Direct Effects No LOC exceedances occurred for acute or chronic effects to fish. Therefore, the effects determination for potential direct effects on the DS is “no effect”.
		Potential for Indirect Effects Labeled uses of atrazine have the potential to adversely affect the delta smelt and modify critical habitat either by reducing available food (marine copepods and aquatic plants), by impacting the riparian habitat of grassy and herbaceous riparian areas, or by impacting water quality via effects to aquatic vegetation.

¹ May affect, likely to adversely affect (LAA)

Table 7.2 Effects Determination Summary for Atrazine Use and CRLF and DS Critical Habitat Impact Analysis

Assessment Endpoint	Effects Determination	Basis for Determination
Modification of aquatic-phase PCEs (DS and CRLF)	Habitat Modification	As described in Table 7.1., the effects determination for the potential for atrazine to affect aquatic phase CRLFs and the DS is LAA. This determination is based on the potential for atrazine to affect the DS and CRLF’s food and habitat. Potential effects to aquatic invertebrates, aquatic plants, and terrestrial (riparian) plants identified in this assessment could result in aquatic habitat modification.
Modification of terrestrial-phase PCE (CRLF)		As described in Table 7.1., the effects determination for the potential for atrazine to affect terrestrial phase CRLFs is LAA. This determination is based on the potential for atrazine to directly affect terrestrial phase CRLFs and indirectly affect CRLFs by potentially impacting food supply and vegetative habitat. These potential effects could result in modification of critical habitat.

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

When evaluating the significance of this risk assessment's direct/indirect and adverse habitat modification effects determinations, it is important to note that pesticide exposures and predicted risks to the listed 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.

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 and the DS life stages within the action area and/or applicable designated critical habitat. 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 assessed species.
- Quantitative information on prey base requirements for the assessed species. While existing information provides a preliminary picture of the types of food sources utilized by the assessed species, 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 species and potential modification to critical habitat.

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