Risks of Trifluralin Use to the Federally Listed California Red-legged Frog (*Rana aurora draytonii*), Delta Smelt (*Hypomesus transpacificus*), San Francisco Garter Snake (*Thamnophis sirtalis tetrataenia*), and San Joaquin Kit Fox (*Vulpes macrotis mutica*)

Pesticide Effects Determination

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

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Primary Authors:
Christine Hartless, Ph.D., Wildlife Biologist
Marie Janson, M.S., Environmental Scientist
Robert Miller, M.E.S.M., Environmental Protection Specialist

Secondary Review:
Faruque Khan, Ph.D., Senior Scientist
Brian Anderson, Ph.D., RAPL

Branch Chief, Environmental Risk Assessment Branch 1
Nancy Andrews, Ph.D.

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1. Executive Summary

The purpose of this assessment is to evaluate potential direct and indirect effects on the following species arising from FIFRA regulatory actions regarding use of trifluralin:

California red-legged frog (CRLF), *Rana aurora draytonii*
Delta smelt (DS), *Hypomesus transpacificus*,
San Francisco Garter Snake (SFGS), *Thamnophis sirtalis tetrataenia*,
San Joaquin Kit Fox (SJKF), *Vulpes macrotis mutica*,

In addition, this assessment evaluates whether these actions can be expected to result in modification of the species' designated critical habitat. This assessment was completed in accordance with the U.S. Fish and Wildlife Service (USFWS) and National Marine Fisheries Service (NMFS) *Endangered Species Consultation Handbook* (USFWS/NMFS 1998 and procedures outlined in the Agency's Overview Document (USEPA 2004).

The listing date and a general description of the range of each assessed species are as follows.

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

The DS was listed as threatened on March 5, 1993 (58 FR 12854) by the U.S. Fish and Wildlife Service (USFWS) (USFWS, 2007a). It is only found in Suisun Bay and the Sacramento-San Joaquin estuary near San Francisco Bay. Critical habitat has been designated for the DS.

The SFGS was listed as an endangered species by the USFWS in 1967 and was grandfathered under the Endangered Species Act (ESA) when it was signed into law in 1973. The SFGS is endemic to the San Francisco Peninsula and San Mateo County and historically inhabited densely vegetated ponds or shallow marshlands near open hillsides found from San Francisco to Santa Cruz, including the San Francisco Peninsula. The current distribution of the SFGS is unknown because most of their historic range is now privately owned; however, it appears that the SFGS can still be found in much of its historic range. Critical habitat has not been designated for the SFGS.

The SJKF was listed as endangered by the USFWS on March 11, 1967. Its current range includes Alameda, Contra Costa, Fresno, Kern, Kings, Madera, Merced, Monterey, San Benito, San Joaquin, San Luis Obispo, Santa Barbara, Santa Clara, Stanislaus, Tulare and Ventura counties in California. The SJKF inhabits a variety of habitats, including grasslands, scrublands, vernal pool areas, oak woodland, alkali meadows and playas, and an agricultural matrix of row crops, irrigated pastures, orchards, vineyards, and grazed annual grasslands. Critical habitat has not been designated for the SJKF.

Trifluralin is a pre-emergent herbicide used to control annual grasses and broadleaf weeds on a variety of food crops and non-food uses. The herbicide is formulated as an emulsifiable concentrate and granular products. Trifluralin is typically applied dormant, semi-dormant, preplant, pre-transplant, post-plant, pre-emergence, post-emergence, lay-by, or postharvest. Trifluralin can be applied by aerial spray, ground spray or by granular spreaders. Some labels require soil incorporation, while others do not require soil incorporation.

Trifluralin is a dinitroaniline herbicide that enters plants through developing roots and stops plant cells from dividing and elongating (meristematic inhibitor).Trifluralin is readily absorbed by young roots. Established weeds are not controlled.

Trifluralin uses that are considered in this federal action relevant to the CRLF, DS, SFGS, and SJKF are all registered uses in California. These uses include many agricultural crops, non-food commodities, residential, and other non-agricultural uses.

The major dissipation routes of trifluralin are volatilization and photodegradation and to a lesser extent by biotic degradation. Trifluralin is most effective when it is incorporated into the soil at the time of application, which reduces the rate of volatilization and photodegradation on soil. Trifluralin has a very low propensity to leach in the vast majority of soils because of its strong adsorption to soil colloids and organic matter. Trifluralin is persistent and immobile in aerobic soil studies and in adsorption/desorption studies. Trifluralin residues in the atmosphere of remote, non-use regions have been reported, indicating its potential for long range transport.

Data indicate that trifluralin has potential to bioaccumulate in aquatic ecosystems. Trifluralin has an octanol-water partitioning coefficient (log K_{ow}) of 5.27. In a bioconcentration study, trifluralin residues in bluegill sunfish resulted in bioconcentration factors of 2041, 9586, and 5674 for edible, nonedible, and whole fish tissues, respectively. The half-life of elimination (depuration) was estimated to be 14 days. The accumulation and depuration rates of trifluralin in fish cannot be fully assessed because radioactive residues in fish tissues were incompletely characterized. In addition, other chemical properties of trifluralin, such as its volatility and short aqueous photolysis half-life, may mediate the realization of bioaccumulation in the environment.

Trifluralin is described by the US EPA (1999) as bioaccumulative, persistent, and toxic; and therefore presents itself as a contaminant of concern. Based on available bioaccumulation data, trifluralin has the potential to accumulate in aquatic organisms. KABAM (K_{OW} (based) Aquatic BioAccumulation Model) v.1.0 is used to estimate potential bioaccumulation of trifluralin residues in a freshwater aquatic food web and subsequent risks these residues pose to aquatic-phase CRLF and SFGS via consumption of contaminated aquatic prey (i.e., aquatic invertebrates and fish).

The effects determinations for each of the listed species assessed is based on a weight-of-evidence method that relies heavily on an evaluation of risks to each taxon 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). Since some of the the assessed species exist within both aquatic and terrestrial habitats, exposure of the listed species, their prey and their habitats to trifluralin are assessed separately for the two habitats. Tier-II aquatic exposure models are used to estimate high-end exposures of trifluralin in aquatic habitats resulting from runoff and spray drift from different uses. Peak model-estimated environmental concentrations resulting from different trifluralin uses range from 0.0002 to 6.55 µg/L. These estimates are supplemented with an analysis of California surface water monitoring data from the California Department of Pesticide Regulation. The maximum concentration of trifluralin reported by the California Department of Pesticide Regulation surface water database (1.74 µg/L) is roughly four times lower than the highest peak model-estimated environmental concentration. No trifluralin monitoring data are available from the U. S. Geological Survey's National Water Quality Assessment (NAWQA).

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

Based on the best available information, the Agency makes a Likely to Adversely Affect (LAA) determination for the CRLF, DS, SJKF and SFGS from the use of trifluralin. 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. A summary of the risk conclusions and effects determinations for each listed species assessed here 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, DS, SFGS, and SJKF and potential modification of designated critical habitat for the CRLF and DS, 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 Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination** [1]	**Basis for Determination**
		Potential for Direct Effects
CRLF	LAA	***Aquatic-phase (Eggs, Larvae, and Adults)***: *Fish, Adult survival:* Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. The chance of individual effects (*i.e*., mortality) for freshwater fish (surrogate for aquatic-phase CRLFs) is as high as ~1 in 3240. Buffer distance required to remove all LOC exceedances under the evaluated labels ranged from 45.9 to 2359 feet. Available aquatic-phase amphibian data suggests trifluralin is less acutely toxic to aquatic-phase amphibians than to fish; however, there are Listed Species LOC exceedances for the most sensitive aquatic-phase amphibian data. *Fish, Growth and reproduction*: There were no chronic LOC exceedances. However, fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS. *Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the maximum observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09).
		Terrestrial-phase (Juveniles and Adults): *Adult survival:* There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater than expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e*., short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories.
		Potential for Indirect Effects
		Aquatic prey items, aquatic habitat, cover and/or primary productivity *Fish, Adult survival:* Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out

Table 1.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination [1]**	**Basis for Determination**
		of 25 crop scenarios with at least one of the application methods. The chance of individual effects (*i.e.*, mortality) for freshwater fish (surrogate for aquatic-phase CRLFs) is as high as ~1 in 3240. Buffer distance required to remove all LOC exceedances under the evaluated labels ranged from 45.9 to 2359 feet. Available aquatic-phase amphibian data suggests trifluralin is less acutely toxic to aquatic-phase amphibians than to fish; however, there are Listed Species LOC exceedances for the most sensitive aquatic-phase amphibian data. *Fish, Growth and reproduction*: There were no chronic LOC exceedances. However, fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS. *Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the maximum observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09). *Freshwater invertebrates:* There were no exceedances of the Listed Species Acute or Chronic LOCs. *Vascular and non-vascular plants:* There were no exceedances of the Acute LOCs. *Terrestrial plants*: LOCs were exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs were exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
		Terrestrial prey items, riparian habitat *Birds, Adult survival:* There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater then expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Birds, Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the

Table 1.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination [1]**	**Basis for Determination**
		nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e.*, short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories. *Mammals: adult survival:* There were no mortalities or sublethal effects in the acute dose. Acute risks to mammals from spray or granular applications of trifluralin are unlikely as the calculated exposures do not exceed the toxicity values. *Mammals, growth and reproduction*: Chronic dose-based RQs for 15 g mammals range from 20.82 to 58.20 for short grass; 9.54 to 26.68 for tall grass; 11.71 to 32.74 broadleaf plants/small insects; 1.30 to 3.64 for fruits/pods/seeds/large insects. Chronic RQs did not exceed LOCs for 15g granivores. Chronic dietary-based RQs exceed the Agency's chronic LOC (1.0) in both crop scenarios (liquid application) for mammals consuming short grass, tall grass, and broadleaf plants/small insects; they were not exceeded for fruits/pods/seeds/large insects. *Terrestrial invertebrates*: For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortality at this dose). Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios. *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
DS	LAA	**Potential for Direct Effects**
		Fish, Adult survival: Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. The chance of individual effects (*i.e.*, mortality) for freshwater fish (surrogate for aquatic-phase CRLFs) is as high as ~1 in 3240. Buffer distance required to remove all LOC exceedances under the evaluated labels ranged from 45.9 to 2359 feet. *Fish, Growth and reproduction*: There were no chronic LOC exceedances. However, fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the

Table 1.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination [1]**	**Basis for Determination**
		monitoring data and many of the chronic EECs estimated using PRZM/EXAMS. *Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09).
		Potential for Indirect Effects
		Freshwater invertebrates: There were no exceedances of the Listed Species Acute or Chronic LOCs. *Vascular and non-vascular plants:* There were no exceedances of the Acute LOCs. *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
SFGS	LAA	**Potential for Direct Effects**
		Adult survival: There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater then expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e.*, short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories.
		Potential for Indirect Effects
		Fish, Adult survival: Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. The chance of individual effects (*i.e.*, mortality) for

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Species	**Effects Determination [1]**	**Basis for Determination**
		freshwater fish (surrogate for aquatic-phase CRLFs) is as high as ~1 in 3240. Buffer distance required to remove all LOC exceedances under the evaluated labels ranged from 45.9 to 2359 feet. *Fish, Growth and reproduction*: There were no chronic LOC exceedances. However, fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS. *Freshwater invertebrates:* There were no exceedances of the Listed Species Acute or Chronic LOCs. *Vascular and non-vascular plants:* There were no exceedances of the Acute LOCs. *Birds, Adult survival:* There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater then expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Birds, Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e.*, short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories. *Mammals: adult survival:* There were no mortalities or sublethal effects in the acute dose. Acute risks to mammals from spray or granular applications of trifluralin are unlikely as the calculated exposures do not exceed the toxicity values. *Mammals, growth and reproduction*: Chronic dose-based RQs for 15 g mammals range from 20.82 to 58.20 for short grass; 9.54 to 26.68 for tall grass; 11.71 to 32.74 broadleaf plants/small insects; 1.30 to 3.64 for fruits/pods/seeds/large insects. Chronic RQs did not exceed LOCs for 15g granivores. Chronic dietary-based RQs exceed the Agency's chronic LOC (1.0) in both crop scenarios (liquid application) for mammals consuming short grass, tall grass, and broadleaf plants/small insects; they were not exceeded for fruits/pods/seeds/large insects. *Terrestrial invertebrates*: For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortality at this dose). Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios. *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to

Table 1.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination [1]**	**Basis for Determination**
		monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
SJKF	LAA	**Potential for Direct Effects**
		Mammals: adult survival: There were no mortalities or sublethal effects in the acute dose. Acute risks to mammals from spray or granular applications of trifluralin are unlikely as the calculated exposures do not exceed the toxicity values. *Mammals, growth and reproduction*: Chronic dose-based RQ values representing trifluralin exposures (spray applications) to large mammals (1000 g) indicate risks resulting from both the alfalfa and nursery application scenarios (for alfalfa, LOC exceedances for mammals consuming short grass, tall grass, and broadleaf plants/small insects; for nursery, LOC exceedences for mammals consuming short grass, tall grass, broadleaf plants/small insects, and fruits/pods/seeds/large insects. Chronic diet-based RQ values representing trifluralin exposures to all mammals indicate risks to mammals consuming short grass, tall grass, and broadleaf plants/small insects for both the alfalfa and nursery scenarios. Chronic risk to mammals from trifluralin through consumption of contaminated earthworms is unlikely.
		Potential for Indirect Effects
		Birds, Adult survival: There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater then expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Birds, Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e*., short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories. *Mammals: adult survival:* There were no mortalities or sublethal effects in the acute dose. Acute risks to mammals from spray or granular applications of trifluralin are unlikely as the calculated exposures do not exceed the toxicity values. *Mammals, growth and reproduction*: Chronic dose-based RQs for all weight classes of mammals exceeded the Chronic LOC

Table 1.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination [1]**	**Basis for Determination**
		(1.0) for all weights and feeding guilds except for large mammals consuming fruits/pods/seeds/large insects (alfalfa application) and all granivores. Exceeding RQs ranged from 1.11 to 58.20. Chronic dietary-based RQs exceed the Agency's Chronic LOC (1.0) in for alfalfa and nursery scenarios (liquid application, non-incorporated) for short grass, tall grass, and broadleaf plants/small insects. *Terrestrial invertebrates*: For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortality at this dose). Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios. *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
[1] No effect (NE); May affect, but not likely to adversely affect (NLAA); May affect, likely to adversely affect (LAA)		

Table 1.2 Effects Determination Summary for the Critical Habitat Impact Analysis (CRLF and DS)[1]		
Designated Critical Habitat for:	**Effects Determination [2]**	**Basis for Determination**
CRLF	HM	***Aquatic-phase PCEs:*** *Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09). *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses. There is a potential for direct effects to aquatic-phase CRLF and indirect effects via reduction of aquatic-phase prey items (fish and aquatic-phase amphibians) as described in **Section 5**. ***Terrestrial-phase PCEs:*** *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85 % were incidents were classified as registered uses. There is a potential for direct effects to terrestrial–phase CRLF and indirect effects via reduction of terrestrial-phased prey items (mammals, terrestrial invertebrates, and frogs) as described in **Section 5**.
DS	HM	*Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09). *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one

		of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses. There is a potential for direct effects to the DS as described in **Section 5**.
[1] Critical habitat has not been designated for the SFGS or the SJKF. [2] Habitat Modification (HM) or No effect (NE)		

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

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

- Enhanced information on the density and distribution of CRLF, DS, SFGS, and SJKF 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 listed threatened California red-legged frog *(Rana aurora draytonii*) (CRLF), and delta smelt (*Hypomesus transpacificus*) (DS), and the federally listed endangered San Francisco garter snake (*Thamnophis sirtalis tetrataenia*) (SFGS), and San Joaquin kit fox (*Vulpes macrotis*) (SJKF) from FIFRA regulatory actions regarding use of trifluralin on cultivated fields for agricultural crops (*e.g*. stone fruits, cucumbers, collards, melons, tree nuts) and non-food commodities (*e.g.*, forest trees, nurseries, turf), residential uses, and other non-agricultural uses (*e.g*., industrial areas, rights-of-ways). In addition, this assessment evaluates whether use on these use sites is expected to result in modification of designated critical habitat for the CRLF and DS. No critical habitat has been designated for the SFGS and SJKF. 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, SJKF, and SFGS for which trifluralin was alleged to be of concern in a separate suit (*Center for Biological Diversity (CBD) vs. EPA et al*. (Case No. 07-2794JCS)).

In this assessment, direct and indirect effects to the CRLF, DS, SFGS, and SJKF potential and modification to designated critical habitat for the CRLF and DS 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 taxon 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, AgDRIFT, and AGDISP, all of which are described at length in the Overview Document (U.S. EPA, 2004). Additional refinements include an analysis of the usage data, use of the T-HERPS model (characterizes potential risks to terrestrial phase amphibians and reptiles from dietary exposure), use of the KABAM model (an aquatic bioaccumulation model), and use of an earthworm fugacity model (predicts concentrations of trifluralin in soil as well as terrestrial invertebrates as food items for the terrestrial-phase CRLF, SFGS, and SJKF). 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 Endangered Species Act (ESA), and the Services' *Endangered Species Consultation Handbook*, the assessment of effects associated with registrations of trifluralin is based on an action area. The action area is the area directly or indirectly affected by the federal action, as indicated by the exceedence of the Agency's Levels of Concern (LOCs). It is acknowledged that the action area for a national-level FIFRA regulatory decision associated with a use of trifluralin 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, DS, SFGS, and SJKF 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 trifluralin in accordance with current labels:

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

Critical habitat has been designated for the CRLF and DS, but not for the SJKF and SFGS. 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 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 trifluralin 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 trifluralin.

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

2.2.1 Chemicals Assessed

This assessment evaluates potential risks from exposure to trifluralin. Trifluralin is a pre-emergent dinitroaniline herbicide used to control annual grasses and broadleaf weeds on a variety of food crops and non-food uses (see **Table 2.3** for a list of labeled uses). The herbicide is formulated as an emulsifiable concentrate and granular products. Trifluralin is typically applied dormant, semi-dormant, pre-plant, pre-transplant, post-plant, pre-emergence, post-emergence, lay-by, or postharvest. Trifluralin can be applied by aerial spray, ground spray or by granular spreaders. Some labels require soil incorporation, while others do not require soil incorporation.

As discussed in **Section 2.4.1**, there are three primary degradates for trifluralin. There is insufficient data on the major degradates of trifluralin to adequately assess their persistence and mobility. Available toxicity data for these degradates suggests that degradates are no more toxic than the parent with respect to aquatic organisms (**Section 4**). No degrade toxicity data were available for terrestrial organisms.

2.2.2 Scope of the Action

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

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

The Agency does not routinely include, in its risk assessments, an evaluation of mixtures of active ingredients, either those mixtures of multiple active ingredients in product formulations or those in the applicator's tank. In the case of the product formulations of active ingredients (that is, a registered product containing more than one active ingredient), each active ingredient is subject to an individual risk assessment for regulatory decision regarding the active ingredient on a particular use site. If effects data are available for a formulated product containing more than one active ingredient, they may be used qualitatively or quantitatively in accordance with the Agency's Overview Document and the Services' Evaluation Memorandum (U.S., EPA 2004; USFWS/NMFS 2004). This analysis can be conducted using acute rat toxicity data submitted to the Agency (**Appendix A**). In this mixture evaluation an LD_{50} with associated 95% CI is needed for the formulated product. The same quality of data is also required for each component of the mixture. In the case of trifluralin, given that there is no 95% CI associated with the oral LD_{50} (>5000 mg/kg), it is not possible to undertake a quantitative or qualitative analysis for potential interactive effects. However, because the active ingredients are not expected to have similar mechanisms of action, metabolites, or toxicokinetic behavior, it is reasonable to conclude that an assumption of dose-addition would be inappropriate. Consequently, an assessment based on the toxicity of trifluralin is the only reasonable approach that employs the available data to address the potential acute risks of the formulated products.

Several papers were cited in ECOTOX that were classified as evaluating mixtures that contained trifluralin. However, none provided sufficient information for inclusion into this assessment.

2.3 Previous Assessments

Trifluralin has a long regulatory history and has been the subject of numerous assessments. It was first registered in the United States in 1963 for use as a selective preemergent herbicide. In April, 1996, the USEPA completed its Reregistration Eligibility Decision on the active ingredient trifluralin (USEPA 1996, Reregistration Eligibility Decision (RED) Trifluralin). The Agency determined all labeled uses of trifluralin at the time of the assessment were eligible for reregistration with the exception of nongrass forage/fodder/straw/hay and dill. Residue data were not generated to support these uses and these uses were to be deleted from all labels. Relevant ecological conclusions from the RED include the following:

- For aquatic animals (fish and invertebrates), trifluralin was ranked as having moderate to high toxicity according to the hazard classification scheme. Due to trifluralin's toxicity to fish, aquatic invertebrates and estuarine/marine organisms, the Agency required aquatic impact labeling on all trifluralin end-use products. In addition, laboratory and field studies suggest exposure-related abnormalities in vertebral development, at concentrations below those where acute effects are anticipated. Also, the LOC determination is based on trifluralin dissolved in the water column and does not take into account trifluralin adsorbed to sediment. Trifluralin adsorbed to sediment may pose a risk for fish species that forage by feeding from sediment, particularly since trifluralin has a moderate tendency to bioaccumulate. The Agency will explore further monitoring efforts or additional analyses with the registrants of technical trifluralin in order to obtain more refined characterization of the risk to fish.

- Trifluralin ranks as practically non-toxic to birds and mammals on an acute basis. Two of four laboratory bird studies indicate chronic risk, as evidenced by egg shell cracking. Chronic risks to mammals were not evaluated.
- For terrestrial and semi-aquatic plants the Agency did find a concern for the semiaquatic category. The Agency did not find concerns for aquatic plants resulting from use of trifluralin.
- For control of effects caused by spray drift, the RED indicated that the Agency will require precautionary labeling, which is standard for pesticides with aerial applications.

The Agency also completed an Effects Determination for 26 threatened and endangered Pacific anadromous salmon and steelhead in April 2004 based on trifluralin uses in a variety of field, fruit, and vegetable crops, ornamentals, and non-crop sites in the Pacific Northwest consistent with a court order in WTC v. EPA (Case No. 1:04-Cv-00126-Ckk, 2004). The results of that endangered species risk assessment showed that that the use of trifluralin may affect and was likely to adversely affect 11 Evolutionary Significant Units (ESUs), may affect but is not likely to adversely affect 4 ESUs, and has no effect on 11 ESUs of Pacific salmon and steelhead when used according to labeled application directions (http://www.epa.gov/oppfead1/endanger/litstatus/effects/#trifluralin). The National Marine Fisheries Service has indicated it will review EPA's determination regarding effects of trifluralin to the Pacific salmon and steelhead, and complete consultation with issuance of a Biological Opinion in February 2012.

2.4 Stressor Source and Distribution

2.4.1 Environmental Fate Properties

The major dissipation routes of trifluralin are volatilization and photodegradation and to a lesser extent by biotic degradation. The physicochemical properties (Table 2.1a) suggest that trifluralin is hardly soluble in water. The vapor pressure (1.10E-4 Torr) indicates that trifluralin is a volatile chemical and Henry's law constant (1.6×10^{-4} atm-m3/mol) indicates that trifluralin has potential to volatilize from moist soil and water surfaces. Trifluralin is stable to hydrolysis at various pHs, but is very prone to phototransformation in air and water ($T_{1/2}$ of < 1 day) and to a lesser extent in soil ($T_{1/2}$ of 41 days; **Table 2.1b**). Soil incorporation of trifluralin can also inhibit the rate of photodegradation in soil. Laboratory aerobic biotransformation studies indicate that trifluralin is moderately persistent to persistent in soil. Trifluralin degraded with half-lives of 189, 202, and 116 days in sandy loam, clay loam, and loam soils, respectively. Anaerobic soil biotransformation of trifluralin is faster than aerobic biotransformation with half-lives of 22-59 days. Field dissipation half-life has been reported as low as 29 days and as high as 149 days for trifluralin. Published field dissipation half-lives range from 60 to 132 days (Wauchope et al., 1992). Several major and minor degradates were detected in fate studies.

Trifluralin has a very low propensity to leach in a majority of soils because of its strong adsorption to soil colloids and organic matter. Adsorption/desorption and leaching studies indicate that trifluralin is strongly adsorbed to most soils and classified as immobile according to FAO Classification System (FAO, 2000). The log K_{ow} (5.27) indicates trifluralin has the potential to bioaccumulate. Trifluralin is described by the US EPA as bioaccumulative, persistent, and toxic; and therefore presents itself as a contaminant of concern (US EPA Toxic Release Inventory, 2007).

Tables 2.1a and **2.1.b** list the environmental fate properties of trifluralin detected in submitted environmental fate and transport studies. The detailed descriptions of guideline fate studies are located in **Appendix B**.

Table 2.1a Summary of Trifluralin Physical and Chemical Properties

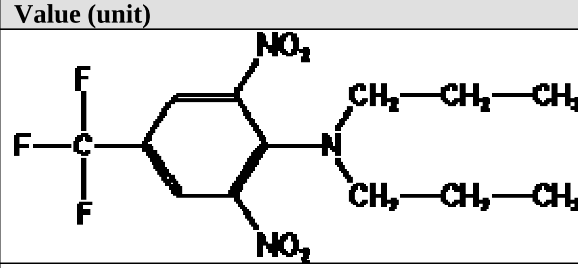
Physical/Chemical Property	**Value (unit)**
Chemical Structure	
IUAPC	2,6-Dinitro-*N*,*N*-dipropyl-4-(trifluoromethyl)aniline
CAS	1582-09-9
Molecular Formula	$C_{13}H_{16}F_3N_3O_4$
Molecular Weight	335.28 g/mole
Physical State	orange crystalline solid
Melting Point	42-49^{o} C
Vapor Pressure (25^{o}C)	1.10E-4 Torr
Water Solubility (25^{o}C)	0.3 mg/L
Log K_{ow} at 20^{o}C	5.27
Henry's Law Constant at 25^{o}C	1.6×10^{-4} atm-m3/mol

Table 2.1b Summary of Environmental Fate Properties of Trifluralin				
Study	**Value (units)**	**Major Degradates[1]** ***Minor Degradates***	**MRID #**	**Study Status**
Hydrolysis	Stable at pH 5, pH 7, pH 9	None	00131135	Supplemental
Aqueous Photolysis	$T_{1/2}$ = 0.371 days	TR-15, ≤47.4% TR-6, ≤29.8% *TR-2, ≤9.6%*	40560101	Supplemental
Photodegra-dation on Soil	$T_{1/2}$ = 41 days	*TR-2 ≤ 6.0%* *TR-12, ≤ 7.1%*	40597801 40751301	Supplemental
Aerobic Soil Metabolism	189 days (sandy loam) 201 days (clay loam) 116 days (loam soils) 22°C	*TR-2, ≤ 4.6%* *TR-5, ≤ 2.1%* *TR-11, ≤ 0.3%* *TR-13, ≤ 1.0%* *TR15, ≤ 2.6%* *TR-20, ≤ 2.7%* *TR-28, ≤ 3.0%*	41240501	Supplemental
Anaerobic Soil Metabolism	59 days (sandy loam) 25 days loam 35 clay loam soils 22° C	TR-4, ≤ 13.2% *TR-14, ≤ 8.3%* *TR-7, ≤ 4.1%* *Other degradates identified at ≤ 2.1% :* *TR-2* *TR-5* *TR-13* *TR-28*	41240502	Supplemental
Anaerobic Aquatic Metabolism	N/A	N/A	N/A	N/A
Aerobic Aquatic Metabolism	N/A	N/A	N/A	N/A
Leaching & Adsorption/ Desorption	sand: K_d = 18.6, L/kg (K_{oc}=6,413 g/mL) sandy loam: K_d = 54.8, L/kg (K_{oc}=6,748 g/mL) loam: K_d = 88.3, L/kg (K_{oc}= 8,457 g/mL) clay loam: K_d = 155.6, L/kg (K_{oc}=13,413 g/mL) Average Koc = 8,757.9 g/mL		40673501	Supplemental
Terrestrial Field Dissipation	Granular $T_{1/2}$ = 49 days; EC formulations $T_{1/2}$ = 149 days from California loam soil; $T_{1/2}$ = 93 days from Alabama clay soil; $T_{1/2}$ = 29 days, coarse sandy loam soil, Shellman, GA; $T_{1/2}$ = 35 days when applied to fine (silty clay loam soil, Mansfield, IL); trifluralin did not appear in depths greater than 6 inches		41781901 41661101 42309101	Supplemental Supplemental Supplemental
Bioconcentrat ion	Trifluralin residues accumulated in a bluegill sunfish (*Lepomis macrochirus*) exposed to 0.0059 mg/L of trifluralin with		40673801	Supplemental

Table 2.1b Summary of Environmental Fate Properties of Trifluralin				
Study	**Value (units)**	**Major Degradates[1]** *Minor Degradates*	**MRID #**	**Study Status**
	maximum mean bioconcentration factor of 5,674x for whole fish tissues. The maximum mean concentration of total [^{14}C] residues occurred at 28 days for the whole fish sample was 67.0 mg/L. Depuration occurred with 86.34-88.01% of the [^{14}C] eliminated from the fish tissues after 14 days of exposure to pesticide free water. The time to reach 90% of steady state for whole fish was 15.8 days.			

[1] *TR-2 α,α,α-trifluoro-2,6-dinitro-N-propyl-p-toluidine*
TR-4 α,α,α -trifluoro-5-nitro-N4,N4-dipropyl-toluene- 3,4-diamine
TR-5 α,α,α-trifluoro-5-nitro-4-propyl-toluene- 3,4-diamine
TR-6 5-trifluoromethyl-3-nitro-1,2-benzenediamine
TR-7 α,α,α-trifluoro-N4,N4-dipropyltoluene-3,4,5-triamine
TR-11 2-ethyl-7-nitro-1-propyl-5-(trifluoromethyl)benzimidazole-3-oxide
TR-12 2-ethyl-7nitro-5- trifluoromethylbenzimidazole- 3-oxide
TR-13 2-ethyl-7-nitro-1-propyl-5-(trifluoromethyl) benzimidazole
TR-14 7-amino-2-ethyl-1-propyl-5-(trifluoromethyl) benzimidazole
TR-15 2-ethyl-7-nitro-5-(trifluoromethyl)benzimidazole
TR-20 α,α,α-trifluoro-2,6-dinitro-p-cresol
TR-28 2,2'azoxybis(, , -trifluoro-6-nitro-N-propyl-p- toluidine

2.4.1.1 Degradation

Trifluralin is hydrolytically stable under most environmental conditions. The vapor-phase of trifluralin is degraded in the atmosphere by reaction with photochemically-produced hydroxyl radicals, and the estimated half-life for this reaction in air is estimated to be 5.3 hours or 0.22 days (Convention for the Protection of the Marine Environment of the North-East Atlantic Convention (OSPAR, 2005). Trifluralin is also susceptible to direct aqueous photolysis (half-life 8.9 hours) which should limit its persistence in the top segment of the water column. The photodegradation on soil study reports that trifluralin degraded with a half-life of 41 days. **Appendix B** provides nomenclature and chemical structures of trifluralin degradates.

Trifluralin is persistent in aerobic soil with half-lives of 189, 201, and 116 days in sandy loam, clay loam, and loam soils, respectively. No major metabolites were formed. In anaerobic soil metabolism studies trifluralin's half-lives ranged from 25-59 days in sandy loam, loam, and clay loam soils, respectively. One major metabolite formed in the anaerobic soil studies, TR-4 (α,α,α -trifluoro-5-nitro-N4,N4-dipropyl-toluene-3,4-diamine), up to13.2%. Aerobic and anaerobic aquatic guideline studies have not been submitted.

The major degradates reported in the aqueous photolysis study include: TR-6, 29.8% (5-trifluoromethyl-3-nitro-1,2-benzenediamine) and TR-15, 47.4% (2-ethyl-7-nitro-5-trifluromethylbenzimidazole).

There is insufficient data on the major degradates of trifluralin to adequately assess their persistence and mobility.

2.4.1.2 Mobility

Volatility may be a major route of dissipation for trifluralin if not incorporated. Trifluralin volatilizes when applied to the surface of soil with an amount of 41-68% of the applied radioactivity after 24 hours (OSPAR, 2005). However, volatilization is minimal (< 2% application rate) when trifluralin is incorporated into the soil immediately after application. Trifluralin rapidly dissipates from surface water due to volatilization, photo-oxidation and adsorption to suspended matter in water column and sediments.

The relatively high soil/water partitioning of trifluralin (Koc of 8000; Kads = 18-19 for sandy soil and 53-156 for finer soils) indicates that the concentration of trifluralin adsorbed to eroding soil will be 1 to over 2 orders of magnitude greater than the dissolved concentration in runoff water. However, the sediment yield off many fields varies anywhere from 1 to > 3 orders of magnitude less than the mass equivalent of the runoff volume. Therefore, the mass percentage of trifluralin runoff occurring via dissolution in runoff water may be somewhat comparable to or sometimes greater than that occurring via adsorption to eroding soil in cases where the sediment yield is 1 to > 3 orders of magnitude less than the runoff volume.

Trifluralin's relatively high soil/water partitioning indicates that the concentration of trifluralin adsorbed to suspended and bottom sediment will be substantially greater than its dissolved concentration in the water column.

Even though phototransformation of trifluralin in air and water is rapid, residues have been detected in air, precipitation (rain and snow) and fog in remote areas such as the Canadian Arctic, Greenland and the Bering Sea (Canadian Arctic Contaminants Program, 2006). These detections in remote regions indicate that trifluralin sorbed to airborne particulate is more resistant to phototransformation; thus, transport of particulates might be the primary transport mode for deposition in remote areas.

In field dissipation studies, trifluralin (Treflan, 44.1 or 50.7% EC) applied at 2.78 lbs/acre dissipated with a half-life of 149 days from loam soil planted with cotton in California and 93 days from clay soil planted with soybeans in Alabama. Trifluralin did not appear to leach below the 0- to 6-inch soil depth. Trifluralin granular formulation dissipated with a reported half-life of 49 days in the top six inches of soil when applied to loamy sand soil in California. Emulsifiable concentrate trifluralin formulations were reported to dissipate with a half-lives ranging from 29 to 35 days when applied to coarse sandy loam soil (Shellman, GA site) and fine silty clay loam soil (Mansfield IL site).

2.4.1.3 Accumulation

Trifluralin residues accumulated in bluegill sunfish exposed to 0.0059 ppm of trifluralin for 28 days under flow through conditions. The maximum mean bioconcentration factors were 2041x, 9586x, and 5674x for edible, nonedible, and whole fish tissues, respectively. Maximum mean concentrations of total residues occurred at 28 days for edible, non-edible, and whole fish samples and were 12.9 ppm, 67.0 ppm and 39.6 ppm, respectively. Depuration occurred with 86.34-88.01% of the [^{14}C]residues eliminated from the fish tissues after 14 days of exposure to pesticide free water. The accumulation and depuration of trifluralin in fish cannot be fully assessed because radioactive residues in fish tissues were incompletely characterized. Radioactivity attributed to a total of 10 metabolites at a maximum of 0.80 ppm was not identified; up to 1.27 ppm was described as only “polar radioactivity.”

Based on the available results, trifluralin poses a high potential for bioaccumulation and may biomagnify at higher trophic levels. For this reason, this assessment considers additional exposure pathways resulting from bioaccumulation for the CRLF and SFGS.

2.4.2 Environmental Transport Mechanisms

Potential transport mechanisms or routes of pesticide exposure for trifluralin include surface water runoff/erosion, spray drift, and secondary drift of volatilized or soil-bound residues leading to deposition onto nearby or more distant ecosystems.

A number of studies have documented atmospheric transport and re-deposition of pesticides from the Central Valley to the Sierra Nevada Mountains (Fellers et al., 2004, Sparling *et al*., 2001, LeNoir *et al*., 1999, and McConnell *et al*., 1998). Several sections of the range and critical habitat for the CLRF and habitat of the SJKF are located east of the Central Valley. The magnitude of transport via secondary drift depends on the ability of trifluralin to be mobilized into air and its eventual removal through wet and dry deposition of gases/particles and photochemical reactions in the atmosphere. Therefore, physicochemical properties of trifluralin that describe its potential to enter the air from water or soil (*e.g.*, Henry’s Law constant and vapor pressure), pesticide use data, modeled estimated concentrations in water and air, and available air monitoring data from the Central Valley and the Sierra Nevada are qualitatively considered in evaluating the potential for atmospheric transport of trifluralin to locations where it could impact the CRLF, DS, SFGS and SJKF.

In general, deposition of spray drift pesticides is expected to be greatest close to the site of application. Computer models of spray drift (AgDRIFT and/or AGDISP) are used to determine potential exposures to aquatic and terrestrial organisms via spray drift.

2.4.3 Mechanism of Herbicidal Action

Trifluralin is a synthetic fluorinated dinitroaniline herbicide that enters plants through developing roots preventing the alignment and separation of chromosomes during mitosis (mitosis disruptor). Trifluralin binds to the major microtubule protein tubulin leading to microtube loss and the absence of the spindle apparatus preventing alignment and separation of chromosomes during mitosis. Dinitroaniline-induced microtubule loss typically results in the swelling of root tips as cells in this region fail to divide or elongate. Trifluralin is readily absorbed by young roots. Established weeds are not controlled.

2.4.4 Use Characterization

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

Trifluralin is a preemergent herbicide used to manage broadleaf weeds and annual grasses and on an array of food crops and is also registered for non-food uses, including residential uses. Formulations include emulsifiable concentrates and granulars.

Trifluralin may be applied with a wide range of application equipment including aircraft, ground spray, soil incorporation treatment, hand held granule applicator, shaker jar and soil broadcast treatment. Trifluralin may be applied at various stages including pre-plant, pre-emergence, emergence, dormant stage, established plantings, post-emergence, and/or post harvest. The set of registered trifluralin products evaluated for the Label Use Information System (LUIS) Report (Biological and Economic Analysis Division (BEAD), dated 3-24-09) is provided in **Table 2.2**. This set of evaluated labels does not constitute all registered trifluralin products; however, it is expected to be a representative set.

Table 2.2 Summary of Labels included in the LUIS Report for Currently Registered Trifluralin Uses

Product Name	Registration Number	Type of Formulation[a]	Trifluralin (%)	Other Active Ingredients	Label Date
MACCO TRIFLURALIN E.C. HERBICIDE	019713-00254	EC[a]	44.5	none	06/17/99
TREFLAN TR-10 GRANULES	062719-00131	G	10	none	12/01/05
TREFLAN HFP	062719-00250	EC	43	none	01/21/02
DREXEL TRIFLURALIN	019713-00543	G	5	none	08/27/02
TREFLAN 5 G	062719-00098	G	5	none	11/20/01
SNAPSHOT 2.5 TG	062719-00175	G	2	isoxaben	02/27/02
TREFLAN E.C. WEED AND GRASS PREVENTER	062719-00097	EC	43	none	02/10/09
GOWAN TRIFLURALIN 10G	CA94000300	G	10	none	06/23/05
SHOWCASE	062719-00516	G	2	isoxaben, oxyfluorefen	09/05/07
TURF FERTILIZER CONTAINS TEAM 1.15%	062719-00192	G	0.27	benefin, isoxaben	06/13/08
Turf Fertilizer - Contains Gallery Plus Team Pro	062719-00565	G	0.5	benefin, isoxaben	04/09/08
SUPER TEAM 1.25%	062719-00327	G	0.43	flumetsulam	02/05/99

Table 2.2 Summary of Labels included in the LUIS Report for Currently Registered Trifluralin Uses					
Product Name	**Registration Number**	**Type of Formulation[a]**	**Trifluralin (%)**	**Other Active Ingredients**	**Label Date**
BROADSTRIKE + TREFLAN	062719-00222	EC	36.35	benefin	07/21/08
T&O FERTILIZER-CONTAINS GALLERY PLUS TEAM	62719-00280	G	0.39	benefin, isoxaben	08/04/08
TURF FERTILIZER – CONTAINS TEAM PRO	62719-00289	G	0.86	benefin	04/09/08
[a] EC = emulsifiable concentrate, G =granular					

The LUIS report from the Biological and Economic Analysis Division (BEAD) was reviewed for active trifluralin label registrations. Because of the large number of labeled uses for trifluralin, categories were created for subsets of crops and non-agriculture uses with similar growing conditions and application rates to make exposure modeling and the resulting analysis workable. These assigned categories are assumed to be representative of the maximum application rates and uses for trifluralin. **Table 2.3** lists the uses and corresponding application rates and methods considered in this assessment. The highest application rates, maximum number of applications per year, and shortest application interval are employed for PRZM/EXAMS modeling. In the instances when application rates, maximum number of applications per year, and shortest application intervals were not specified in the LUIS Report, representative rates and intervals were assumed based on management practices and use rates on similar crops. Additional sources of data include California Pesticide Use Reporting (Cal PUR) data and USDA crop profile information. Twenty-five crop categories were designated to estimate EECs and risk as surrogates for all of the uses included in a given crop category. A master label was not provided by SRRD; however, SRRD did confirm that the use table (**Table 2.3**) developed for this assessment was correct (**Appendix Q**).

Table 2.3 Trifluralin Uses Assessed in California

General crop category	Representa-tive Crops	Form-ulation[1]	Appli-cation Method	Soil Incorp-oration (Y/N)[4]	Maximum Single Application Rate (lbs/acre)	Max Number of Applications per Year (Minimum Interval)	Represent-ative label
Orchard Uses							
Avocado	Avocado	G[1]	Ground	N	4.0	3[2] (60)	Snapshot 2.5 TG 062719-00175
Citrus	Grapefruit, lemon, orange, tangelo, tangerine	G	Ground	N	4.0	3[2] (60)	Snapshot 2.5 TG 062719-00175
		EC	Aerial Ground	Y	2.0	1[3] (NA)	Treflan HFP 062719-00250
Grape and berry	Blackberry, blueberry, boysenberry, currant, dewberry, elderberry, gooseberry, grapes, kiwi fruit, loganberry, mulberry, raspberry (black, red)	G	Ground	N	4.0	3[2] (60)	Snapshot 2.5 TG 062719-00175
Grape	Grape	EC	Aerial Ground	Y	2.0	1[3] (NA)	Treflan HFP 062719-00250
Olive	Olive	G	Ground	N	4.0	3[2] (60)	Snapshot 2.5 TG 062719-00175
Stone and pome fruit	Apples, apricot, cherry, fig, fruits unspecified, nectarine, peach, pear, plum, pomegranate, prune	G	Ground	N	4.0	3[2] (60)	Snapshot 2.5 TG 062719-00175
		EC	Aerial Ground	Y	2.0	1[3] (NA)	Treflan HFP 062719-00250
Tree nuts	Almond, filbert, macadamia,	G	Ground	N	4.0	3[2] (60)	Showcase 062719-00516

Table 2.3 Trifluralin Uses Assessed in California							
General crop category	**Representa-tive Crops**	**Form-ulation[1]**	**Appli-cation Method**	**Soil Incorp-oration (Y/N)[4]**	**Maximum Single Application Rate (lbs/acre)**	**Max Number of Applications per Year (Minimum Interval)**	**Represent-ative label**
	pecan, pistachio, tree nuts, walnut (English/black)	EC	Aerial Ground	Y	2.0	1[3] (NA)	Treflan HFP 062719-00250
Non-Orchard Agricultural Uses							
Alfalfa	Alfalfa, clover	G	Aerial Ground	Y	2.0	2 (60)	Treflan TR-10 Granules 062719-00131
		EC	Aerial Ground	N	1.0	1[3] (NA)	Treflan HFP 062719-00250
Potato	White/Irish potato, turnip	EC	Ground	Y	1.0	1	Treflan HFP 062719-00250
Tomato	Okra, tomato						
Melon	Cucumber, melon, watermelon						
Cole Crops	Broccoli, broccoli raab, Brussels spouts, cabbage, cauliflower, collards, crambe, kale, mustard, canola/rape	EC	Aerial Ground	Y	1.0	1[3] (NA)	Treflan HFP 062719-00250
Corn	Corn field, kenaf, sunflower	EC	Aerial Ground	Y	1.0	1[3] (NA)	Treflan HFP 062719-00250
Cotton	Cotton	EC	Aerial Ground	Y	2.0	1[3] (NA)	Treflan HFP 062719-00250
Lettuce	Chicory, endive	EC	Aerial Ground	Y	1.0	1[3] (NA)	Treflan HFP 062719-00250
Onion	Onion (dry bulb) Radish,	EC	Aerial Ground	Y	0.8	1[3] (NA)	Treflan HFP 062719-00250

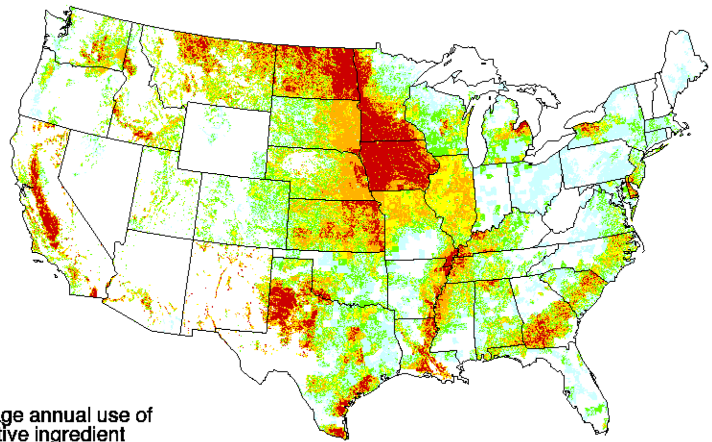
Table 2.3 Trifluralin Uses Assessed in California							
General crop category	**Representa-tive Crops**	**Form-ulation[1]**	**Appli-cation Method**	**Soil Incorp-oration (Y/N)[4]**	**Maximum Single Application Rate (lbs/acre)**	**Max Number of Applications per Year (Minimum Interval)**	**Represent-ative label**
Rangeland Hay	Bermuda grass, forage, fodder, hay, straw, un-cultivated agriculture	G	Ground	N	2.0	1[3] (NA)	Turf Fertilizer – contains Team Pro 0.86% 062719-00289
Row Crop	Asparagus, bean (castor) bean (lima), bean (mung), bean succulent, bean (snap), carrot, celery, guar, lentil, peas(southern) peas (succulent), pepper, sugar cane	EC	Aerial Ground	Y	2.0	1[3] (NA)	Treflan HFP 062719-00250
Sugar Beet	Sugar beet	EC	Aerial Ground	Y	0.75	1[3] (NA)	Treflan HFP 062719-00250
Wheat	Barley, hops, flax, safflower, sorghum, unspecified grains, wheat	EC	Aerial Ground	Y	1.0	1[3] (NA)	Treflan HFP 062719-00250
Non-agricultural Uses							
Nursery	Banana, greenhouse- in use ornamental shade trees, ornamental ground cover, ornamental herbaceous plants, ornamental	G	Ground	N	4.0	3 (60)	Snapshot 2.5 TG 062719-00175

Table 2.3 Trifluralin Uses Assessed in California							
General crop category	**Representa-tive Crops**	**Form-ulation[1]**	**Appli-cation Method**	**Soil Incorp-oration (Y/N)[4]**	**Maximum Single Application Rate (lbs/acre)**	**Max Number of Applications per Year (Minimum Interval)**	**Represent-ative label**
	lawns and turf, ornamental non-flowering plants, ornamental woody shrubs and vines residential lawns	EC	Ground	N	4.0	3[2] (60)	062719-00097
Resident-ial	Ornamental and/or shade trees, ornamental ground cover, ornamental herbaceous plants, ornamental lawns and turf, ornamental non-flowering plants, ornamental woody shrubs and vines residential lawns	G	Ground	N	1.5	2 (56)	Showcase 062719-00516
Rights-of-way	Pre-paving, private roads/side-walks, non-agriculture rights-of-way, fencerows, hedgerows	G	Ground	N	4.0	3[2] (60)	Showcase 062719-00516
Turf	Golf course turf, recreat-ional, residential lawns, commercial/in dustrial lawns	G	Ground	N	1.5	2 (56)	Turf Fertilizer – contains Team Pro 0.86% 062719-00289
Forestry	Christmas tree plantations	G	Ground	N	4.0	3[2] (60)	Showcase 062719-00516

Table 2.3 Trifluralin Uses Assessed in California							
General crop category	**Representa-tive Crops**	**Form-ulation[1]**	**Appli-cation Method**	**Soil Incorp-oration (Y/N)[4]**	**Maximum Single Application Rate (lbs/acre)**	**Max Number of Applications per Year (Minimum Interval)**	**Represent-ative label**
	Cottonwood (forest/shelterbelt), poplar	EC	Aerial Ground	Y	2.0	1[3] (NA)	Treflan HFP 062719-00250

[1] EC = Emulsifiable Concentrate, G = Granular
[2] Maximum number of applications per year is not specified on label or in LUIS Report. For this assessment it is assumed that a maximum of 3 applications of 4 lbs trifluralin at a minimum of 60 day intervals is applied. This application rate is comparable to numerous annual maximum application rates for granular nursery and orchard uses.
[3] The LUIS Report listed many of the number of applications per year and maximum lbs trifluralin per year as NA (not available). EFED looked at the representative label (next column) and inferred that a single application per crop cycle was intended as a specific time in the growing cycle of the crop was provided for trifluralin application (*e.g.*, if label specified triflluralin application was to be pre-plant, it was assumed this product applied once per crop cycle).
[4] Y = soil incorporation is required within 24 hours, N = soil incorporation is greater than 24 hours or not at all.

A national map **(Figure 2.1)** showing the estimated poundage of trifluralin uses across the United States is provided below. The map was downloaded from a U.S. Geological Survey (USGS), National Water Quality Assessment Program (NAWQA) website (http://water.usgs.gov/nawqa/pnsp/usage/maps/). These data suggest that at a national level, trifluralin is primarily applied to soybeans, alfalfa hay, cotton, and wheat for grain. Use intensity is highest in several parts of the country including areas of the Southeast, upper mid-west, Texas, and the central valley of California. Use data in California is discussed in greater detail below.



Crops	Total pounds applied	Percent national use
soybeans	2999382	33.69
cotton	2672127	30.02
alfalfa hay	1063635	11.95
wheat for grain	715160	8.03
sugarcane	304406	3.42
sunflower seed	274123	3.08
dry beans	260349	2.92
tomatoes	108776	1.22
sorghum	62492	0.70
green beans	58753	0.66

Figure 2.1 Estimated Trifluralin Use in 2002, Total Pounds Based on Data at the County Level

The Agency's Biological and Economic Analysis Division (BEAD) provides an analysis of both national- and county-level usage information using state-level usage data[1] obtained from USDA-NASS[2], 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) database[3]. CDPR PUR is considered a more comprehensive source of usage data than USDA-NASS or EPA proprietary databases, and thus the usage data reported for trifluralin 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. Data from CDPR PUR were obtained for every agricultural pesticide application made on every use site at the section level

[1] Memo from Carter and Kaul (BEAD) to Galavotti (EFED), June 3, 2009: County-Level Usage for strychnidin; strychnine; triclopyr, butoxyethyl ester; triclopyr, triethylamine salt; diflubenzuron; trifluralin; thiobencarb; chlorpyrifos; vinclozolin; iprodione in California in Support of a Red Legged Frog Endangered Species Assessment, DP # TBD

[2] United States Depart of Agriculture (USDA), National Agricultural Statistics Service (NASS) Chemical Use Reports provide summary pesticide usage statistics for select agricultural use sites by chemical, crop and state. See http://www.usda.gov/nass/pubs/estindx1.htm#agchem.

[3] 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.

(approximately one square mile) of the public land survey system.[4] BEAD summarized these data to the county level by site, pesticide, and unit treated. Calculating county-level usage involved summarizing across all applications made within a section and then across all sections within a county for each use site and for each pesticide. The county level usage data that were calculated include: average annual pounds applied, average annual area treated, and average and maximum application rate across all eight years. The units of area treated are also provided where available.

A summary of all trifluralin uses in California based on the modeled scenarios is provided in **Table 2.4**. Use data for the 20 counties with the highest usage is provided in **Table 2.5**. Complete data tables can be found in **Appendix C**.

From 1999 to 2006, trifluralin was used on 81 crops or sites in 52 counties in California. The herbicide was used in the greatest quantity on alfalfa with an average yearly application of 601,216 lbs/year. The greatest quantity of trifluralin was applied in Imperial County with a yearly average application of 259,148 lbs/year. The highest average application rate over the eight year period was 1.7 lbs/acre applied in Imperial County.

Nearly all the maximum application rates recorded in the 1999 to 2006 CDPR PUR data exceed the maximum application rates permitted on trifluralin labels (see **Tables 2.4** and **2.5**). This likely indicates data entry errors in the pounds applied or the acres treated. The 95th and 99th percentile estimations of application rates aggregated by cropping category were, for the majority, less than the maximum labeled rates. **Figure 2.2** shows the scenarios with the highest usage as a fraction of the entire average annual usage.

[4] Most pesticide applications to parks, golf courses, cemeteries, rangeland, pastures, and along roadside and railroad rights of way, and postharvest treatments of agricultural commodities. The primary exceptions to the reporting requirement are home-and-garden use and most industrial and institutional uses (http://www.cdpr.ca.gov/docs/pur/purmain.htm).

Table 2.4 Summary of California Department of Pesticide Registration (CDPR) Pesticide Use Reporting (PUR) Data from 1999 to 2006 for Currently Registered Trifluralin Uses Based on Aggregated Scenarios[1]

General Crop Category	Average Annual Application (lbs/year) [2]	Application Rate (lbs/acre) [2]		
		AVG	95th % ile	99th % ile
Alfalfa	601,216	2.0	2.8	3.8
Cotton	180,135	0.8	1.2	1.4
Tomato	108,155	0.6	1.2	1.5
Row crop	53,306	0.7	0.9	1.4
Wheat	23,195	0.8	1.3	1.6
Rangeland hay	22,828	1.5	1.8	1.8
Tree nuts	20,301	1.6	3.1	3.4
Sugar beet	10,166	0.8	1.0	1.5
Grapes	9,248	1.0	1.8	3.1
Melon	7,528	1.0	3.5	3.8
Grapes (wine)	7,086	4.1	4.9	6.8
Corn	5,625	0.9	1.1	1.2
Cole crop	3,833	0.6	1.1	1.3
Right of way	3,393	1.8	NA	NA
Citrus	3,132	0.6	0.9	1.1
Nursery	2,835	1.2	2.2	3.0
Stone and pome fruit	1,001	1.1	1.5	1.7
Potato	568	14.3	14.3	14.8
Onion	356	0.7	0.8	1.2
Lettuce	121	0.5	0.6	0.6
Forestry	2	NA	NA	NA
Turf	2	NA	NA	NA
Olive	1	NA	NA	NA
Others	8,281			
TOTAL FOR ALL SCENARIOS	**1,072,315**			

1- Based on data supplied by BEAD (Memo from Carter and Kaul (BEAD) to Galavotti (EFED), June 3, 2009)
2- The average annual pounds applied and average application rate was calculated as the weighted average of the average application rate for one county or average annual pounds applied for one county. The values reflect the average annual pounds applied to that site across all counties and the average application rate for that site across all counties.

Figure 2.2 Scenarios with Highest Trifluralin Use Based on CDPR PUR Data from 1999-2006

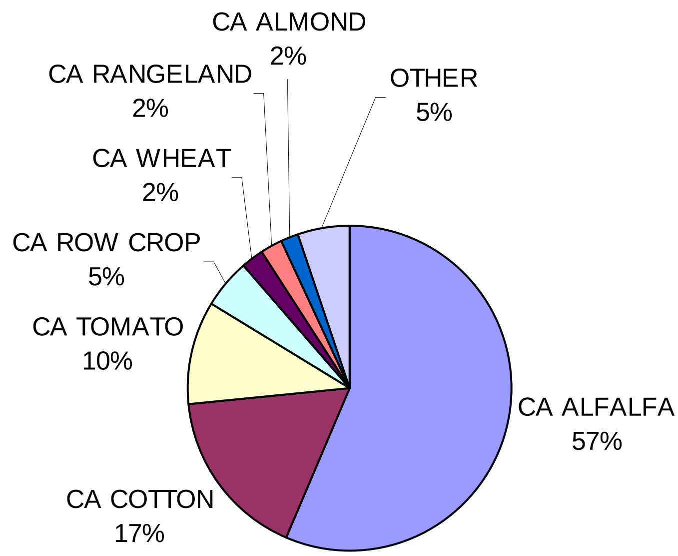


Table 2.5 Summary of California Department of Pesticide Registration (CDPR) Pesticide Usage Reporting (PUR) Data from 1999 to 2006 for 20 Counties with Highest Trifluralin Usage[1]		
County	**Average Annual Application (lbs/year)**	**Application Rate (lbs/acre)**
		Average
IMPERIAL	259,148	1.7
FRESNO	169,941	1.0
MERCED	117,817	0.8
KINGS	106,406	1.1
KERN	74,838	1.0
SAN JOAQUIN	55,881	1.3
TULARE	55,235	1.3
RIVERSIDE	50,637	NA
YOLO	49,212	0.8
MADERA	27,182	1.2
STANISLAUS	26,827	0.8
SOLANO	13,741	0.9
SUTTER	11,693	0.8
SACRAMENTO	9,595	1.0
COLUSA	8,401	0.7
LOS ANGELES	7,772	1.2

Table 2.5 Summary of California Department of Pesticide Registration (CDPR) Pesticide Usage Reporting (PUR) Data from 1999 to 2006 for 20 Counties with Highest Trifluralin Usage[1]		
County	**Average Annual Application (lbs/year)**	**Application Rate (lbs/acre)**
		Average
GLENN	7,581	1.5
SAN LUIS OBISPO	5,153	1.0
SANTA BARBARA	4,280	0.6
SAN BERNADINO	3,685	0.9
All Other Counties	7,290	NA
TOTAL	**1,072,315**	NA
1- Based on data supplied by BEAD (Memo from Carter and Kaul (BEAD) to Galavotti (EFED), June 3, 2009)		

2.5 Assessed Species

Table 2.6 provides a summary of the current distribution, habitat requirements, and life history parameters for the four listed species being assessed. More detailed life history and distribution information can be found for CRLF in **Attachment 1** and for DS, SFGS, and SJKF in **Attachment 3**. **Figures 2.3 to 2.6** provide maps of the current range and habitats of the assessed listed species.

Table 2.6 Summary of Current Distribution, Habitat Requirements, and Life History Information for the Assessed Listed Species[1]						
Assessed Species	**Size**	**Current Range**	**Habitat Type**	**Desig-nated Critical Habitat?**	**Reproductive Cycle**	**Diet**
California red-legged frog (*Rana aurora draytonii*)	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	Breeding: Nov. to Apr. Tadpoles: Dec. to Mar. Young juveniles: Mar. to Sept.	Aquatic-phase[2]: algae, freshwater aquatic invertebrates Terrestrial-phase: aquatic and terrestrial invertebrates, small mammals, fish and frogs
Delta smelt (*Hypomesus transpacificus*)	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.	Primarily planktonic copepods, cladocerans, amphipods, and insect larvae. Larvae feed on phytoplankton; juveniles feed on zooplankton.
San Francisco garter snake (*Thamnophis sirtalis tetrataenia*)	Adult (46-131 cm in length), Females – 227 g, Males – 113 g; Juveniles (18–20 cm in	San Mateo County	Densely vegetated freshwater ponds near open grassy hillsides; emergent vegetation; rodent burrows	No	Oviparous Reproduction[3] Breeding: Spring (Mar. and Apr.) and Fall (Sept. to Nov.) Ovulation and Pregnancy: Late spring and early summer Young: Born 3-4	Juveniles: frogs (Pacific tree frog, CRLF, and bullfrogs depending on size) and insects Adults: primarily frogs (mainly CRLFs; also bullfrogs, toads); to a lesser extent newts; freshwater fish and invertebrates; insects and small mammals

Table 2.6 Summary of Current Distribution, Habitat Requirements, and Life History Information for the Assessed Listed Species[1]						
Assessed Species	**Size**	**Current Range**	**Habitat Type**	**Desig-nated Critical Habitat?**	**Reproductive Cycle**	**Diet**
	length)				months after mating	
San Joaquin kit fox (*Vulpes macrotis mutica*)	Adult ~2 kg	Alameda, Contra Costa, Fresno, Kern, Kings, Madera, Merced, Monterey, San Benito, San Joaquin, San Luis Obispo, Santa Barbara, Santa Clara, Stanislaus, Tulare and Ventura counties	A variety of habitats, including grasslands, scrublands (*e.g.*, chenopod scrub and sub-shrub scrub), vernal pool areas, oak woodland, alkali meadows and playas, and an agricultural matrix of row crops, irrigated pastures, orchards, vineyards, and grazed annual grasslands. Kit foxes dig their own dens, modify and use those already constructed by other animals (ground squirrels, badgers, and coyotes), or use human-made structures (culverts, abandoned pipelines, or banks in sumps or roadbeds). They move to new dens within their home range often (likely to avoid predation by coyotes)	No, but has designated core areas	Mating and conception: late December - March. Gestation period: 48 to 52 days. Litters born: February - late March Pups emerge from their dens at about 1-month of age and may begin to disperse after 4 – 5 months usually in Aug. or Sept.	Small animals including blacktailed hares, desert cottontails, mice, kangaroo rats, squirrels, birds, lizards, insects and grass. It satisfies its moisture requirements from prey and does not depend on freshwater sources.

[1] For more detailed information on the distribution, habitat requirements, and life history information of the assessed listed species, see Attachments 1 and 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.

[3] Oviparous = eggs hatch within the female's body and young are born live.

Recovery Units

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

Core Areas

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

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

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

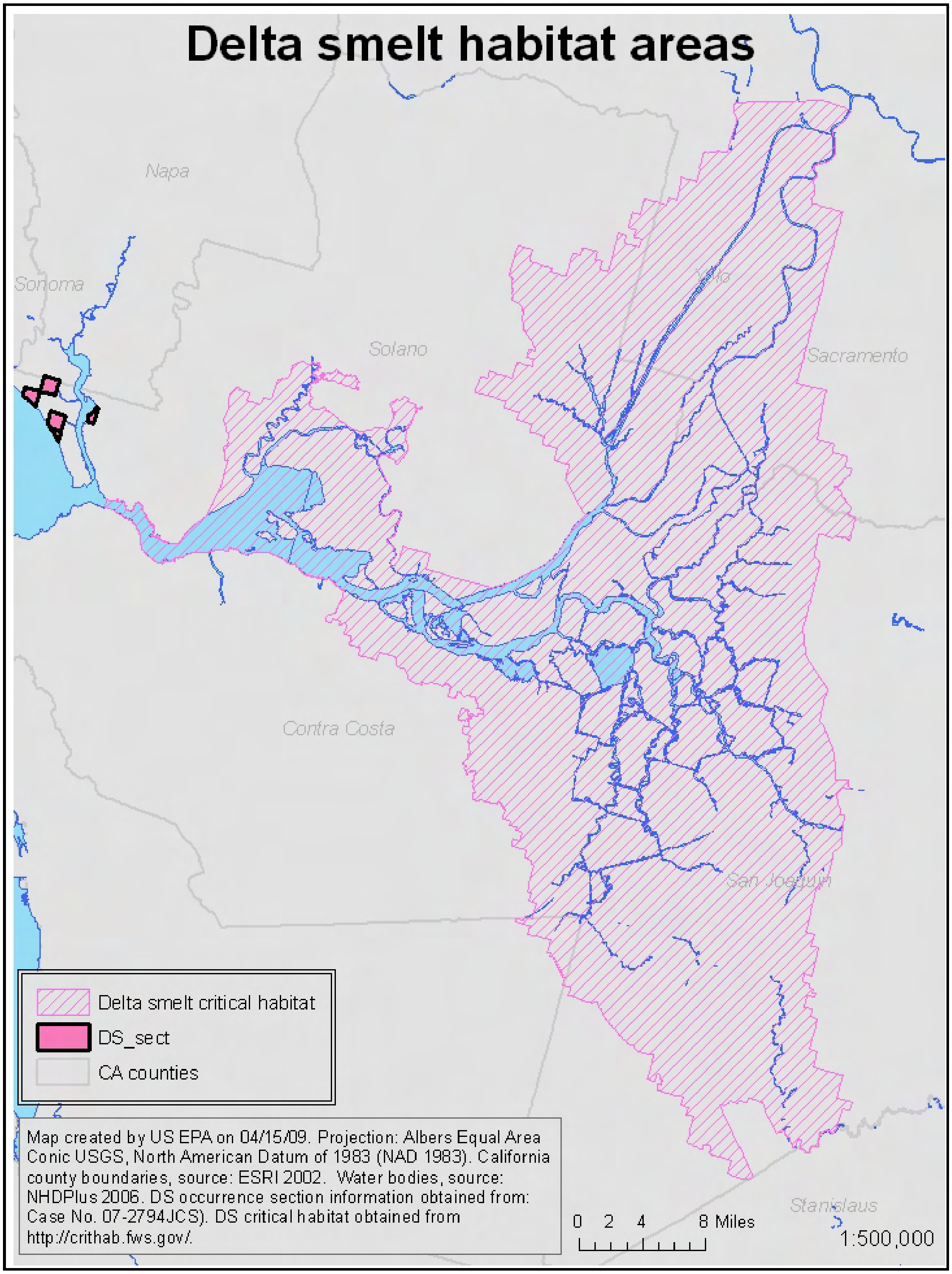


Figure 2.4 Delta Smelt Habitat Areas

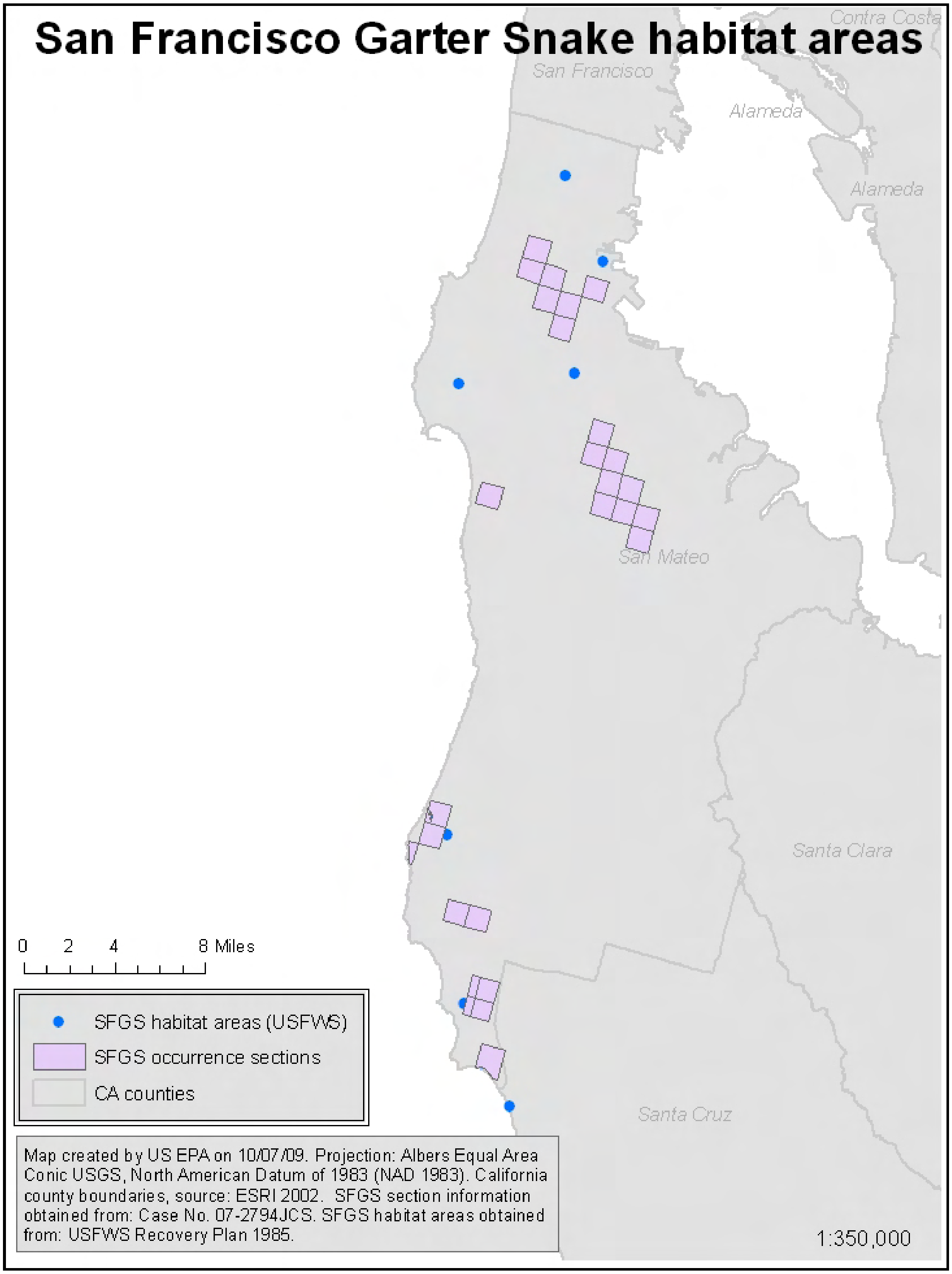


Figure 2.5 San Francisco Garter Snake Habitat Areas

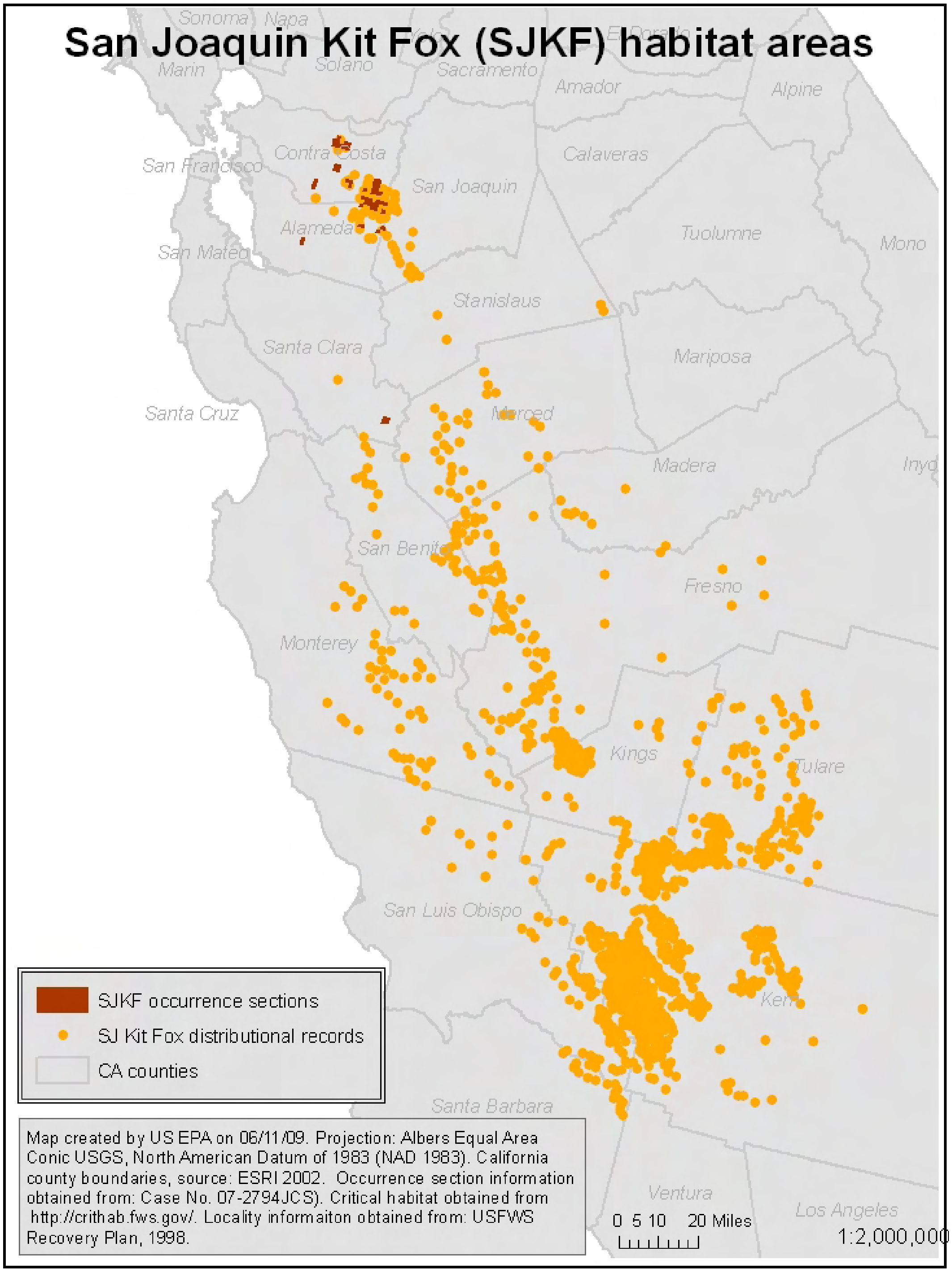


Figure 2.6 San Joaquin Kit Fox Habitat Areas

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 2.7** describes the PCEs for the critical habitats designated for the CRLF and the DS.

Table 2.7 Designated Critical Habitat PCEs for the CRLF and the DS

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 (*e.g.*, 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.	

Table 2.7 Designated Critical Habitat PCEs for the CRLF and the DS		
Species	**PCEs**	**Reference**
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.e.*, low "concentrations of pollutants) and substrates for egg attachment (*e.g.*, 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 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.	
[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.		

More detail on the designated critical habitat applicable to this assessment can be found in **Attachment 1** (CRLF) and **Attachment 3** (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 trifluralin that may alter the PCEs of the designated critical habitat for the CRLF and the DS form the basis of the critical habitat impact analysis.

As previously noted in **Section 2.1**, the Agency believes that the analysis of direct and indirect effects to listed species provides the basis for an analysis of potential effects on the designated critical habitat. Because trifluralin is expected to directly impact living organisms within the action area, critical habitat analysis for trifluralin 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 trifluralin is likely to encompass considerable portions of the United States based on the large array of agricultural and/or non-agricultural uses. However, the scope of this assessment limits consideration of the overall action area to those portions that may be applicable to the protection of the CRLF, DS, SFGS, and SJKF and their designated critical habitat. For the purposes of this assessment, attention will be focused on the footprint of the action (*i.e.*, the area where pesticide application occurs), plus all areas where offsite transport (*i.e.*, spray drift, downstream dilution, etc.) may result in potential exposure that exceeds the Agency's LOCs. Although the watershed for the San Francisco Bay extends northward into the very southwestern portion of Lake County, Oregon, and westward into the western edge of Washoe County, Nevada, the non-California portions of the watershed are small and very rural with little, if any, agriculture. Therefore, no use of trifluralin is expected in these areas, and they are not considered as part of the action area applicable to this assessment.

The definition of action area requires a stepwise approach that begins with an understanding of the federal action. The federal action is defined by the currently labeled uses for trifluralin. An analysis of labeled uses and review of available product labels was completed. Several of the currently labeled uses are special local needs (SLN) uses or are restricted to specific states and are excluded from this assessment. In addition, a distinction has been made between food use crops and those that are non-food/non-agricultural uses. For those uses relevant to the assessed species, the analysis indicates that, for trifluralin, the following agricultural uses are considered as part of the federal action evaluated in this assessment:

(1) Orchard Agricultural uses

Almond, apple, apricot, avocado, blackberry, blueberry, boysenberry, cherry, currant, dewberry, elderberry, fig, filbert, fruits unspecified, gooseberry, grape, grapefruit, kiwi fruit, lemon, loganberry, macadamia, mulberry, nectarine, orange, olive, peach, pear, pecan, pistachio, plum, pomegranate, prune, raspberry (black, red), tangerine, tangelo, tree nuts, (English/black) walnut

(2) Non-Orchard Agricultural uses

Alfalfa, asparagus, barley, broccoli, broccoli rabbinni, beans (succulent), beans (lima), beans (mung), beans (snap), Bermuda grass, Brussels spouts, cabbage, cauliflower, canola/rape, carrot, castor, celery, chicory, clover, collards, corn field, cotton, crambe, cucumber, endive, flax, guar, hops, kale, kenaf, lentil, melon, mustard, okra, onion (dry bulb), peas(southern), peas (succulent), pepper, radish, safflower, sorghum, sugar beet, sugar cane, sunflower, tomato, turnip, uncultivated agriculture, uncultivated areas/soils, unspecified grains, watermelon, white/Irish potato, wheat

(3) Non-agricultural uses

> Banana (ornamental), commercial/industrial lawns, Christmas tree plantations, cottonwood (forest/shelterbelt), golf course turf, greenhouse- in use, industrial areas, non-agriculture outdoor buildings and structures, non-agriculture rights-of-way, fencerows, hedgerows, ornamental and/or shade trees, ornamental ground cover, ornamental herbaceous plants, ornamental lawns and turf, ornamental non-flowering plants, ornamental woody shrubs and vines, residential lawns, pre-paving, poplar, private roads/sidewalks, recreational, residential lawns

Following a determination of the assessed uses, an evaluation of the potential "footprint" of trifluralin use patterns (*i.e.*, the area where pesticide application occurs) is determined. This "footprint" represents the initial area of concern, based on an analysis of available land cover data for the state of California. The initial area of concern is defined as all land cover types and the stream reaches within the land cover areas that represent the labeled uses described above. Because of the wide variety of labeled trifluralin uses ranging from orchard, agricultural, nursery, turf, rights-of-way, and residential, the initial area of concern was defined as the entire state of California. Precluding geographic areas of the state given these uses is not possible.

Once the initial area of concern is defined, the next step is to define the potential boundaries of the action area by determining the extent of offsite transport via spray drift and runoff where exposure of one or more taxonomic groups to the pesticide exceeds the listed species LOCs. The environmental fate properties of trifluralin along with monitoring data identifying its presence in surface waters, air and precipitation in California indicate that runoff, spray drift, volatilization and atmospheric transport and (wet) deposition represent significant potential transport mechanisms of trifluralin to the aquatic and terrestrial habitats of the CRLF, DS, SJKF and SFGS.

The Agency's approach to defining the action area under the provisions of the Overview Document (USEPA 2004) considers the results of the risk assessment process to establish boundaries for that action area with the understanding that exposures below the Agency's defined Levels of Concern (LOCs) constitute a no-effect threshold. Deriving the geographical extent of this portion of the action area is based on consideration of the types of effects that trifluralin may be expected to have on the environment, the exposure levels to trifluralin that are associated with those effects, and the best available information concerning the use of trifluralin and its fate and transport within the state of California. Specific measures of ecological effect for the assessed species that define the action area include any direct and indirect toxic effect to the assessed species and any potential modification of its critical habitat, including reduction in survival, growth, and fecundity as well as the full suite of sublethal effects available in the effects literature. Therefore, the action area extends to a point where environmental exposures are below any measured lethal or sublethal effect threshold for any biological entity at the whole organism, organ, tissue, and cellular level of organization. In situations where it is not possible to determine the threshold for an observed effect, the action area is not spatially limited and is assumed to be the entire state of California.

Because a NOAEC value was not established for estuarine/marine chronic fish study and because trifluralin has been recognized as a possible human carcinogen (Group C Carcinogen, HED Human Health Risk Assessment, May 7, 2004, DP barcode 296628; **Appendix G**), the spatial extent of the action area (*i.e.*, the boundary where exposures and potential effects are less than the Agency's LOC) for trifluralin cannot be determined. Therefore, it is assumed that the action area encompasses the entire state of California, regardless of the spatial extent (*i.e.*, initial area of concern or footprint) of the pesticide use(s). In addition, it is reasonable to assume that the action area encompasses the entire state of California given the broad range of labeled uses and the large geographic coverage of the state for those uses.

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, DS, SFGS, and SJKF), 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 trifluralin (*e.g.*, runoff, spray drift, *etc*.), and the routes by which ecological receptors are exposed to trifluralin *(e.g.,* direct contact, *etc.).*

2.8.1 Assessment Endpoints

Assessment endpoints for the CRLF, DS, SFGS, and SJKF 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 trifluralin is provided in **Section 2.5** and **Table 2.9**.

[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 taxon is used for risk estimation. For this assessment, evaluated taxa include freshwater fish (surrogate for aquatic-phase amphibians), freshwater invertebrates, aquatic plants, birds (surrogate for terrestrial-phase amphibians), mammals, terrestrial invertebrates, and terrestrial plants. Additional taxa evaluated for the DS are estuarine/marine fish and estuarine/marine invertebrates; the most sensitive of the freshwater and estuarine/marine species are used for the risk assessment. 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 trifluralin.

Table 2.8 identifies the taxa used to assess the potential for direct and indirect effects from the uses of trifluralin for each listed species assessed here. The specific assessment endpoints used to assess the potential for direct and indirect effects to each listed species are provided in **Table 2.9**.

Table 2.8 Taxa Used in the Analyses of Direct and Indirect Effects for the Assessed Listed Species.

Listed Species	Birds	Mammals	Terr. Plants	Terr. Inverts.	FW Fish	FW Inverts.	E/M Fish	E/M Inverts.	Aquatic Plants[1]
CRLF[3]	Direct Indirect (prey)	Indirect (prey)	Indirect (habitat)	Indirect (prey)	Direct Indirect (prey)	Indirect (prey)	N/A	N/A	Indirect (food/ habitat)
SFGS[3]	Direct Indirect (prey)	Indirect (prey/ habitat)	Indirect (habitat)	Indirect (prey)	Indirect (prey)	Indirect (prey)	N/A	N/A	Indirect (habitat)
SJKF	Indirect (prey)	Direct Indirect (prey)	Indirect (food/ habitat)	Indirect (prey)	N/A	N/A	N/A	N/A	N/A
DS	N/A	N/A	Indirect (habitat)	N/A	Direct [2]	Indirect (prey) [2]	Direct[2]	Indirect[2] (prey)	Indirect (food/ habitat)

[1] Vascular and non-vascular aquatic plants are assessed separately. Data from the most sensitive plant species of each group (vascular and non-vascular) will be used, regardless of habitat (freshwater or estuarine/marine).
[2] The most sensitive species in either freshwater or estuarine/marine environments was used for the DS assessment.
[3] Consumption of residues in aquatic organisms may result in direct effects to the CRLF and SFGS.

N/A = Not applicable
Terr. = Terrestrial
Invert. = Invertebrate
FW = Freshwater
E/M = Estuarine/Marine

Table 2.9 Taxa and Assessment Endpoints Used to Evaluate the Potential for the Use of Trifluralin to Result in Direct and Indirect Effects to the Assessed Listed Species.			
Taxa Used to Assess Direct and/or Indirect Effects to Assessed Species	**Assessed Listed Species**	**Assessment Endpoints**	**Measures of Ecological Effects[1]**
1. Freshwater Fish and Aquatic-phase Amphibians	Direct Effect – -Aquatic-phase CRLF[2] - DS	Survival, growth, and reproduction of individuals via direct effects	1a. Bluegill sunfish acute LC_{50} 1b. Bluegill sunfish chronic NOAEC (estimated by ACR)
	Indirect Effect (prey) -Aquatic-phase CRLF -SFGS	Survival, growth, and reproduction of individuals via indirect effects on aquatic prey food supply (*i.e.*, fish and aquatic-phase amphibians)	
2. Freshwater Invertebrates	Indirect Effect (prey) -Aquatic-phase CRLF -DS -SFGS	Survival, growth, and reproduction of individuals via indirect effects on aquatic prey food supply (*i.e.*, freshwater invertebrates)	2a. Daphnid acute EC_{50} 2b. Daphnid chronic NOAEC
3. Estuarine/Marine Fish	Direct Effect – - DS	Survival, growth, and reproduction of individuals via direct effects	3a. Bluegill sunfish acute LC_{50} 3b. Bluegill sunfish chronic NOAEC (estimated by ACR) *Since the DS inhabits freshwater and brackish waters, the most sensitive of the freshwater and estuarine/marine organisms is used for the assessment.*
	Indirect Effect (prey) -DS	Survival, growth, and reproduction of individuals via indirect effects on aquatic prey food supply (*i.e.*, estuarine/marine fish)	
4. Estuarine/Marine Invertebrates	Indirect Effect (prey) -DS	Survival, growth, and reproduction of individuals via indirect effects on aquatic prey food supply (*i.e.*, estuarine/marine invertebrates)	4a. Daphnid acute EC_{50} 4b. Daphnid chronic NOAEC *Since the DS inhabits freshwater and brackish waters, the most sensitive of the freshwater and estuarine/marine organisms is used for the assessment.*
5. Aquatic Plants (freshwater/marine)	Indirect Effect (food/habitat) -Aquatic-phase CRLF -DS -SFGS	Survival, growth, and reproduction of individuals via indirect effects on habitat, cover, food supply, and/or primary productivity (*i.e.*, aquatic plant community)	5a. Duckweed IC_{50} 5b. *Skeletonema costatum* IC_{50}
6. Birds, Terrestrial-phase Amphibians, and Reptiles	Direct Effect -Terrestrial-phase CRLF -SFGS	Survival, growth, and reproduction of individuals via direct effects	6a. Northern bobwhite quail and mallard duck acute oral LD_{50} 6b. Northern bobwhite quail and mallard duck acute dietary LC_{50} 6c. Mallard duck chronic NOAEC
	Indirect Effect (prey) -Terrestrial-phase CRLF -SFGS -SJKF	Survival, growth, and reproduction of individuals via indirect effects on terrestrial prey (birds)	

Table 2.9 Taxa and Assessment Endpoints Used to Evaluate the Potential for the Use of Trifluralin to Result in Direct and Indirect Effects to the Assessed Listed Species.

Taxa Used to Assess Direct and/or Indirect Effects to Assessed Species	Assessed Listed Species	Assessment Endpoints	Measures of Ecological Effects[1]
7. Mammals	Direct Effect -SJKF	Survival, growth, and reproduction of individuals via direct effects	7a. Laboratory rat acute LD$_{50}$ 7b. Laboratory rat chronic NOAEL
	Indirect Effect (prey/habitat from burrows) -Terrestrial-phase CRLF -SFGS -SJKF	Survival, growth, and reproduction of individuals via indirect effects on terrestrial prey (mammals)	
8. Terrestrial Invertebrates	Indirect Effect (prey) -Terrestrial-phase CRLF - SFGS - SJKF	Survival, growth, and reproduction of individuals via indirect effects on terrestrial prey (terrestrial invertebrates)	8a. Honey bee acute LD$_{50}$
9. Terrestrial Plants	Indirect Effect (food/habitat) (non-obligate relationship) -Terrestrial-phase CRLF - SFGS - SJKF	Survival, growth, and reproduction of individuals via indirect effects on food and habitat (*i.e.*, riparian and upland vegetation)	9a. Monocot EC$_{25}$: sorghum (seedling emergence) and corn (vegetative vigor) 9b. Dicot EC$_{25}$: cucumber (seedling emergence and vegetative vigor)

[1]Citations for all registrant-submitted and open literature toxicity data reviewed for this assessment are included in **Appendix F** and **Appendix H**.
[2]Adult frogs are no longer in the "aquatic phase" of the amphibian life cycle; however, submerged adult frogs are considered "aquatic" for the purposes of this assessment because exposure pathways in the water are considerably different than exposure pathways on land.

2.8.2 Assessment Endpoints for Designated Critical Habitat

As previously discussed, designated critical habitat is assessed to evaluate actions related to the use of trifluralin 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. It should be noted that evaluation of PCEs as assessment endpoints is limited to those of a biological nature (*i.e.*, the biological resource requirements for the listed species associated with the critical habitat) and those for which trifluralin effects data are available.

Some components of these PCEs are associated with physical abiotic features (*e.g.*, presence and/or depth of a water body, or distance between two sites), which are not expected to be measurably altered by use of pesticides. Measures of ecological effect used to assess the potential for modification to the critical habitat of the CRLF and the DS are described in **Table 2.10**. The SFGS and the SJKF do not have designated critical habitat.

Table 2.10 Summary of Assessment Endpoints and Measures of Ecological Effect for Primary Constituent Elements of Designated Critical Habitat for CRLF and the DS.			
Taxon Used to Assess Modification of PCE	**Assessed Listed Species Associated with the PCE**	**Assessment Endpoints**	**Measures of Ecological Effects [1]**
1. Freshwater Fish and Aquatic-phase Amphibians	Direct Effect – -Aquatic-phase CRLF [2] - DS	Survival, growth, and reproduction of individuals via direct effects	1a. Bluegill sunfish acute LC_{50} 1b. Bluegill sunfish chronic NOAEC (estimated by ACR)
	Indirect Effect (prey) -Aquatic-phase CRLF -DS	Modification of critical habitat via change in aquatic prey food supply (*i.e.*, fish and aquatic-phase amphibians)	
2. Freshwater Invertebrates	Indirect Effect (prey) -Aquatic-phase CRLF -DS	Survival, growth, and reproduction of individuals via indirect effects on aquatic prey food supply (*i.e.*, freshwater invertebrates)	2a. Daphnid acute EC_{50} 2b. Daphnid chronic NOAEC
3. Estuarine/Marine Fish	Direct Effect - DS Indirect Effect (prey) - DS	Modification of critical habitat via change in aquatic prey food supply (*i.e.*, estuarine/marine fish)	3a. Bluegill sunfish acute LC_{50} 3b. Bluegill sunfish chronic NOAEC (estimated by ACR) *Since the DS inhabits freshwater and brackish waters, the most sensitive of the freshwater and estuarine/marine organisms is used for the assessment.*
4. Estuarine/Marine Invertebrates	Indirect Effect (prey) - DS	Modification of critical habitat via change in aquatic prey food supply (*i.e.*, estuarine/marine invertebrates)	4a. Daphnid acute EC_{50} 4b. Daphnid chronic NOAEC *Since the DS inhabits freshwater and brackish waters, the most sensitive of the freshwater and estuarine/marine organisms is used for the assessment.*
5. Aquatic Plants (freshwater/marine)	Indirect Effect (food/habitat) -Aquatic-phase CRLF - DS	Modification of critical habitat via change in habitat, cover, food supply, and/or primary productivity (*i.e.*, aquatic plant community)	5a. Duckweed IC_{50} 5b. *Skeletonema costatum* IC_{50}
6. Birds	Direct Effect -Terrestrial-phase CRLF	Survival, growth, and reproduction of individuals via direct effects	6a. Northern bobwhite quail and mallard duck acute oral LD_{50} 6b. Northern bobwhite quail and mallard duck acute dietary LC_{50} 6c. Mallard duck chronic NOAEC
	Indirect Effect (prey) -Terrestrial-phase CRLF	Modification of critical habitat via change in terrestrial prey (birds)	

Table 2.10 Summary of Assessment Endpoints and Measures of Ecological Effect for Primary Constituent Elements of Designated Critical Habitat for CRLF and the DS.			
Taxon Used to Assess Modification of PCE	**Assessed Listed Species Associated with the PCE**	**Assessment Endpoints**	**Measures of Ecological Effects [1]**
7. Mammals	Indirect Effect (prey/habitat from burrows) -Terrestrial-phase CRLF -SFGS -SJKF	Modification of critical habitat via change in terrestrial prey (mammals)	7a. Laboratory rat acute LD_{50} 7b. Laboratory rat chronic NOAEL
8. Terrestrial Invertebrates	Indirect Effect (prey) -Terrestrial-phase CRLF	Modification of critical habitat via change in terrestrial prey (terrestrial invertebrates)	8a. Honey bee acute LD_{50}
9. Terrestrial Plants	Indirect Effect (food/habitat) (non-obligate relationship) -Terrestrial-phase CRLF	Modification of critical habitat via change in food and habitat (*i.e.*, riparian and upland vegetation)	9a. Monocot EC_{25}: sorghum (seedling emergence) and corn (vegetative vigor) 9b. Dicot EC_{25}: cucumber (seedling emergence and vegetative vigor)

[1]Citations for all registrant-submitted and open literature toxicity data reviewed for this assessment are included in **Appendix F** and **Appendix H**.
[2]Adult frogs are no longer in the "aquatic phase" of the amphibian life cycle; however, submerged adult frogs are considered "aquatic" for the purposes of this assessment because exposure pathways in the water are considerably different than exposure pathways on land.

2.9 Conceptual Model

2.9.1 Risk Hypotheses

Risk hypotheses are specific assumptions about potential 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 trifluralin to the environment. The following risk hypotheses are presumed for each assessed species in this assessment:

The labeled use of trifluralin within the action area may:

- directly affect the CRLF, DS, SFGS, and/or SJKF by causing mortality or by adversely affecting growth or fecundity;
- indirectly affect the CRLF, DS, SFGS, and/or SJKF and/or modify designated critical habitat of the CRLF or DS by reducing or changing the composition of food supply;

- indirectly affect the CRLF, DS and/or SFGS 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, DS, SFGS, and/or SJKF 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 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 trifluralin release mechanisms, biological receptor types, and effects endpoints of potential concern. The conceptual models for aquatic and terrestrial phases of the CRLF, DS, SFGS, and SJKF and the conceptual models for the aquatic and terrestrial PCE components of critical habitat for the CRLF and DS are shown in **Figures 2.7** and **2.8**. 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, DS, SFGS, and SJKF and modification to designated critical habitat for the CRLF and DS is expected to be negligible.

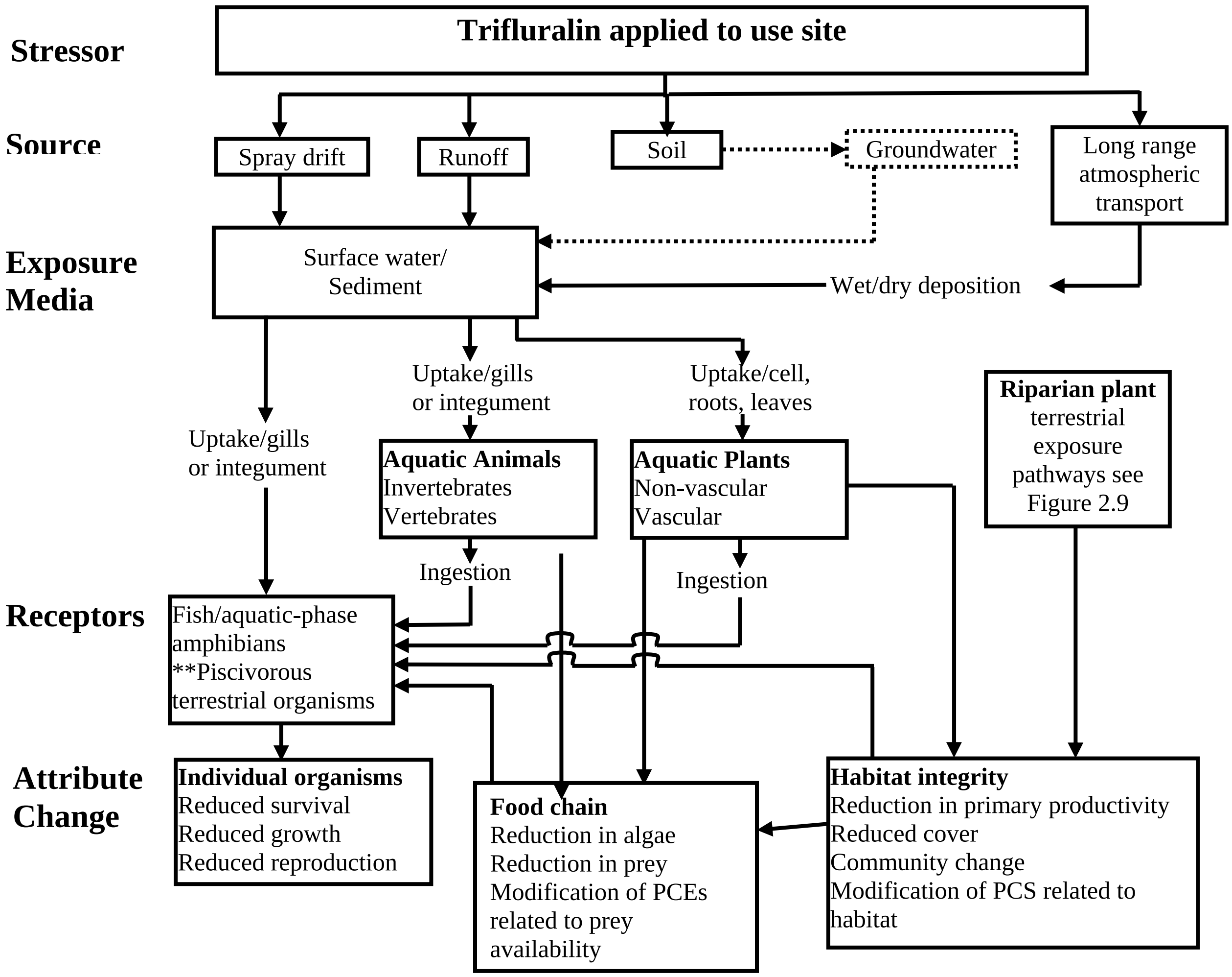


Figure 2.7 Conceptual Model for Trifluralin Effects on the Assessed Species and Their Critical Habitat in the Aquatic Environment

** Route exposure includes only ingestion of aquatic fish and invertebrates; piscivorous terrestrial organisms may include mammals, birds, reptiles, terrestrial-phase amphibians

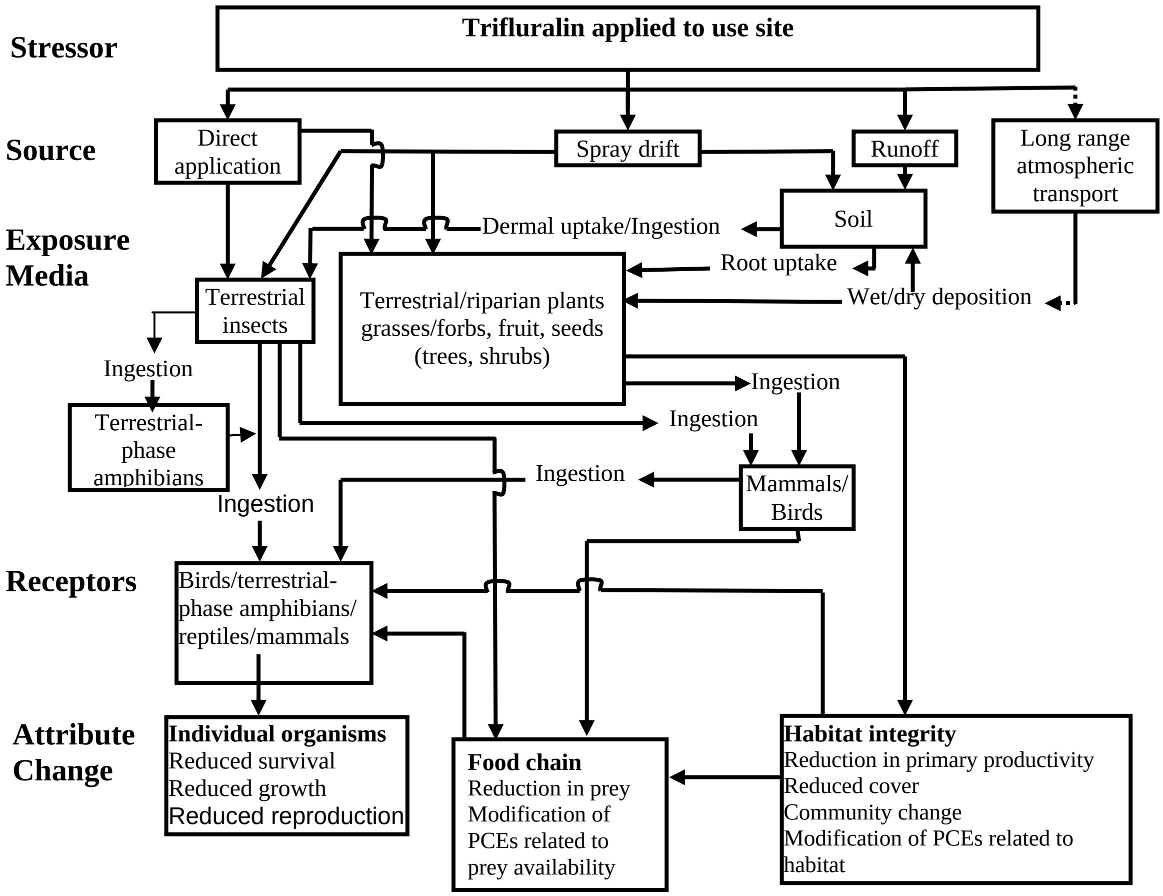


Figure 2.8 Conceptual Model for Trifluralin Effects on the Assessed Species and Their Critical Habitat in the Terrestrial Environment

2.10 Analysis Plan

In order to address the risk hypothesis, the potential for direct and indirect effects to the CRLF, DS, SFGS, and SJKF prey items, and habitat is estimated based on a taxon-level approach. In the following sections, the use, environmental fate, and ecological effects of trifluralin 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 trifluralin is estimated using the probit dose-response slope and either the level of concern (discussed below) or actual calculated risk quotient value.

2.10.1 Measures to Evaluate the Risk Hypothesis and Conceptual Model

2.10.1.1 Measures of Exposure

The environmental fate properties of trifluralin along with available monitoring data indicate that runoff, spray drift, and long-range atmospheric transport are the principle potential transport mechanisms of trifluralin to the aquatic and terrestrial habitats of the CRLF, DS, SFGS, and SJKF. In this assessment, transport of trifluralin through runoff and spray drift is considered in deriving quantitative estimates of CRLF, DS, SFGS, and SJKF exposure to trifluralin, in addition to exposure of their prey and habitats. Assessment of long-range atmospheric transport is addressed qualitatively through a review of available monitoring data from California, but not explicitly modeled due to the lack of models that quantitatively predict far-field pesticide concentrations resulting from near-field loadings.

Measures of exposure are based on aquatic and terrestrial models that predict estimated environmental concentrations (EECs) of trifluralin 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 trifluralin that may occur in surface water bodies adjacent to application sites receiving trifluralin through runoff and spray drift. PRZM simulates pesticide application, movement and transformation on an agricultural field and the resultant pesticide loadings to a receiving water body via runoff, erosion and spray drift. EXAMS simulates the fate of the pesticide and resulting concentrations in the water body. The standard scenario used for ecological pesticide assessments assumes application to a 10-hectare agricultural field that drains into an adjacent 1-hectare water body, 2-meters deep (20,000 m^3 volume) with no outlet. PRZM/EXAMS was used to estimate screening-level exposure of aquatic organisms to trifluralin. 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, Oct. 9, 2008). This model incorporates the Kenega nomograph, as modified by Fletcher *et al.* (1994), which is based on a large set of actual field residue data. The upper limit values from the nomograph represented the 95th percentile of residue values from actual field measurements (Hoerger and Kenega, 1972). For the purposes of this assessment, upper-bound Kenaga nomogram estimates reported by T-REX are used for derivation of the EECs for the terrestrial-phase CRLF, SFGS, and SJKF and their potential prey.

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

For modeling purposes, direct exposures of the SJKF to trifluralin through contaminated food are estimated using the EECs for the large mammal (1000 g) which consumes all food categories. Dietary-based and dose-based exposures of potential prey (mammals and birds) are assessed using the small mammal (15 g) and small bird (20 g) which consumes short grass. The small bird (20g) 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 its prey items. Estimated exposures of terrestrial insects to trifluralin 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. Therefore, T-REX (version 1.4.1) has been refined to the T-HERPS model (v. 1.0), which allows for an estimation of food intake for poikilotherms using the same basic procedure as T-REX to estimate avian food intake.

Because there is evidence of the potential for bioaccumulation of trifluralin in aquatic organisms, an additional exposure pathway that will be considered in this assessment is the consumption of contaminated fish or aquatic invertebrates that have bioaccumulated trifluralin dissolved in water and their aquatic diet. The potential risk from this pathway will be evaluated and discussed further in the "Risk Description" section of the document. Multiple lines of evidence will be used to evaluate bioaccumulation potential, including measured bioconcentration factors (BCF), bioaccumulation factors (BAF) and a food web bioaccumulation model (Kow-Based Aquatic Bioaccumulation Model or KABAM ver 1.0). The bioaccumulation assessment relies on predicted water and sediment concentrations from PRZM/EXAMS to estimate concentrations of trifluralin in aquatic organisms. These estimated tissue concentrations will be compared to toxicity values for various taxonomic groups that may eat aquatic organisms in order to evaluate potential risk.

The current version of KABAM estimates risks only to terrestrial birds and mammals consuming aquatic prey. For this assessment, risks to the CRLF and the SFGS were assessed using KABAM since birds can be used as a surrogate for both terrestrial-phase amphibians and reptiles. Risks to the DS from the bioaccumulation pathway are not assessed with KABAM as this species does not have a terrestrial life phase. Risks to the SJKF from the bioaccumulation pathway are not assessed with KABAM as this species does not consume aquatic prey.

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.

AgDRIFT is the spray drift model used to assess exposures of the assessed species and their prey to trifluralin deposited on terrestrial habitats by spray drift. In addition to the buffered area from the spray drift analysis, the downstream extent of trifluralin that exceeds the LOC for the effects determination is also considered.

2.10.1.2 Measures of Effect

Data identified in **Section 2.8** are used as measures of effect for direct and indirect effects to the CRLF, DS, SFGS, and SJKF. 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 and the SFGS makes the assumption that toxicity of trifluralin 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 (again, this also applies to potential prey items).

The acute measures of effect used for animals in this screening level assessment are the LD_{50}, LC_{50}, EC_{50}, and IC_{50}. LD stands for "Lethal Dose", and LD_{50} is the dose of material that is estimated to cause the death of 50% of the test organisms. LC stands for “Lethal Concentration” and LC_{50} is the concentration of a chemical that is estimated to kill 50% of the test organisms. EC stands for “Effective Concentration” and the EC_{50} is the concentration of a chemical that is estimated to produce a specific effect (*e.g.*, immobility) in 50% of the test organisms. IC_{50} stands for “Inhibition Concentration” and is the concentration estimated to cause a 50% reduction in a response variable (*e.g.,* plant height) relative to the control. 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.*, EC_{25} for terrestrial plants and EC_{50} for aquatic plants).

It is important to note that 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.

2.10.1.3 Integration of Exposure and Effects

Risk characterization is the integration of exposure and ecological effects characterization to determine the potential ecological risk from agricultural and non-agricultural uses of trifluralin, and the likelihood of direct and indirect effects to CRLF, DS, SFGS, and SJKF in aquatic and terrestrial habitats, as appropriate. 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 trifluralin 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 J)**.

For this endangered species assessment, listed species LOCs are used for comparing RQ values for acute and chronic exposures of trifluralin directly to the CRLF, DS, SFGS, and SJKF. If estimated exposures directly to the assessed species of trifluralin 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 trifluralin 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".

2.10.1.4 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 trifluralin 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.

2.10.2 Data Gaps

Uncertainties, Limitations, and Assumptions

- There are no aerobic aquatic degradation data for trifluralin. A default aerobic aquatic degradation half-life was calculated as twice the aerobic soil metabolism half-life for PRZM/EXAMS modeling.

- There are no anaerobic aquatic degradation data for trifluralin. A default anaerobic aquatic degradation half-life was calculated as 0.5 times the aerobic aquatic metabolism rate for PRZM/EXAMS modeling.

- There are no estuarine/marine fish data for evaluation of acute toxicity of trifluralin. For acute estuarine/marine fish, data from ethafluralin (a herbicide in the same chemical class with the same mode of action as trifluralin) are used.

- There are no estuarine/marine invertebrate data for evaluation of chronic toxicity of trifluralin. For chronic toxicity, the acute-to-chronic ratio (ACR) calculations are conducted using daphnid toxicity data and applying to the acute grass shrimp data.

- There are no acute or chronic data for benthic invertebrates. This is of concern for trifluralin as it sorbs strongly to sediment and pore water concentrations are expected to be higher than in the water column.

3. Exposure Assessment

Trifluralin is formulated as an emulsifiable concentrate and as a granular. Application equipment and methods include aircraft, groundspray, chemigation, soil broadcast, sprinkler irrigation, directed spray, and granular applications. Risks from ground boom and aerial applications are considered in this assessment because they tend to result in the highest off-target levels of trifluralin due to generally higher spray drift levels. Ground boom and aerial modes of application tend to use lower volumes in finer sprays than applications coincident with sprayers and spreaders and thus have a higher potential for off-target movement via spray drift.

3.1 Application Rates, Dates and Intervals

Trifluralin labels may be categorized into two types: labels for manufacturing uses (including technical grade trifluralin and its formulated products) and end-use products. While technical products, which contain trifluralin of high purity, are not used directly in the environment, they are used to make formulated products which can be applied in specific areas to control pre- and post-emergent broadleaf and monocot weeds. The formulated product labels legally limit trifluralin's use to only those sites that are specified on the labels. These products, their use sites, and rates are discussed in **Section 2.4.4**.

Crop-specific management practices for all of the assessed uses of trifluralin were used for modeling (**Table 3.1**). These parameters included application rates, number of applications per year, application intervals, and the first application date for each crop.

Maximum application rates and numbers of application as specified by current labels for each crop were modeled in PRZM/EXAMS. In instances when maximum annual application rates and/or number of applications were not specified in the LUIS Report, EFED assumed representative rates and application intervals based on management practices and use rates on similar crops. For some crops (*e.g.*, avocado and orchard), it was assumed that a maximum of 3 applications of 4 lbs trifluralin at a minimum of 60 day intervals is applied. This application rate is comparable to numerous annual maximum application rates for granular nursery and orchard uses. For other crops (*e.g.*, corn, cotton, and wheat), it was inferred that a single application per crop cycle was intended as a specific time in the growing cycle of the crop. For more details on application rates for assessed trifluralin uses, see **Table 2.3**.

The date of first application was developed based on several sources of information including data provided by BEAD, a summary of individual applications from the CDPR PUR data, and Integrated Management Pest Center Crop Profiles maintained by the USDA. If an application pattern emerged and a typical application date from a crop scenario could be established, then that date was employed. In general, peak trifluralin use occurred during the months February through April. If no application date pattern emerged from the data, a March 1st application date was selected to result in a conservative EEC that would result from increased rainfall experienced in California during the winter months. More detail on the crop profiles and the previous assessments may be found at: http://www.wrpmc.ucdavis.edu/Ca/CaCrops /calendar.html and http://www.ipmcenters.org/cropprofiles/.

Table 3.1 Crop Specific Application Input Data for PRZM/EXAMS for Trifluralin Uses in California					
Crop Scenario (Application Date)	**Crops/Uses**	**Method**	**Soil Incorp-oration (Y/N)[1]**	**Max Application Rate kg/ha (Min Interval)**	**1st App. Date (month/day)**
Orchard Uses					
CA Almond _WirrigSTD	Almond, filbert, macadamia, pecan, pistachio, tree nuts, walnut (English/black)	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
		Aerial Ground	Y	2 app @ 2.24 kg/ha (NA)	
CA Avocado RLF_V2	Avocado	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
CA Citrus _WirrigSTD 03/01	Fruits unspecified, grapefruit, lemon, orange, tangerine, tangelo	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
		Arial Ground	Y	1 app @ 2.24 kg/ha (NA)	
CA Fruit _WirrigSTD	Apple, apricot, cherry, fig, nectarine, peach, pear, plum, pomegranate, prune	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
		Aerial Ground	Y	1 app @ 2.24 kg/ha (NA)	
CA Grapes _WirrigSTD (Grapes and Berries)	Blackberry, blueberry, boysenberry, currant, dewberry, elderberry, gooseberry, grape, kiwi fruit, loganberry, mulberry, raspberry (black, red)	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
CA Grapes _WirrigSTD	Grape	Aerial Ground	Y	1 app @ 2.24 kg/ha (NA)	03/01
CA OliveRLF_V2	Olive	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
Agricultural Uses					
CA Alfalfa _WirrigOP	Alfalfa, clover	Aerial Ground (EC)	N	1 app @ 2.24 kg/ha (NA)	15/03
		Aerial Ground (Granular)	Y	2app @ 2.24 kg/ha (NA)	

Table 3.1 Crop Specific Application Input Data for PRZM/EXAMS for Trifluralin Uses in California					
Crop Scenario (Application Date)	**Crops/Uses**	**Method**	**Soil Incorp-oration (Y/N)[1]**	**Max Application Rate kg/ha (Min Interval)**	**1st App. Date (month/day)**
CA Cole Crop RLF_V2	Broccoli, broccoli rabbinni, Brussels spouts, cabbage, canola/rape, cauliflower, collards, crambe, kale, mustard,	Aerial Ground	Y	1 app @ 1.12 kg/ha (NA)	03/01
CA CornOP	Corn field, kenaf, sunflower	Aerial Ground	Y	1 app @ 1.12 kg/ha (NA)	09/05
CACotton_WirrigSTD	Cotton	Aerial Ground	Y	1 app @ 2.24 kg/ha (NA)	03/01
CA LettuceSTD	Chicory, endive	Aerial Ground	Y	1 app @ 1.12 kg/ha (NA)	04/01
CA Melons RLF_V2	Cucumber, melon, watermelon	Ground	Y	1 app @ 1.12 kg/ha (NA)	11/20
CA Onion	Onion (dry bulb) Radish	Aerial Ground	Y	1 app @ 0.9 kg/ha (NA)	11/01
CA Potato RLF_V2 (09/05)	White/Irish potato¸ turnip	Ground	Y	1 app @ 1.12 kg/ha (NA)	09/05
CA Rangeland Hay RLF_V2	Bermuda grass for seed, uncultivated agriculture, Un-cultivatedareas/soils	Ground	N	1 app @ 2.24 kg/ha (NA)	03/01
CA Row Crop RLF_V2	Asparagus, bean (castor) Bean succulent, bean (lima), bean (mung), bean succulent, bean (snap), carrot, celery, guar lentil, peas(southern), peas (succulent), pepper, sugar cane	Aerial Ground	Y	1 app @ 2.24 kg/ha (NA)	05/15
CA Sugar Beet _WirrigSTD	Sugar beet	Aerial Ground	Y	1 app @ 0.8 kg/ha (NA)	05/15
CA Tomato_WirrigSTD	Okra, tomato	Ground	Y	1 app @ 1.12kg/ha (NA)	04/20

Table 3.1 Crop Specific Application Input Data for PRZM/EXAMS for Trifluralin Uses in California					
Crop Scenario (Application Date)	**Crops/Uses**	**Method**	**Soil Incorp-oration (Y/N)[1]**	**Max Application Rate kg/ha (Min Interval)**	**1st App. Date (month/day)**
CA Wheat RLF_V2	Barley, flax, hops, safflower, sorghum, unspecified grains, wheat	Aerial Ground	Y	1 app @ 1.12 kg/ha (NA)	12/15
Non-agricultural Uses					
CA Nursery STD_V2	Banana, greenhouse-in use ornamental shade trees, ornamental ground cover, ornamental herbaceous plants, ornamental lawns and turf, ornamental non-flowering plants, ornamental woody shrubs and vines residential lawns	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
CA ResidentialRLF.txt	Ornamental and/or shade trees, ornamental ground cover, ornamental herbaceous plants, ornamental lawns and turf, ornamental non-flowering plants, ornamental woody shrubs and vines residential lawns	Ground	N	2 app @ 1.68 kg/ha (56)	03/01
CA Rights-of-way RLF_V2	Pre-paving, private roads/sidewalks, non-agriculture rights-of-way, fencerows, hedgerows	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
CA TurfRLF	Golf course turf, recreational, commercial/industrial lawns	Ground	N	2 app @ 1.68 kg/ha (56)	03/01
CA ForestryRLF	Christmas tree plantations Cottonwood (forest/shelterbelt), poplar	Ground	N	3 app @ 4.48 kg/ha (60)	03/01
		Aerial Ground	Y	1 app @ 2.24 kg/ha (NA)	
[1] Y – Soil incorporation required within 24 hours, N soil incorporation is greater than 24 hours or is not required.					

3.2 Aquatic Exposure Assessment

3.2.1 Modeling Approach

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

Crop specific management practices for all of the assessed uses of trifluralin were used for modeling. These parameters included application rates, number of applications per year, application intervals, and the first application date for each crop. The date of first application was developed based on several sources including data provided by EPA/OPP/BEAD LUIS Report, a summary of individual applications from the CDPR PUR data, and Crop Profiles maintained by the USDA.

3.2.2 Post-processing of PRZM/EXAMS Outputs to Develop EECs for Residential and Rights-of-ways

The LUIS Report lists trifluralin uses that are modeled using the rights-of-way and residential RLF scenarios. Rights-of-way uses include fencerows, hedgerows, paved areas, and private roads/sidewalks applications. Residential uses include ornamental and/or shade trees, ornamental ground cover, ornamental herbaceous plants, ornamental lawns and turf, ornamental non-flowering plants, ornamental woody shrubs and vines applications. Both these scenarios contain areas with impervious (*i.e.,* cement, asphalt, metal surfaces) and pervious surfaces. It is assumed that trifluralin will be applied to the pervious surfaces where weeds are expected to grow. It is also assumed that trifluralin is not applied to impervious surfaces in rights-of way, but that there is a 1% incidental spray onto impervious surfaces surrounding rights-of-way.

In a standard PRZM scenario, it is assumed that an entire 10-hectare field is composed only of the identified crop and that the field has uniform surface properties throughout the field. In the rights-of-way and residential scenarios, this is not a reasonable assumption because rights-of-way and residential areas generally contain both impervious and pervious surfaces. Since the two surfaces have different properties (especially different curve numbers influencing the runoff from the surfaces) and different amounts of trifluralin applied, the standard approach for deriving aquatic EECs is revised using the following approach:

1) Aquatic EECs are derived for the pervious portion of the rights-of-way, using the maximum use rate of trifluralin on the CA rights-of-way and CA residential scenarios. At this point, it is assumed that 100% of both the rights-of-way and residential scenarios are composed of pervious surface. Specific inputs for this modeling are defined below.
2) Aquatic EECs are derived for the impervious portion of the rights-of-way using 1% for liquid formulation of the maximum use rate of trifluralin on the CA impervious and CA Residential scenarios. At this point, it is assumed that 100% of the rights-of-way and residential scenarios are composed of impervious surface.
3) The daily aquatic EECs (contained in the PRZM/EXAMS output file with the suffix "TS") are input into a Microsoft® Excel® worksheet.
4) Daily aquatic EECs for the impervious surface are multiplied by 50%. Daily aquatic EECs for the pervious surface are multiplied by 50%. The resulting EECs for impervious and pervious surfaces are added together to get an adjusted EEC for each day of the 30-year simulation period (**Equation 3.1**).

Equation 3.1

Revised EEC = (*impervious EEC* * 50%) + (*pervious EEC* * 50%)

5) Rolling averages for the relevant durations of exposure (21-day and 60-day averages) are calculated. The 1-in-10 year peak, 21-day, and 60-day values are used to define the acute and chronic EECs for the aquatic habitat.

In this modeling approach, it is assumed that both the rights-of-way and residential scenarios are composed of equal parts pervious and impervious surfaces (*i.e.*, in step 4, the EECs of both surfaces are multiplied by 50%). However, in reality, it is likely that rights-of-way and residential areas contain different ratios of the two surfaces. In general, incorporation of impervious surfaces into the exposure assessment results in increasing runoff volume in the watershed, which tends to reduce overall pesticide exposure (when assuming 1% overspray to the impervious surface).

3.2.3 Model Inputs

PRZM scenarios used to model aquatic exposures resulting from applications of specific uses are identified in **Table 3.1.** In cases where a scenario does not exist for a specific use, it is necessary to assign a surrogate scenario. The surrogates are assigned based on the same or similar families or similar cultural practices and to be most representative of the use being considered.

In cases where the LUIS Report presents numerous possible variations of application rates, application methods, and formulations for a modeled crop scenario, EFED modeled the rates, methods and formulations that would likely result in the highest EECs in PRZM/EXAMS. Aerial emulsifiable applications generally produce higher EECs than aerial granular applications; therefore, aerial emulsifiable concentration applications producing higher EECs were modeled.

The appropriate chemical-specific PRZM and EXAMS input parameters are selected from reviewed environmental fate data submitted by the registrant in **Table 3.2.** These input parameters are in accordance with EFED water model PRZM/EXAMS input parameter guidance (U.S. EPA 2002). The input parameters employed are similar to those used in the 1996 trifluralin RED (U.S. EPA, 1996).

In PRZM, the method of pesticide application is defined by Chemical Application Method (CAM). CAM 1 was used to represent surface applied broadcast when soil incorporation was not required within 24 hours by the label. When the label required incorporation within 24 hours, CAM 4 was employed to represent uniform soil incorporation to the default depth of 5 cm.

Trifluralin is a volatile compound; therefore, PRZM/EXAMs manual default input parameters DAIR (vapor phase diffusion coefficient) of 4300 cm^2/day and enthalpy of vaporization coefficient of 20 kcal/mol were inputted into PRZM/EXAMs to account for volatilization from soil.

Table 3.2 Summary of PRZM/EXAMS Environmental Fate Data Used for Aquatic Exposure Inputs for Trifluralin Endangered Species Assessment

Model Parameter	Value	Comment	Data Source
Maximum Single Application Rate	Dependant on Scenario	See **Table 2.3** for values for each scenario	Product labels
Hydrolysis	Stable at pH 5, pH 7, pH 9	Stable at pH 5, pH 7, pH 9 at 25 ºC	MRID 00131135
Aqueous Photolysis	$T_{1/2}$ = 0.371 days		MRID 40560101
Molecular Weight	335.28 g/mol		Product Chemistry
Solubility in Water	0.3 mg/L		Product Chemistry
Vapor Pressure	1.10E-4 Torr		Product Chemistry
Henry's Law Constant	1.62E-4 atm m^3/mole		RED
Aerobic Soil Metabolism	$T_{1/2}$ = 219 days	90% upper confidence bound on the mean metabolism half-lives, 189, 201, 116 days[1]	MRID 41240501
Anaerobic Aquatic Metabolism	$T_{1/2}$ = 29.5 days	Anaerobic aquatic metabolism 59 days (0.5 x anaerobic soil metabolism rate)[1]	N/A
Aerobic Aquatic Metabolism	$T_{1/2}$ = 438 days	Aerobic aquatic metabolism 438 days (2x aerobic soil metabolism)	EFED Guidance (Feb 28, 2002)
K_{oc}	8,757.9	Mean of 4 Koc values (6,413.3, 6,748.2, 8,457.2, and 13,413 L/kg)[1]	MRID 40673501
Application Method (CAM)	CAM 1 used when not incorporated; CAM 4 employed when incorporated	Soil incorporation depth 5cm when incorporated	Product labels

Table 3.2 Summary of PRZM/EXAMS Environmental Fate Data Used for Aquatic Exposure Inputs for Trifluralin Endangered Species Assessment			
Model Parameter	**Value**	**Comment**	**Data Source**
Application Efficiency	0.99 ground spray 0.95 aerial spray 1.00 granular	Spray Drift Task Force (SDTF) application efficiency data	SDTF
Spray Drift Fraction	0.01 ground spray 0.05 aerial spray 0.0 granular	SDTF application efficiency data	SDTF
DIAR (vapor diffusion phase coefficient)	4300 $cm^{2/day}$		PRZM/EXAMs manual
Enthalpy	20 kcal/mol		PRZM/EXAMs manual

3.2.4 Results

The aquatic EECs for the various scenarios and application practices are listed in **Table 3.3.** The peak EECs range from 0.0002 µg/L (residential) to 6.53 µg/L (EC nursery), with a median of 1.18 µg/L. The 21-day EECs range from 0.0001 µg/L (residential) to 1.83 µg/L (nursery), with a median of 0.32 µg/L. The predicted 60-day average EECs range from 0.00005 µg/L (residential) to 0.87 µg/L (nursery), with a median of 0.18 µg/L for all scenarios modeled.

In general, the 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. However, trifluralin concentrations do not increase across the 30-year time series probably due to its susceptibility to direct aqueous photolysis (half-life = 8.9 hours) and to volatilization (Henry's Law constant = 1.6 × 10-4atm.m3/mol) which should limit its persistence in the water column of well mixed, surface water. An example PRZM/EXAMS output file is located in **Appendix D**.

Table 3.3 Aquatic EECs (µg/L) for Trifluralin Uses in California					
Crop Scenario/ Formulation	**Application Method**	**Soil Incorporation**	**Peak EEC µg/L**	**21 day EEC µg/L**	**60 day EEC µg/L**
Orchard Uses					
CA Almond (tree nuts)					
Granular	Ground	N	2.23	0.78	0.72
EC[1]	Aerial	Y	5.65	1.31	0.60
	Ground	Y	1.23	0.32	0.18
CA Avocado					
Granular	Ground	N	5.81	1.45	0.73
CA Citrus					
Granular	Ground	N	0.74	0.20	0.14
EC[1]	Aerial	Y	5.48	1.28	0.54
	Ground	Y	1.10	0.26	0.11
CA Fruit					
Granular	Ground	N	2.52	0.75	0.40
EC[1]	Aerial	Y	5.51	1.30	0.55

Table 3.3 Aquatic EECs (µg/L) for Trifluralin Uses in California					
Crop Scenario/ Formulation	**Application Method**	**Soil Incorporation**	**Peak EEC µg/L**	**21 day EEC µg/L**	**60 day EEC µg/L**
	Ground	Y	1.12	0.30	0.13
CA Grape (grapes and berries)					
Granular	Ground	N	4.00	1.18	0.62
CA Grape					
EC[1]	Aerial	Y	5.50	1.29	0.54
	Ground	Y	1.18	0.28	0.12
CA Olive					
Granular	Ground	N	4.45	1.35	0.77
Agricultural Uses - Non-Orchard					
CA Alfalfa					
EC	Aerial	N	5.14	1.32	0.58
	Ground	N	1.12	0.32	0.17
Granular	Aerial	Y	0.32	0.09	0.07
	Ground	Y	0.32	0.09	0.07
CA Cole					
EC	Aerial	Y	2.79	0.65	0.29
	Ground	Y	0.59	0.19	0.10
CA Corn					
EC	Aerial	Y	2.74	0.62	0.26
	Ground	Y	0.55	0.13	0.07
CA Cotton					
EC	Aerial	Y	5.55	1.46	0.65
	Ground	Y	1.48	0.47	0.24
CA Melon					
EC	Ground	Y	0.73	0.27	0.13
CA Lettuce					
EC	Aerial	Y	2.77	0.64	0.27
	Ground	Y	0.58	0.17	0.08
CA Onion					
EC	Aerial	Y	1.96	0.48	0.21
	Ground	Y	0.39	0.10	0.05
CA Potato					
EC	Ground	Y	0.55	0.12	0.05
CA Rangeland					
Granular	Ground	N	0.68	0.23	0.12
CA Tomato					
EC	Ground	Y	0.56	0.13	0.06
CA Row Crop					
EC	Aerial	Y	5.49	1.09	0.45
	Ground	Y	1.01	0.22	0.09
CA Sugar Beet					
EC	Aerial	Y	1.96	0.42	0.17
	Ground	Y	0.39	0.08	0.03
CA Wheat					
EC	Aerial	Y	2.77	0.78	0.29
	Ground	Y	0.57	0.18	0.11
Non-agricultural Uses					
CA Nursery					
Granular	Ground	N	6.55	1.80	0.86

Table 3.3 Aquatic EECs (µg/L) for Trifluralin Uses in California					
Crop Scenario/ Formulation	**Application Method**	**Soil Incorporation**	**Peak EEC µg/L**	**21 day EEC µg/L**	**60 day EEC µg/L**
EC	Ground	N	6.53	1.83	0.87
CA Residential[2]					
Granular	Ground	N	0.0002	0.0001	0.00005
CA Right-of-way[3]					
Granular	Ground	N	0.002	0.0007	0.0005
CA Turf					
Granular	Ground	N	0.16	0.05	0.02
CA Forestry					
Granular	Ground	N	2.25	0.77	0.39
EC	Aerial	Y	5.50	1.39	0.63
	Ground	Y	1.17	0.35	0.19

[1] Due to the lack of spray drift aerial granular EECs are generally lower than aerial emulsifiable concentration EECs; therefore, the higher, and more conservative aerial emulsifiable concentration applications were modeled for this scenario.
[2] Modeled with 1% overspray using Impervious Scenario, post-processed with Residential scenario with 50/50 ratio. See **Section 3.2.1** for explanation of initial application date selection.
[3] Modeled with 1% overspray using Impervious Scenario, post-processed with Right-of-Way scenario with 50/50 ratio.

3.2.5 Existing Monitoring Data

A critical step in the process of characterizing predicted EECs is comparing the modeled estimates with available surface water monitoring data. An evaluation of the surface water monitoring data was conducted to assess the occurrence of trifluralin in California surface and ground waters. Most of this data, however, is non-targeted (*i.e.*, study was not specifically designed to capture trifluralin concentrations in high-use areas). Typically, sampling frequencies employed in these monitoring studies are insufficient to document peak exposure values. This, coupled with the fact that these data are not temporally or spatially correlated with pesticide applications, limits the utility of these data for estimating exposure concentrations for risk assessment purposes. These monitoring data are characterized in terms of general statistics including number of samples, frequency of detection, maximum concentration, and mean value of all detections, where that level of detail is available.

3.2.5.1 Federal and Cal EPA Monitoring Data

Data from the CDPR surface water monitoring database website for trifluralin occurrence were obtained on Jul 15, 2009 (http://www.cdpr.ca.gov/docs/emon/surfwtr/surfcont.htm). A total of 3,915 surface water samples were analyzed for trifluralin spanning a period from 1991 to 2006. Of these, a total of 600 samples detected trifluralin (detection frequency of 15%). The two highest concentrations detected were 1.74 and 1.5 µg/L from Butte County on 01/16/01. The maximum concentration of trifluralin reported by the CDPR surface water database (1.74 µg/L) is roughly 3.75 times lower than the highest peak model-estimated environmental concentration.

No trifluralin data were available from the EPA STORET database for the state of California (http://www.epa.gov/storet/). USGS's National Water-Quality Assessment Program has not collected groundwater or surface samples for trifluralin in California (http://infotrek.er.usgs.gov/traverse/f?p=NAWQA:HOME:3249496365037407).

3.2.5.2 Open Literature Monitoring Data

Water samples collected from DS habitat in Suisun Bay during the spring and summer of 2000 detected trifluralin in 32 of 54 samples where 44 ng/L was the highest measured concentration (Kuivila, 2002).

A study was conducted during the first flush of suspended sediments into Suisun Bay at Mallard Island in December 1995 (Bergamaschi and others 2001). Trifluralin was detected on 4 days of the 16 day study with values ranging from 0.2 to1.3 ng/g per dry weight of sediment.

The transport of suspended sediments may be an important mechanism for the movement of trifluralin into estuaries. Based on sampling of suspended sediments during February 1992 in the San Joaquin River (at Vernalis, California), trifluralin concentrations in ranged from 4.6 to 31.3 ng/L in dry weight of sediment. (Bergamaschi *et al*. 1997).

In a study of pesticide inputs to Yolo Bypass in 2004 and 2005 trifluralin was detected in surface water at 17 of 44 sites with values ranging from 4.1 to 66.4 ng/L (Smalling and others 2005). In the same study, trifluralin was detected in sediment at 4 of 6 sites with a high concentration of 24 µg/kg.

3.3 Long Range Transport Exposure Assessment

3.3.1 Background

Exposure to trifluralin through long range atmospheric transport may also occur. Prevailing winds blow across the Central Valley eastward to the Sierra Nevada Mountains, transporting airborne industrial and agricultural pollutants into the Sierra Nevada ecosystems (Fellers *et al.*, 2004, LeNoir *et al.*, 1999, and McConnell *et al.*, 1998). Trifluralin can be present in air or precipitation due to spray drift, volatilization and/or wind erosion of soil containing residues (Unsworth *et al*, 1999). Precipitation and dry particulate matter can contribute to trifluralin deposits in aquatic systems (LeNoir *et al*, 1999). Therefore, deposition of trifluralin could potentially be transported to the habitats of CRLF, DS, SFGS, and SJKF.

No approved model currently exists for estimating atmospheric transport of pesticides and the resulting exposures to organisms in areas receiving pesticide deposition. Therefore, potential mechanisms of atmospheric transport for trifluralin such as volatilization, wind erosion of soil and spray drift can only be discussed qualitatively. Given the presence of trifluralin in air and precipitation in California monitoring data, exposure to CRLF, DS, SFGS, and SJKF habitats through atmospheric transport of trifluralin cannot be precluded. Although it is not possible to quantify trifluralin exposure via long-range transport, the amount of trifluralin deposition into

CRLF, DS, SFGS, and SJKF habitats based on measured trifluralin concentrations in rain samples are considered in combination with California precipitation data.

3.3.2 Qualitative Discussion of Potential Transport Mechanisms for Long-Range Transport of Trifluralin

A number of long-range mechanisms transport trifluralin from an initial application site to the atmosphere via wet or dry deposition of trifluralin to distant locales. The mechanisms are: 1) volatilization from soil and plant surfaces in treated areas, 2) wind erosion of soil containing sorbed trifluralin, and 3) spray drift of trifluralin during application.

Factors influencing volatilization of trifluralin from a treated area are: vapor pressure, adsorption to soil, incorporation depth, and Henry's law constant. The vapor pressure (1.1×10^{-4} torr) and reported Henry's law constant of 1.6×10^{-4} atm-m^3/mol indicate that trifluralin will volatilize from soil and water.

Soil incorporation of trifluralin affects post-application volatilization. A laboratory experiment that placed trifluralin 1.27 cm below the surface reduced vapor loss by a factor of about 25 times (Bardsley *et al*, 1968). Another laboratory study that incorporated trifluralin into the top 10 cm of soil recorded vapor loss rate at 51.7 g/ha/d in the first 24 hours, while surface applied losses occurred at 4,000 g/ha/d (Spencer and Cliath, 1974). A field study indicated that trifluralin incorporated at a depth of 5 cm volatilized at a maximum rate of 3 g/ha/d during the first 4 to 6 hours after application (Grover and others, 1988). The volatilization rate of trifluralin applied to the surface of agricultural sites was 70 g/ha/d (Majewski et al, 1993)

3.3.3 Long-Range Air and Precipitation Monitoring Data

Trifluralin has been extensively used in wheat production since the 1960s and has been detected in air, rain and snow in California (Majewski and Capel, 1995). Available air and precipitation California monitoring data for trifluralin are reported in **Table 3.4**.

Table 3.4 Detections of Trifluralin in Air, Precipitation, and Snow Samples Taken in California

Location	Year	Sample type	Maximum Conc.[1]	Detection frequency	Source
CA	1970s-1990s	Air	0.063	NA	Reported in Majewski and Capel, 1995
Sequoia National Park, CA	1996	Air	0.00064	100%	LeNoir *et al.* 1999
Sacramento, CA (Franklin Field Airport)	1996-1997	Air	0.014	71.4%	Majewski and Baston 2002
Sacramento, CA (Sacramento Metropolitan Area)	1996-1997	Air	0.010	67.6%	Majewski and Baston 2002
Sacramento, CA (Sacramento International Airport)	1996-1997	Air	0.019	81.5%	Majewski and Baston 2002
Sequoia National Park, CA (Ash Mountain)	1995-1996	Rain	0.0012	NA	McConnell *et al.* 1998
San Joaquin River Basin, CA	2001	Rain	0.039	78%	Zamora et al. 2003

Table 3.4 Detections of Trifluralin in Air, Precipitation, and Snow Samples Taken in California					
Location	**Year**	**Sample type**	**Maximum Conc.[1]**	**Detection frequency**	**Source**
CA	1970s-1990s	Rain	0.97	NA	Reported in Majewski and Capel, 1995
Sequoia National Park, CA (Lower Kaweah)	1995-1996	Snow	0.041	NA	McConnell *et al.* 1998
[1] For Air, μg/m^3; for rain, snow and μg/L					

Detected concentrations of trifluralin in air, snow and rain vary depending on proximity to use areas and timing of applications. In air, trifluralin has been detected at concentrations up to 0.063 μg/m^3. Measured concentrations of trifluralin in rain in California have been measured up to 0.97 μg/L.

The extent to which trifluralin will be deposited from the air to the habitat of the CRLF, DS, SFGS, and SJKF is unknown. In an attempt to estimate the amount of trifluralin deposited into aquatic and terrestrial habitats, trifluralin concentrations measured in rain samples taken in California are considered below in combination with California specific precipitation data and runoff estimates from PRZM.

Air monitoring data from the September, 2007 Environmental Justice Pilot Study conducted by the California EPA in Parlier, a small agricultural community located in California's San Joaquin Valley was evaluated. Trifluralin was detected in 24% (112 samples) of 468 air monitoring samples collected within 5 miles of Parlier (http://www.cdpr.ca.gov/docs/envjust/pilot_proj/ interim/narrative.pdf).

Trifluralin has been measured in arctic air, seawater, and freshwater sediments (Canadian Arctic Contaminants Program, 2006). Trifluralin was observed in air at three Arctic monitoring stations, Tagish, Alert and Dunai at concentrations up to 2.92; 0.64 and 0.13 pg/m^3, respectively.

Trifluralin is an herbicide whose occurrence and concentrations in air have been found to correlate with local use. The highest concentration of trifluralin occurred in May and June at several locations throughout Saskatchewan, Canada (Grover *et al.*, 1981). Air concentrations increased slightly during late October and November, which corresponds with a second application season. Grover *et al.* observed during that air concentrations of trifluralin decreased during dry periods and increased after rain events. Presumably this was due to the absorption of trifluralin from the remoistened soil, which resulted in an increase in volatilization.

3.3.4 Deposition Data

In a 3.5 year study (from 2001-2004) in the central San Joaquin Valley, wet deposition of pesticides, including trifluralin, were monitored at 6 sites, including some with agricultural and urban landcovers. Trifluralin was detected in 78% of rain samples (n=137), with mean and maximum concentrations of 0.010 and 0.039 μg/L, respectively (Majewski *et al*. 2006).

3.3.5 Monitoring Data from Lakes Assumed to Only Receive Atmospheric Deposition of Trifluralin

Studies are available involving monitoring of trifluralin concentrations in California lakes which are removed from agricultural areas and are presumed to receive inputs of trifluralin from atmospheric deposition only. Two 1997 studies (Fellers *et al.* 2004; LeNoir *et al.* 1999) measured trifluralin concentrations in lake water in Kings Canyon and Sequoia National Parks (located in the Sierra Nevada Mountains in California). Fellers *et al*. (2004) reported a concentration of 0.0025 µg/L, and LeNoir *et al.* (1999) reported a maximum concentration of 0.11 µg/L in lake water. The authors attributed these detections to atmospheric deposition from dry deposition and/or gas exchange from air samples of trifluralin originating from agricultural sites located in California's Central Valley, which is up wind of the lakes.

3.3.6 Modeling of Contributions of Wet Deposition to Aquatic and Terrestrial Habitats

In an attempt to estimate the amount of trifluralin deposited into aquatic and terrestrial habitats, trifluralin concentrations measured in rain samples taken in California were considered in combination with California specific precipitation data and runoff estimates from PRZM. Precipitation and runoff data associated with the PRZM scenarios used to model aquatic EECs were used to determine relevant 1-in-10 year peak runoff and rain events. The scenarios included were: CA almond, CA lettuce, CA wine grape, CA row crop, CA fruit, CA nursery, and CA onion. The corresponding meteorological data were from the following locations: Sacramento, Santa Maria, San Francisco, Monterey County, Fresno, San Diego, and Bakersfield, respectively.

To estimate concentrations of trifluralin in the aquatic habitat resulting from wet deposition, the daily PRZM-simulated volume of runoff from a 10 ha field is combined with an estimate of daily precipitation volumes over the 1 ha farm pond relevant to the EXAMS environment. This volume is multiplied by the maximum concentration of trifluralin in precipitation reported in monitoring data, which is 0.97 µg/L. The result is a daily mass load of trifluralin into the farm pond. This mass is then divided by the volume of water in the farm pond (2.0 x10^7 L) to achieve a daily estimate of trifluralin concentration in the farm pond, which represents the aquatic habitat (**Table 3.5**). From the daily values, the 1-in-10 year peak estimate of the concentration of trifluralin in the aquatic habitat is determined for each PRZM scenario (**Table 3.6**). Concentrations estimated using this approach are 1-2 orders of magnitude greater than those reported by Fellers et al. (2004) and LeNoir et al. (1999) in mountain lakes assumed to be receiving trifluralin loading only from atmospheric deposition. This difference in concentrations is reasonable since the mountain lakes where trifluralin was detected were spatially removed from trifluralin use areas; while the location where the maximum detected concentration of trifluralin was observed in precipitation was in close proximity to agricultural uses of trifluralin. There are several assumptions associated with this approach, including: 1) the concentration of trifluralin in the rain event is spatially and temporally homogeneous (e.g. constant over the 10 ha field and 1 ha pond for the entire rain event); 2) the entire mass of trifluralin contained in the precipitation runs off the pond or is deposited directly into the pond; 3) there is no degradation of trifluralin between the time it leaves the air and the time it reaches the pond.

Table 3.5 Input Parameters Relevant to Estimation of Trifluralin Load through Wet Deposition		
Parameter Description	**Value**	**Comment**
Area of PRZM field (ha)	10	-
Area of EXAMS pond (ha)	1	-
Volume of EXAMS farm pond (L)	20,000,000	Dimensions: 1 ha area x 2 m deep
Concentration of pesticide in precipitation (µg/L)	0.97	Reported in Majewski and Capel, 1995

Table 3.6 1-in-10 Year Peak Estimates of Trifluralin Concentrations in Aquatic and Terrestrial Habitats Resulting from Wet Deposition.		
Met Station	**Scenario(s)**	**Concentration in aquatic habitat (µg/L)[1]**
Sacramento	CA almond	0.181
Santa Maria	CA lettuce, CA cole crops, CA strawberry	0.195
San Francisco	CA wine grape	0.170
Monterey Co.	CA row crop	0.156
Fresno	CA fruit, CA tomato, CA melon	0.071
San Diego	CA nursery	0.131
Bakersfield	CA onion, CA potato	0.052

3.4 Aquatic Bioaccumulation Assessment

3.4.1 Empirical BCF Data

Trifluralin residues accumulated in a bluegill sunfish (*Lepomis macrochirus*) exposed to 0.0059 mg/L of trifluralin with maximum mean bioconcentration factor of 5,674x for whole fish tissues. The maximum mean concentration of total [^{14}C] residues that occurred at 28 days for the whole fish sample was 67.0 mg/L. Depuration occurred with 86.34-88.01% of the [^{14}C] eliminated from the fish tissues after 14 days of exposure to pesticide free water. The time to reach 90% of steady state for whole fish was 15.8 days (MRID 40673801). The accumulation and depuration of trifluralin in fish cannot be fully assessed because radioactive residues in fish tissues were incompletely characterized.

3.4.2 Fish Tissue Monitoring Data

The National Lake Fish Tissue Study (NLFTS) was the first survey of fish contamination in lakes and reservoirs in the 48 conterminous states based on a probability survey design (http://www.epa.gov/waterscience/fish/study/, Stahl *et al.* 2009, and Olsen *et al*. 2009). This study included the largest set (268) of persistent, bioaccumulative, and toxic (PBT) chemicals ever studied in predator and bottom-dwelling fish species; trifluralin was one of the chemicals evaluated. The USEPA implemented the study in cooperation with states, tribal nations, and other federal agencies, with field collection occurring at 500 lakes and reservoirs over a four-year period (2000–2003). The sampled lakes and reservoirs were selected using a spatially balanced unequal probability survey design from 270,761 lake objects in US EPA's River Reach File Version 3 (RF3).

There were a total of 33 composite fish samples in California that were evaluated for trifluralin. Of the 12 samples of bottom-dwelling fish (homogenized whole), six (50%) had detected levels of trifluralin (3.01, 16.4, 19.6, 22.3, 41.9, and 42.1 μg/kg-wet weight). Of the 21 samples of predator fish (filleted prior to homogenization), two (10%) had detected levels of trifluralin (3.21 and 3.33 μg/kg-wet weight). The Method Detection Limit (MDL) was 2.98 μg/kg-wet weight, and the Minimum Level of quantification (ML) was 10 μg/kg-wet weight.

Nationally, there were 1003 composite fish samples evaluated for trifluralin. Of the 448 samples of bottom-dwelling fish (homogenized whole), 157 (35%) had detected levels of trifluralin (minimum=3.01, maximum=96.3, and median=9.5 μg/kg-wet weight). Of the 555 samples of predator fish (filleted prior to homogenization), 41 (7%) had detected levels of trifluralin (minimum=3.01, maximum=11.3, and median=4.27 μg/kg-wet weight).

3.4.3 Bioaccumulation Modeling

The KABAM model (K_{OW} (based) Aquatic BioAccumulation Model) version 1.0 is used to evaluate the potential exposure and risk of direct effects to the CRLF and SFGS via bioaccumulation and biomagnification in aquatic food webs. KABAM is used to estimate potential bioaccumulation of hydrophobic organic pesticides in freshwater aquatic ecosystems and risks to mammals and birds consuming aquatic organisms which have bioaccumulated these pesticides.

The bioaccumulation portion of KABAM is based upon work by Arnot and Gobas (2004) who parameterized a bioaccumulation model based on PCBs and some pesticides (e.g., lindane, DDT) in freshwater aquatic ecosystems. KABAM relies on a chemical's octanol-water partition coefficient (K_{OW}) and K_{oc} to estimate uptake and elimination constants through respiration and diet of organisms in different trophic levels. Pesticide tissue residues are calculated for different levels of an aquatic food web. The model then uses pesticide tissue concentrations in aquatic animals to estimate dose- and dietary-based exposures and associated risks to mammals and birds consuming aquatic organisms.

The KABAM model operated primarily in the default mode (see user's guide for full description). The CA Nursery scenario was selected because it had the highest 60 day EEC (0.87 µg/L) of the modeled scenarios.

For the CRLF and SFGS analysis, the ecosystem components for avian consumers were modeled, as birds are considered surrogates for aquatic-phase amphibians. However, the default avian species used in KABAM were altered to more accurately represent the size range and dietary preferences of the CRLF and SFGS. Specifically, three size classes were modeled for the CRLF (1.4g, 37g, and 238g) and SFGS (2g, 113g, and 227g) each with a different dietary composition that would be representative of its body size and likely feeding pattern (see **Table 5.2**). For the two larger size classes of the CRLF and ther SFGS, two different dietary preferences were selected to bound the range of trophic levels at which the CRLF and SFGS could potentially feed.

Physical-chemical parameters and PRZM/EXAMS output parameters required for input into KABAM are listed in **Table 3.7**. The resulting concentrations of trifluralin in tissues of aquatic organisms are provided in **Table 3.8.** The input and output data tables from KABAM are provided in **Appendix N**.

Table 3.7 Summary of KABAM Environmental Fate and PRZM/EXAMS Data for Aquatic Bioaccumulation Modeling

Model Parameter	Value	Data Source
Log Kow	5.27	Product Chemistry
K_{oc}	8,757.9 L/kg [1]	PRZM/EXAM Guidelines
Water Column EEC (60-day)	0.87 µg/L	CA Nursery PRZM/EXAMS Output File
Pore Water Concentration (60-day)	0.36 µg/L	CA Nursery PRZM/EXAMS Output File
Dissolved Oxygen Concentration	5.0 mg O_2/L	CA Nursery PRZM/EXAMS Output File
Water Temp	15.0°C	CA Nursery PRZM/EXAMS Output File

[1] Mean of four Koc values (6,413.3, 6,748.2, 8,457.2, and 13,413 L/kg) used in PRZM/EXAMS (see **Table 3.2**).

Table 3.8 Concentrations of Trifluralin in Tissues of Aquatic Organisms (Estimated Using KABAM Model)

Trophic level	Total concentration (µg/kg-ww)
Phytoplankton	7,201
Zooplankton	5,825
Benthic Invertebrates	6,612
Filter Feeders	4,299
Small Fish	10,938
Medium Fish	14,787
Large Fish	26,226

3.5 Accumulation of Trifluralin Residue on Soil

Because trifluralin is persistent (90% upper confidence bound is 219 days in aerobic soil) and does not have a tendency to leach from soil, a screening analysis was conducted to characterize levels in the soil of a treated site after 30 years of applications. The CA nursery scenario was selected for the screening analysis, as it was expected to result in highest soil concentrations of trifluralin. The 1-in-10-year peak concentration in the soil for total trifluralin was 30.1 g/m^3 for the nursery scenario. The annual average trifluralin soil concentration in soil is 6.41 g/m^3 (**Figure 3.1**).

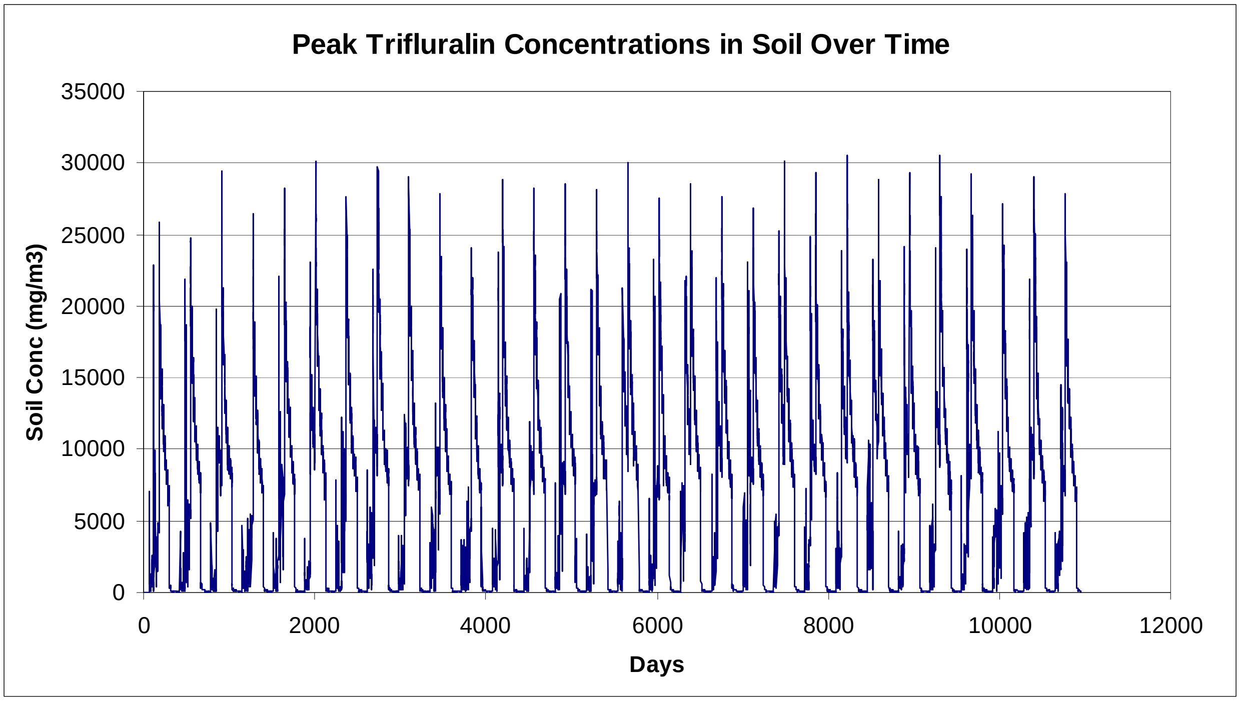


Figure 3.1 Concentration of trifluralin in soil that has been treated with trifluralin for 30 years.

3.6 Terrestrial Animal Exposure Assessment

T-REX (Version 1.4.1) is used to calculate dietary and dose-based EECs of trifluralin for birds, mammals, and terrestrial invertebrates. T-REX simulates a 1-year time period. For this assessment, spray and granular applications of trifluralin are considered, as discussed in below.

Given that no data on interception and subsequent dissipation from foliar surfaces is available for trifluralin, a default foliar dissipation half-life of 35 days is used based on the work of Willis and McDowell (1987). An example output from T-REX is available in **Appendix K**.

Spray applications (not soil-incorporated)

Upper-bound Kenega nomogram values reported by T-REX are used for derivation of dietary- and dose-based EECs for the terrestrial-phase CRLF, SFGS and SJKF and their potential prey (**Table 3.9**). Potential direct acute and chronic effects of trifluralin to the terrestrial-phase CRLF and SFGS are derived by considering dietary-based exposures for a bird consuming small invertebrates. Potential direct acute and chronic effects for the SJKF are derived by considering dose- and dietary-based EECs modeled in T-REX for a large mammal (1,000 g) consuming a variety of dietary items (**Table 3.9**).

T-REX is also used to calculate EECs for terrestrial insects exposed to trifluralin. Dietary-based EECs calculated by T-REX for small and large insects (µg/g-bee) are used to bound an estimate of exposure to bees. Available acute contact toxicity data for bees exposed to trifluralin (in units of µg/bee), are converted to µg/g-bee by multiplying by 1 bee/0.128 g. The EECs are later compared to the adjusted acute contact toxicity data for bees in order to derive RQs. Dietary-based EECs for small and large insects reported by T-REX as well as the resulting adjusted EECs are available in **Table 3.10**.

Table 3.9 Upper-bound Kenega Nomogram EECs for Dietary- and Dose-based Exposures of Birds and Mammals to Trifluralin (liquid applications, not soil incorporated)							
Use Category	**Application Rate (Interval days)**	**Dietary Category**	**Direct Effects for SJKF (1000 g mammal only) Indirect Effects for SJKF (mammalian prey, all weight classes) Indirect Effects for CRLF and SFGS (mammalian prey, 15g only)**			**Direct Effects for CRLF and SFGS (Avian dietary small insects EECs) Direct Effects for SJKF (mammal dietary EECs) Indirect Effects for CRLF, SFGS and SJKF (mammalian and avian dietary EECs)**	
			Mammalian Dosed based EECs (mg/kg-bwt)			**Mammalian Dietary based EECs (mg/kg-diet)**	**Avian Dietary based EECs (mg/kg-diet)**
			15 g	**35 g**	**1000 g**		
Alfalfa	1 app @ 2 lbs/acre	Short Grass	457.64	316.29	73.33	480.00	480.00
		Tall Grass	209.75	144.97	33.61	220.00	220.00
		Broadleaf Plants/ Small Insects	257.42	177.91	41.25	270.00	270.00
		Fruits/Pods/ Seeds/ Large Insects	28.60	19.77	4.58	30.00	30.00
		Granivore*	6.36	4.39	1.02	N/A	N/A
Nursery	3 app @ 4 lbs/acre (60 days)	Short Grass	1279.2	884.12	204.99	1341.72	1341.72
		Tall Grass	586.31	405.22	93.95	614.96	614.96
		Broadleaf Plants/ Small Insects	719.57	497.32	115.30	754.72	754.72
		Fruits/Pods/ Seeds/ Large Insects	79.95	55.26	12.81	83.86	83.86
		Granivore*	17.77	12.28	2.85	N/A	N/A

* EECs for granivores are only relevant for indirect effects.

Table 3.10 Upper-bound Kenega Nomogram EECs for Exposures to Terrestrial Invertebrates from Trifluralin (liquid applications, not soil-incorporated) EECs (indirect effects to terrestrial-phase CRLF, SFGS, and the SJKF)			
Use Category	**Application Rate (Interval days) incorporated**	**Small Insect (mg/kg-insect)**	**Large Insect (mg/kg-insect)**
Alfalfa	1 app @ 2 lbs/acre	270	30
Nursery	3 app @ 4 lbs/acre (60 days)	755	84

Granular applications (not soil incorporated)

For granular formulations, an LD_{50}/sq-ft analysis was performed to evaluate potential acute risks to birds and mammals. The exposure used in this analysis is the mass of trifluralin applied to a square foot area (mg/sq-ft). Based on application rates of 1.5, 2, and 4 lbs/acre with no soil incorporation, the exposure values used in the LD_{50}/sq-ft analysis are 15.6, 20.8, and 41.7 mg/sq-ft, respectively. These exposure estimates are used for all terrestrial taxonomic groups.

Spray and granular applications (soil incorporated)

If the granular material is soil incorporated immediately after application, it is assumed that 1% of the material is available to terrestrial organisms (USEPA 1992). It was assumed that the availability of soil-incorporated spray material to terrestrial organisms would be similar to that of soil-incorporated granular pesticides. These EECs are derived by calculating 1% of the EECs based on the LD_{50}/sq ft exposure calculations. Therefore, the EEC for incorporated trifluralin at the highest single application rate of 2 lb/acre is 0.21mg/sq-ft.

3.7 Terrestrial Plant Exposure Assessment

TerrPlant (Version 1.2.2) is used to calculate EECs for non-target plant species inhabiting dry and semi-aquatic areas. Parameter values for application rate, drift assumption and incorporation depth are based upon the use and related application method. A runoff value of 0.1 is utilized based on trifluralin's solubility, which is classified by TerrPlant as <10 mg/L. For aerial and ground liquid applications, drift was assumed to be 5% and 1%, respectively. For granular applications, drift was assumed to be 0%. EECs relevant to terrestrial plants consider pesticide concentrations in drift and in runoff. These EECs are listed by use in **Table 3.11.** An example output from TerrPlant v.1.2.2 is available in **Appendix M.**

Table 3.11 TerrPlant EECs for Monocots and Dicots Inhabiting Dry and Semi-aquatic Areas Exposed to Trifluralin via Runoff and Drift from liquid and granular applications (single application only)							
Scenario	**Formulation[1]**	**Method[1*]**	**Application Rate (lbs/acre)**	**Drift Value (%)**	**Dry Area EEC (lbs/acre)**	**Semi-aquatic Area EEC (lbs/acre)**	**Spray Drift EEC (lbs/acre)**
Avocado Citrus Grapes and berries Olive Stone and Pome fruit Tree nuts Nursery Right of way Forestry	G	Ground	4	0	0.04	0.4	N/A
Alfalfa	G	Aerial * Ground *	2	0	0.01	0.1	N/A
Citrus Grapes Stone and Pome fruit Tree nuts Cotton Row crop Forestry	EC	Aerial* Ground*	2	5 1	0.11 0.03	0.2 0.12	0.1 0.02
Alfalfa	EC	Aerial Ground	2	5 1	0.12 0.04	0.3 0.22	0.1 0.02
Corn Cole Lettuce Wheat	EC	Aerial*	1	5	0.055	0.1	0.05
Corn Lettuce Melon Cole Tomato Potato Wheat	EC	Ground *	1	1	0.015	0.06	0.01
Onion	EC	Aerial * Ground *	0.8	5 1	0.044 0.012	0.08 0.048	0.04 0.008
Sugar beet	EC	Aerial * Ground *	0.75	5 1	0.041 0.011	0.075 0.045	0.038 0.008
Rangeland Hay	G	Ground	2	0	0.02	0.2	N/A
Turf Residential	G	Ground	1.5	0	0.015	0.15	N/A
Nursery	EC	Ground	4	1	0.08	0.44	0.04

* 2 inch incorporation
[1]G = Granular application. EC = Emulsifiable Concentrate.

4. Effects Assessment

This assessment evaluates the potential for trifluralin to directly or indirectly affect the CRLF, DS, SFGS and SJKF or modify their designated critical habitat. As previously discussed in Section 2.7, assessment endpoints for the effects determination for each assessed species include direct toxic effects on the survival, reproduction, and growth, as well as indirect effects, such as reduction of the prey base or modification of its habitat. In addition, potential modification of critical habitat is assessed by evaluating effects to the PCEs, which are components of the critical habitat areas that provide essential life cycle needs of each assessed species. Direct effects to the aquatic-phase CRLF are based on toxicity information for freshwater fish (or amphibian data if appropriate), while terrestrial-phase amphibian effects (terrestrial-phase CRLF) and reptiles (SFGS) are based on avian toxicity data, given that birds are generally used as a surrogate for terrestrial-phase amphibians and reptiles. Direct effects to the DS are based on toxicity information for freshwater and estuarine/marine fish; data from the most sensitive taxonomic group will be used as the DS both freshwater and estuarine habitats during its lifecycle. Direct effects to the SFGS are based on avian toxicity data, given that birds are generally used as a surrogate for terrestrial-phase amphibians and reptiles. Direct effects to the SJKF are based on mammalian toxicity data.

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

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 trifluralin RED (1996) as well as ECOTOX information obtained on March 31, 2009. In order to be included in the ECOTOX database, papers must meet the following minimum criteria:

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

Open literature toxicity data for 'target' terrestrial plant species, which include efficacy studies, are not currently considered in deriving the most sensitive endpoint for terrestrial plants. Efficacy studies do not typically provide endpoint values that are useful for risk assessment (e.g., NOAEC, EC_{50}, *etc*.), but rather are intended to identify a dose that maximizes a particular effect (*e.g.*, EC_{100}). Therefore, efficacy data and non-efficacy toxicological target data are not included in the ECOTOX open literature summary table provided in **Appendix H.** The list of citations including toxicological and/or efficacy data on target plant species not considered in this assessment is also provided in **Appendix H** [include all citations listed under 'Target Species' in the ECOTOX file name 'Other' in the appendix of ECOTOX papers not considered].

Data that pass the ECOTOX screen are evaluated along with the registrant-submitted data, and may be incorporated qualitatively or quantitatively into this endangered species assessment. In general, effects data in the open literature that are more conservative than the registrant-submitted data are considered. The degree to which open literature data are quantitatively or qualitatively characterized for the effects determination is dependent on whether the information is relevant to the assessment endpoints (*i.e.*, survival, reproduction, and growth) identified in **Section 2.8**. For example, endpoints such as behavior modifications are likely to be qualitatively evaluated, because quantitative relationships between modifications and reduction in species survival, reproduction, and/or growth are not available. Although the effects determination relies on endpoints that are relevant to the assessment endpoints of survival, growth, or reproduction, it is important to note that the full suite of sublethal endpoints potentially available in the effects literature (regardless of their significance to the assessment endpoints) are considered to define the action area for trifluralin.

Citations of all open literature not considered as part of this assessment because they were either rejected by the ECOTOX screen or accepted by ECOTOX but not used (*e.g.*, the endpoint is less sensitive) are included in **Appendix H. Appendix H** also includes a rationale for rejection of those studies that did not pass the ECOTOX screen and those that were not evaluated as part of this endangered species risk assessment. A detailed spreadsheet of the available ECOTOX open literature data, including the full suite of lethal and sublethal endpoints is presented in **Appendix H**. Reviews of several of the ECOTOX and open literature studies are also included in **Appendix H**.

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

As discussed in **Section 2.4.1**, TR-4, TR-6 and TR-15 are the primary degradates of exposure concern for trifluralin. Data recently submitted to the Agency suggests that degradates of trifluralin are less toxic than the parent for most taxonomic groups (**Table 4.1**). These recently submitted studies have not undergone a thorough Agency review; however, a preliminary review did not indicate significant scientific issues. Although data suggest that TR-4 is more toxic to earthworms than the parent, the reported toxicity value (LC_{50} = 186 mg/kg-dry soil) is much greater than expected concentrations in the soil after labeled trifluralin usage. No degradate toxicity data for mammals and birds are available. The Agency does not have concerns for any degradates of trifluralin relative to human health issues as the tolerance is expressed in terms of the trifluralin parent only, based on the determination of the Metabolite Assessment Review Committee (MARC) Health Effects Division (HED) of OPP. Based on this analysis, EFED concluded that trifluralin degradates would not result in exposure concerns for direct or indirect effects to the CRLF, DS, SFGS, or SJKF.

Table 4.1. Comparison of the Toxicity of Trifluralin to the Toxicity of its Degradates.

	Trifluralin	TR-6	TR-15	TR-4
Rainbow trout (96-hr LC_{50}, mg/L)	0.041 (MRID 400980-01)	1.0* (MRID 478070-01)	5.46* (MRID 478070-02)	NA
Daphnid (48-hr EC_{50}, mg/L)	0.251 (MRID 478070-07)	3.52* (MRID 478070-04)	9.36* (MRID 478070-03)	NA
Selenastrum (96-hr IC_{50}, mg/L)	0.089 (MRID 419345-02)	4.09* (MRID 478070-06)	1.67* (MRID 478070-05)	NA
Earthworm (14-day LC_{50}, mg/kg-dry soil)	>1000* (MRID 478070-09)	NA	NA	186* (MRID 478070-10)
* Study submitted by the registrant to EPA in 2009, but not yet reviewed.				

Trifluralin has registered products that contain multiple active ingredients. Analysis of the available open literature and acute oral mammalian LD_{50} data for multiple active ingredient products relative to the single active ingredient is provided in **Appendix A**. In this mixture evaluation an LD_{50} with associated 95% CI is needed for the formulated product. The same quality of data is also required for each component of the mixture. In the case of trifluralin, given that there is no 95% CI associated with the oral LD_{50} (>5000 mg/kg), it is not possible to undertake a quantitative or qualitative analysis for potential interactive effects. However, because the active ingredients are not expected to have similar mechanisms of action, metabolites, or toxicokinetic behavior, it is reasonable to conclude that an assumption of dose-addition would be inappropriate. Several papers were cited in ECOTOX that were classified as evaluating mixtures that contained trifluralin. However, none provided sufficient information for inclusion into this assessment. The results of this analysis show that an assessment based on the toxicity of the single active ingredient of trifluralin is appropriate.

4.1 Toxicity of Trifluralin 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.

Toxicity to fish and aquatic invertebrates is categorized using the system shown in **Table 4.3** (U.S. EPA, 2004). Toxicity categories for aquatic plants have not been defined.

Table 4.2 Aquatic Toxicity Profile for Trifluralin				
Assessment Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	**MRID (Author & Date)**	**Status/Comment**
Freshwater Organisms				
Acute fish	Bluegill sunfish	LC_{50} = 18.5 µg/L probit slope = 23.94	400980-01 (Mayer and Ellersieck, 1986)	Supplemental
Chronic fish	Calculated using ACR	NOAEC = 0.93 µg/L LOAEC not calculated	NA	Calculated using the ACR = 20 for trifluralin rainbow trout data, applied to the bluegill sunfish LC_{50}= 18.5 µg/L
Acute invertebrates	Daphnid	EC_{50} = 251 µg/L (moving average) probit slope = NA	478070-07	Acceptable
Chronic invertebrates	Daphnid	NOAEC = 2.4 µg/L LOAEC= 7.2 µg/L	05008271	Acceptable
Estuarine/Marine Organisms				
Acute fish	Sheepshead minnow	(ethalfluralin) LC_{50} = 240 µg/L probit slope = 1.9	416139-04	Acceptable for ethalfluralin, as there were no acceptable/ supplemental studies for trifluralin.
Chronic fish	Sheepshead minnow	NOAEC = <1.3 µg/L LOAEC=1.3 µg/L	424499-01	Supplemental
Acute invertebrates	Grass shrimp	LC_{50} = 638.5 µg/L Probit slope = 3.48	406748-01	Acceptable
Chronic invertebrates	Calculated using ACR	NOAEC = 6.3 µg/L LOAEC not calculated	NA	Calculated using the ACR = 102 for trifluralin daphnid data, applied to the grass shrimp LC_{50}= 638.5 µg/L
Plants				
Vascular Aquatic Plants	*Lemna gibba*	IC_{50} = 49.7 µg/L NOAEC<2.53 µg/L, LOAEC 2.53 µg/L	428341-04	Supplemental
Non-vascular Aquatic Plants	*Skeletonema costatum*	IC_{50} = 21.9 µg/L NOAEC = 14 µg/L, LOAEC = 18.3 µg/L	428341-01	Acceptable

Table 4.3 Categories of Acute Toxicity for Fish and Aquatic Invertebrates	
LC_{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

4.1.1 Toxicity to Freshwater Fish and Aquatic-Phase Amphibians

A summary of acute and chronic freshwater fish data, including data from the open literature, is provided below in **Sections 4.1.1.1** through **4.1.1.3**.

Although data evaluating the acute toxicity to aquatic-phase amphibians were reviewed, EFED determined that the use of freshwater fish data is preferable to the use of aquatic-phase amphibian data because it is unknown where the CRLF would fall on a species sensitivity distribution. Because amphibian data is not required from the registrant, it is EFED's standard approach to use freshwater fish as a surrogate for aquatic-phase amphibians. In addition, because acute amphibian data were less sensitive than acute freshwater fish data, the use of freshwater fish as a surrogate provides a more conservative estimation of risk to the aquatic-phase CRLF. Chronic aquatic-phase amphibian toxicity data were not available.

Freshwater fish toxicity data were also used to assess potential indirect effects of trifluralin to the CRLF. Effects to freshwater fish resulting from exposure to trifluralin have the potential to indirectly affect the CRLF via reduction in available food, as over 50% of the prey mass of the CRLF may consist of vertebrates such as mice, frogs, and fish (Hayes and Tennant, 1985).

To assess potential direct effects of trifluralin to the DS, toxicity data for both the freshwater fish and estuarine/marine fish were evaluated since the DS lives in brackish waters. The most sensitive freshwater or estuarine/marine fish toxicity data was utilized in the risk estimation. Since freshwater fish were more sensitive than estuarine/marine fish, the freshwater fish toxicity data were used to assess potential direct effects of trifluralin to the DS.

To assess potential indirect effects of trifluralin to the SFGS, toxicity data for freshwater fish were utilized, as the SFGS consumes fish and frogs as a component of its diet.

4.1.1.1 Freshwater Fish: Acute Exposure (Mortality) Studies

Acute toxicity to freshwater fish can be summarized as very highly toxic for trifluralin. The most sensitive LC_{50} values for trifluralin were 18.5 and 43.6 µg/L (bluegill sunfish (*Lepomis macrochirus*) and rainbow trout (*Oncorhynchus mykiss*), respectively (MRID 400980-01, Supplemental, see **Appendix F** for details). The LC_{50} value of 18.5 µg/L (bluegill sunfish) is used for risk estimation. Nominal concentrations were reported for these two studies; these values may overestimate the actual exposure concentration due to trifluralin's tendency to volatilize and sorb to surfaces.

The calculated probit slopes for the bluegill and rainbow studies are 23.94 and 4.16, respectively. There is a high uncertainty in the slope value calculated for the bluegill as the probit model was reported as not a good fit for the data, and this value is well outside the range of typical slopes as the default lower and upper bounds used when the slope cannot be calculated are 2 and 9 (with a mean of 4.5). Therefore, the slope = 4.16 (rainbow trout) will be used to calculate the probability of acute individual effects.

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

Two acceptable chronic studies were available to the Agency to evaluate the effects of chronic exposure to trifluralin (technical) on freshwater fish which were conducted on rainbow trout and fathead minnow (*Pimephales promelas*) (see **Appendix F** for details). The early life-stage study conducted on rainbow trout (MRID 413862-02) demonstrated that chronic exposure to a mean-measured concentration as low as 4.32 µg/L has the potential to cause a reduction in larval fish length (3.5% relative to negative control) and reduction in body weight (8.8% relative to negative control). The NOAEC and LOAEC for this study are 2.18 and 4.32 µg/L, respectively. This study was re-evaluated and the data statistically re-analyzed (updated values, justifications and details are provided in DPbarcode 417055 and **Appendix F)**. Based on the results of this analysis, the NOAEC and LOAEC were changed from the values used in previous assessments.

The full life-cycle study, conducted on fathead minnow (MRID 05008271), demonstrated that chronic exposure to a mean-measured concentration at 5.1 µg/L caused a 64% reduction in survival at 61 weeks. The NOAEC and LOAEC for this study are 1.9 and 5.1 µg/L, respectively.

The Acute-to-Chronic Ratio (ACR) approach also was used to obtain an estimate for chronic trifluralin toxicity to freshwater fish. Both acute and chronic data are available for rainbow trout, LC50 = 43.6 µg/L (MRID 400980-01) and NOAEC = 2.18 µg/L (MRID 413862-02) resulting in an ACR = 20. This ACR was applied to the most sensitive acute toxicity data (bluegill, LC50 = 18.5 µg/L resulting in an estimated chronic NOAEC = 0.93 µg/L. This value will be used for risk estimation.

4.1.1.3 Freshwater Fish: Sublethal Effects and Additional Open Literature Information

Poleksic and Karan (1999, E20430) conducted acute and subacute toxicity tests with carp (*Cyprinus carpio* L) to determine the median lethal concentration and effects on relative growth rates, biochemical parameters (alakaline phosphate [ALP], aspartate aminotransferase [AST], alanin aminotransferase [ALT] activities in serum, gills, liver and kidney) and structure of the gills, liver and kidneys. The 96-hr LC_{50} value (with 95% C.I.) was 45 (36, 51) µg/L. The 14-d subacute test resulted in a decrease in relative growth rate and an increase in functional enzyme activities in blood serum and the organs, most notably at the highest concentration (20 µg/L). ALP and AST activity was increased at all treatment levels in either the gills and/or livers. Gill and liver abnormalities were noted in the 10 and 20 µg/L concentrations and kidney abnormalities were observed at the 20 µg/L treatment level only. The 14-day subacute NOAEC was 5 µg/L.

4.1.1.4 Aquatic-phase Amphibian: Acute Studies

Two independent 96-hour acute tests were conducted with Fowler's toad tadpoles (*Bufo w. fouileri*) on trifluralin (95.9% purity) at a temperature of 60.0°F and a water hardness of 44 mg/L. The resulting LC_{50} values (with 95% C.I.) for Fowler's toad tadpoles were 116.15 (CI not available since only one partial mortality) µg/L and 115.04 (62.0-151.0) µg/L (MRID 400980-01).

Sanders (1970, E2891) conducted a study evaluating acute effects to 4- to 5-week old Fowler's toads resulted in a 96-hr LC_{50} (with 95% C.I.) of 100 (80, 490) µg/L. Sanders reported that mortalities were often preceded by irritability and loss of equilibrium.

4.1.2 Toxicity to Freshwater Invertebrates

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

To assess potential indirect effects (prey) of trifluralin to the DS, toxicity data for both the freshwater invertebrates and estuarine/marine invertebrates were evaluated since the DS lives in brackish waters. The most sensitive freshwater or estuarine/marine invertebrate toxicity data was utilized in the risk estimation. Since freshwater invertebrates were more sensitive than estuarine/marine invertebrates, the freshwater invertebrate toxicity data were used to assess potential indirect effects of trifluralin to the DS.

4.1.2.1 Freshwater Invertebrates: Acute Exposure Studies

One acceptable acute toxicity test to freshwater invertebrate was available to the Agency to evaluate acute exposure of trifluralin (technical) which was conducted on daphnids (MRID 478070-07). The resulting EC_{50} was 251 µg/L (calculated using moving average, see **Appendix F** for details). The EC_{50} and slope obtained from the probit model were unreliable (only one partial mortality in the area of the EC_{50}) and not utilized for this assessment.

This EC_{50} of 251 µg/L for *Daphnia magna* is used to quantitatively estimate acute risk to freshwater invertebrates.

4.1.2.2 Freshwater Invertebrates: Chronic Exposure Studies

Two acceptable chronic (life-cycle) exposure studies with trifluralin (technical) involving the freshwater invertebrate, *Daphnia magna,* were submitted to the Agency. The first life-cycle study (MRID 05008271), demonstrated that chronic exposure to a mean-measured concentration at 7.2 µg/L resulted in 100% mortality in the third generation (day 64 of study). The NOAEC and LOAEC for this study are 2.4 and 7.2 µg/L. The second life-cycle study (MRID 413862-01) demonstrated that chronic exposure to a mean-measured concentration as high as 50.7 µg/L (highest level tested) resulted in no effects on survival, reproduction or growth. The NOAEC for this study is 50.7 µg/L and the LOAEC is > 50.7 µg/L (see **Appendix F** for details).

The NOAEC of 2.4 µg/L for *Daphnia magna* is used to quantitatively estimate chronic risk to freshwater invertebrates.

4.1.2.3 Freshwater Invertebrates: Open Literature Data

Naqvi *et. al* (1985, E11476) conducted acute toxicity studies by exposing ostracods (*Cypria* sp.), cladocerans (*Alonella* sp.), calanoids (*Diaptomus* sp.), and cyclopoids (*Eucyclops* sp.) to Treflan (trifluralin) in mixed species test vessels. The 48-hour LC_{50} values for cladocerans, calanoids, cyclopods, and ostracods were 0.06, 0.08, 0.05 and 0.06 mg/L, respectively, based on nominal concentrations. The results of this study should not be used quantitatively because the initial loading of organisms to the test vessels is unknown, it is unclear if interspecies competition impacted the results of this study, the exposure concentrations were not analytically determined, and the purity of the test material was not reported.

4.1.3 Toxicity to Estuarine/Marine Fish

A summary of acute and chronic estuarine/marine fish data, including data published in the open literature, is provided in **Sections 4.1.3.1** through **4.1.3.3**.

4.1.3.1 Estuarine/Marine Fish: Acute Exposure Studies

No acceptable or supplemental studies evaluating the acute toxicity of trifluralin to estuarine/marine fish were submitted to the Agency. Due to lack of submitted acute studies for estuarine/marine fish and no available open literature data, acute toxicity data from other dinitroanline herbicides were evaluated (benfluralin, butralin, ethalfluralin, oryzalin, and pendimethalin) (**Appendix E**). No data were available for benfluralin or oryzalin; sheepshead minnow (*Cyprinodon variegates*) studies for butralin, ethalfluralin, and pendimethalin resulted in 96-hr LC_{50}s of >180, 240, and 710 µg/L, respectively. The most sensitive definitive acute endpoint was the LC_{50} value of 240 µg/L for sheepshead minnow (MRID 416139-04) from the toxicity test conducted using ethalfluralin. In lieu of acute trifluralin data for estuarine/marine fish, the LC_{50} value 240 µg/L for sheepshead minnow is used for risk estimation.

4.1.3.2 Estuarine/Marine Fish: Chronic Exposure Studies

A 56-day early life cycle study was submitted for sheepshead minnow (*Cyprinodon variegates*, MRID 424499-01). The NOAEC was undefined (< 1.3 µg/L) as reproduction (number of eggs spawned/day/female) in the lowest test concentration was significantly less than in the control group (a 27% reduction). This study was classified as Supplemental because insufficient data were submitted for completing a full review.

4.1.3.3 Estuarine/Marine Fish: Open Literature Data

In a study conducted by Couch (1979, E6425), sheepshead minnows exposed to 5.5 to 31 µg/L of trifluralin during the first 28 days of life developed a vertebral dysplasia. It consisted of semi-symmetrical hypertrophy of vertebra, three to 20 times normal. Effects of the abnormal vertebral development were dorsal vertebral growth into the neural canal, ventral compression of renal ducts, and longitudinal fusion of vertebrae. Fish, exposed for 51 days to 16.6 µg/l trifluralin and thereafter depurated for 4l days, showed no increase in vertebral dysplasia during depuration; however, residual spinal column damage was evident. Serum calcium concentrations were elevated in adult fish exposed for 4 days to 16.6 µg/l trifluralin.

In a second study (Couch 1984, E48406), pituitaries of sheepshead minnows, exposed for 19 months to 1-5 µg/L trifluralin in the laboratory exhibited enlargement, pseudocysts, congestion of blood vessels and oedema. Most of the fish with an enlarged pituitary also had induced diffuse and/or focal vertebral hyperostosis and other dysplastic vertebral changes.

Koyama (1996, E17085) estimated 96-hr LC_{50}s and vertebral deformity rates in 10 marine fish species after exposure to trifluralin, as a Japanese end-use product. Two species had non-definitive LD_{50}s <5.0 µg/L, definitive LC50s ranged from 21 to 120 µg/L, and two species had LC_{50}s of >56 and >74 µg/L. Vertebral deformities were observed in eight of the 10 evaluated species, they were not observed in the two species in which an LC_{50} was not reached. For those species which had vertebral lesions, the lesions were observed at concentrations as low as 5 to 30 µg/L, depending on species. Vertebral deformity rates in fish exposed to concentrations at or above the lowest concentration at which any deformities were observed ranged from 14 to 82%.

4.1.4 Toxicity to Estuarine/Marine Invertebrates

A summary of acute and chronic estuarine/marine invertebrate data, including data published in the open literature, is provided below in **Sections 4.1.4.1** through **4.1.4.3**.

4.1.4.1 Estuarine/Marine Invertebrates: Acute Exposure Studies

The acute toxicity studies available to the Agency demonstrate that trifluralin can be classified as highly toxic to estuarine/marine invertebrates. The EC_{50} values were 638.5 µg/L for grass shrimp (*Palaemonetes pugio*) and >136 µg/L for the mysid (MRIDs 406748-01 and 436620-01, see **Appendix F** for details).

4.1.4.2 Estuarine/Marine Invertebrates: Chronic Exposure Studies

No studies evaluating the chronic toxicity of trifluralin to estuarine/marine invertebrates have been submitted to the Agency. Due to lack of submitted chronic studies for estuarine/marine invertebrates and no available open literature data, the ACR approach was used to obtain an estimate for chronic trifluralin toxicity to estuarine/marine invertebrates. An ACR = 102 was calculated from the trifluralin daphnid data (245 µg/L ÷ 2.4 µg/L) and applied to the grass shrimp data (LC_{50}= 638.5 µg/L) for an estimated chronic toxicity NOAEC = 6.3 µg/L.

4.1.4.3 Estuarine/Marine Invertebrates: Open Literature Data

No studies evaluating toxicity of trifluralin to estuarine/marine invertebrates were identified in open literature that had more sensitive toxicity values.

4.1.5 Toxicity to Aquatic Plants

Laboratory studies are used to evaluate the potential of trifluralin to affect vascular and non-vascular aquatic plants For non-vascular plant laboratory data, the toxicity values used for risk estimation were derived based on the lowest endpoint from from either freshwater or estuarine/marine species since guideline studies do not sufficiently explore the relative sensitivity of algae with regards to freshwater or estuarine/marine environment.

4.1.5.1 Aquatic Plants: Laboratory Data

Five acceptable aquatic plant studies with trifluralin (technical) conducted on *Skeletonema costatum, Selanstrum capricornutum, Anabaena flos-aquae and Lemna gibba* were submitted by the registrant (MRIDs 428341-01, 428341-02, 428341-03, 428341-04, and 419345-02). All five aquatic plant studies were re-evaluated and the data statistically re-analyzed (updated values, justifications and details are provided in DPbarcode 417055 and **Appendix F)**. Based on the results of this analysis, toxicity values from all five studies were changed.

This risk assessment will utilize data from the most sensitive vascular plant (*Lemna gibba*) and non-vascular plant (*Skeletonema costatum*) studies. For *Lemna gibba*, the IC_{50} and IC_{05} are 49.7 and 14.7 µg/L (NOAEC not definitive). For *Skeletonema costatum,* the IC_{50} and NOAEC are 21.9 and 14.0 µg/L.

4.2 Toxicity of Trifluralin to Terrestrial Organisms

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

Table 4.4 Terrestrial Toxicity Profile for Trifluralin

Endpoint	Acute/ Chronic	Species	Toxicity Value Used in Risk Assessment	MRID	Status/Comment
Birds (surrogate for terrestrial-phase amphibians and reptiles)	Acute (oral gavage)	Bobwhite quail Mallard duck	LD_{50} > 2000 mg/kg-bwt LD_{50} > 2000 mg/kg-bwt	00137573 00160000	Practically non-toxic Both studies classified Acceptable, no mortalities or sublethal effects
	Acute (dietary)	Bobwhite quail Mallard duck	LC_{50} > 5000 mg/kg-diet LC_{50} > 5000 mg/kg-diet	00138857 00138858	Practically non-toxic Both studies classified Accceptable, no mortalities or sublethal effects
	Chronic	Mallard Duck	NOAEC = 500 mg/kg-diet LOAEC = 1000 mg/kg-diet	403347-04	Acceptable Effects were reductions in egg shell thickness, 14-day hatchling weights.
Mammals	Acute (oral gavage)	Laboratory rat	LD_{50} > 5000 mg/kg-bwt	00157486	Practically non-toxic Acceptable, no mortalities or sublethal effects
	Chronic	Laboratory rat	NOAEL = 10 mg/kg-bwt/day (converted to 200	00151901 00151902 00151903	Acceptable Effects were kidney toxicity (renal lesions

Table 4.4 Terrestrial Toxicity Profile for Trifluralin					
Endpoint	**Acute/ Chronic**	**Species**	**Toxicity Value Used in Risk Assessment**	**MRID**	**Status/Comment**
			mg/kg-diet) LOAEC = 32.5 mg/kg-bwt/day (converted to 650 mg/kg-diet)		and increased relative liver weight), reduced weanling body weights and reduced litter sizes
Terrestrial invertebrates	Acute	Honey bee, contact	LD_{50} > 24.17 µg/bee	00028772	Supplemental (13% mortality at only test level = 24.17 µg/bee)
Terrestrial plants	N/A	Seedling Emergence Monocots	IC_{25} = 0.09 lbs/acre	439844-01	Sorghum, fresh shoot weight Acceptable, conducted using TEP*
	N/A	Seedling Emergence Dicots	IC_{25} = 0.19 lbs/acre	439844-01	Cucumber, fresh shoot weight Acceptable, conducted using TEP*
	N/A	Vegetative Vigor Monocots	IC_{25} = 1.09 lbs/acre	419345-03	Corn, fresh shoot weight Acceptable, conducted using technical
	N/A	Vegetative Vigor Dicots	IC_{25} = 0.796 lbs/acre	419345-03	Cucumber, fresh shoot weight Acceptable, conducted using technical
N/A: not applicable * TEP = typical end-use product, in this case product was Treflan HFP containing 43.8% trifluralin					

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

Table 4.5 Categories of Acute Toxicity for Terrestrial Organisms		
Categories of Acute Toxicity for Birds and Mammals		
Toxicity Category	**Oral LD_{50}**	**Dietary LC_{50}**
Very highly toxic	< 10 mg/kg	< 50 ppm
Highly toxic	10 – 50 mg/kg	50 - 500 ppm
Moderately toxic	51 – 500 mg/kg	501 - 1000 ppm
Slightly toxic	501 - 2000 mg/kg	1001 - 5000 ppm
Practically non-toxic	> 2000 mg/kg	> 5000 ppm

Categories of Acute Toxicity for Non-Target Insects	
Toxicity Category	**Contact LC_{50}**
Highly toxic	< 2 µg/bee
Moderately toxic	2-11 µg/bee
Practically non-toxic	>11 µg/bee

4.2.1 Toxicity to Birds

As specified in the Overview Document, the Agency uses birds as a surrogate for reptiles and terrestrial-phase amphibians when toxicity data for each specific taxon are not available (U.S. EPA, 2004). No terrestrial-phase amphibian or reptile data are available for trifluralin; therefore, acute and chronic avian toxicity data are used to assess the potential direct effects of trifluralin to terrestrial-phase CRLF and SFGS, as well as potential effects on prey of terrestrial-phase CRLF, SFGS, and SJKF. A summary of acute and chronic bird data, including data published in the open literature, is provided below in **Sections 4.2.1.1** through **4.2.1.4**. Additional information on these studies is provided in **Appendix F**.

4.2.1.1 Birds: Acute Exposure (Mortality) Studies

The acute toxicity of the technical grade trifluralin (~96.7% purity) to birds was established with the following guideline tests: two avian single-dose oral (LD_{50}) studies on the bobwhite quail and mallard duck; two sub-acute dietary studies (LC_{50}) on the mallard duck and the bobwhite quail.

The non-definitive LD_{50}s for bobwhite quail and mallard duck were > 2000 mg/kg-bw (MRIDs 00137573 and 00160000). The LC_{50}s for both bobwhite quail and mallard duck were > 5000 mg/kg-diet (MRIDs 00138858 and 00138857). No mortalities or sublethal effects occurred at any treatment level for all the above acute avian studies. Based on these studies, acute exposure of trifluralin to birds was classified as "practically non-toxic".

4.2.1.2 Birds: Chronic Exposure (Growth, Reproduction) Studies

Avian chronic exposure reproduction effects studies were performed for using two species, bobwhite quail and mallard duck.

Two recent studies were classified as acceptable for the bobwhite quail and mallard duck using 96% technical trifluralin. In the bobwhite quail study (MRID 403347-06 and DPbarcode 417055 for revisions), the NOAEC was established at 1000 mg/kg-diet with no significant adverse effects observed. In the mallard duck study (MRID 403347-04), the NOAEC was established at 500 mg/kg-diet with the most sensitive endpoints being reduction in eggshell thickness, 14-day survival body weight of chicks and male body weight for the duck at the highest test concentration of 1000 mg/kg-diet.

Additional bobwhite quail and mallard duck studies using 99.6% technical (MRIDs 00131134 and 00131132, DPbarcode 417055 for revisions) were also submitted to the Agency; these studies were used for previous risk assessments. For the bobwhite quail study, the NOAEC was established at 50 mg/kg-diet (the highest test concentration), as no significant effects were observed for any of the reported endpoints. Although this study was classified as Acceptable, there is concern for the validity of the study as the overall percentage of cracked eggs was high (9.7% of eggs laid in controls). EPA guidance for bobwhite quail reproduction studies states that typically only 0.6 – 2.0% of eggs laid are cracked. For the mallard duck study, the NOAEC was established at 5 mg/kg-diet, as there was a statistically significant reduction in the percentage of eggs not cracked ("eggs not cracked/eggs laid") at the highest test concentration of 50 mg/kg-diet. This effect was relatively small (98.8, 99.7, and 97.0% eggs not cracked/eggs laid in the control, 5 mg/kg-diet, and 50 mg/kg-diet groups, respectively). This effect was not observed in any of the other chronic bird studies including those that had test concentrations up to 1000 mg/kg-diet (MRID 403347-04 and 403347-06). After evaluating the size of the effect and the full suite of avian reproduction studies conducted for trifluralin, EFED determined that the reduction in the percentage of eggs cracked in this study (MRID 00131132) was not biologically significant, and the study NOAEC should be established at 50 mg/kg-diet (the highest concentration tested).

For risk estimation in this assessment, a chronic avian NOAEC of 500 mg/kg-diet is used. This is based on the mallard duck study (MRID 403347-04) with effected endpoints of reduction in eggshell thickness, 14-day survival body weight of chicks and male body weight for the duck at the highest test concentration of 1000 mg/kg-diet. Any of these effects may have an effect on the fitness of individuals and on the overall fitness of wild bird populations exposed to trifluralin. Since birds are used as a surrogate for the terrestrial-phase CRLF and the SFGS, these effects have the potential to result in direct effects to the frog and snake by affecting reproductive fitness or to result in indirect effects to the terrestrial-phase CRLF, SFGS, and SJKF by affecting their prey populations.

4.2.1.3 Birds, Reptiles and Terrestrial-phased Amphibians: Open Literature Studies

No studies evaluating toxicity of trifluralin to birds, reptiles and terrestrial-phase amphibians were identified in open literature that had more sensitive toxicity values.

4.2.2 Toxicity to Mammals

A summary of acute and chronic mammalian data, including data published in the open literature, is provided below in **Sections 4.2.2.1** through **4.2.1.2**. Additional information on these studies is provided in **Appendix F**.

Mammalian toxicity data are used to assess potential direct and indirect effects of trifluralin to the SJKF and indirect effects of trifluralin to the terrestrial-phase CRLF, SJKF and SFGS as discussed in **Section 2.8**. For the purposes of this risk assessment, the available mammalian toxicity data on laboratory rodents as surrogates for mammalian wildlife were used.

4.2.2.1 Mammals: Acute Exposure (Mortality) Studies

There is one-registrant submitted acceptable rat acute oral toxicity study. No mortality or signs of systemic toxicity were reported; based on a laboratory rat LD_{50} value greater than 5000 mg/kg-bw, trifluralin is practically non-toxic to small mammals on an acute oral basis (MRID 00157486). Based on these studies, acute toxicity of trifluralin to mammals was classified as "practically non-toxic".

4.2.2.2 Mammals: Chronic Exposure (Growth, Reproduction) Studies

In 2-generation rat reproduction study (MRIDs 00151901, 00151902 and 00151903, technical grade), toxic effects in Wister KFM-Han rats included kidney toxicity (renal lesions and increased relative liver weight), reduced weanling body weights and reduced litter sizes. The NOAEC for reproductive and developmental toxicity was 200 mg/kg-diet and the LOAEC was 650 mg/kg-diet based on reduced weanling body weights at 650 and 2000 mg/kg-diet and reduced litter sizes at the highest concentration (2000 mg/kg-diet). Since terrestrial-phase CRLF, SFGS and SJKF may consume mammals, these effects have the potential to result in indirect effects by affecting their prey populations. Using standard laboratory rat weights, the NOAEC = 200 mg/kg-diet can be converted to a NOAEL = 10 mg/kg-bwt. This study is used for risk estimation in this assessment.

4.2.2.3 Mammals: Open Literature Studies

No studies evaluating toxicity of trifluralin to mammals were identified in open literature that had more sensitive toxicity values than those studies submitted by to the Agency.

4.2.3 Toxicity to Terrestrial Invertebrates

A summary of acute terrestrial invertebrate data, including data published in the open literature, is provided below in **Sections 4.2.3.1** through **4.2.3.2**.

4.2.3.1 Terrestrial Invertebrates: Acute Exposure (Mortality) Studies

Based on an acute contact LD_{50} of >24.17 µg/bee, trifluralin appears to be “practically non-toxic” to honeybees (MRID 00028772). At the only tested dose, 24.17 µg/bee, 12.85% mortality occurred.

One additional study honeybee study was submitted with an acute contact of LD_{50} of > 100 µg/bee and acute oral of LD_{50} of > 50 µg/bee, trifluralin appears to be “practically non-toxic” to honeybees (MRID 05001991). Mortality at each test dose was not reported.

The non-definitive terrestrial invertebrate toxicity value for acute contact to bees with an LD_{50} of >24.17 µg/bee is used qualitatively to characterize the indirect effects to the SFGS, SJKF and terrestrial-phase CRLF.

4.2.3.2 Terrestrial Invertebrates: Open Literature Studies

Toxicity of trifluralin to mature earthworms (*Eisenia foetida)* was evaluated by Roberts and Dorough (E040531; 1984). Trifluralin was found to be relatively non-toxic to earthworms with a non-definitive LC_{50} >1000 $\mu g/cm^2$, in a 48-hr study.

4.2.4 Toxicity to Terrestrial Plants

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

Commercial crop species have been selectively bred, and may be more or less resistant to particular stressors than wild herbs and forbs. The direction of this uncertainty for specific plants and stressors, including trifluralin, 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.

4.2.4.1 Terrestrial Plants: Registrant-submitted Studies

In general, toxicity tests demonstrate that trifluralin negatively impacts seedling emergence and vegetative vigor of terrestrial plants (MRID 439844-01, 419345-03). Results of Tier II toxicity testing on the technical material and the end-use product are summarized in **Appendix F**.

One terrestrial plant seedling emergence Tier II study was classified as acceptable using the formulation product Treflan HFP (43.8% purity) (MRID 439844-01). In general, the emerged monocots were more sensitive than emerged dicots based on EC_{25} comparisons. Shoot fresh weight was the most sensitive parameter for both monocots and dicots. In the seedling emergence study; sorghum was the most sensitive monocot (IC_{25} = 0.09 lb/acre), and cucumber was the most sensitive dicot (IC_{25} = 0.19 lb/acre).

A Tier II study was submitted that evaluated the effects of technical trifluralin (95.7% purity) on non-target terrestrial plant vegetative vigor (MRID 419345-03). Shoot fresh weight was the most sensitive parameter for both monocots and dicots. For the vegetative vigor test, the most sensitive monocot was corn (IC_{25} = 1.09 lb/acre) and the most sensitive dicot was cucumber (IC_{25} = 0.796 lb/acre).

Table 4.6 Non-target Terrestrial Plant Seedling Emergence Toxicity (Tier II) Data for Trifluralin TEP (Treflan HFP) 43.8% purity (MRID 439844-01)*				
Crop	**Species**	**NOAEC (lb a.i./acre)**	**IC_{25} (lb a.i./acre)**	**Most Sensitive Endpoint**
Monocot	Corn	0.13	0.17	Shoot fresh weight
	Sorghum	0.06	0.09	Shoot fresh weight
	Onion	0.50	0.74	Shoot fresh weight
	Wheat	0.13	0.21	Shoot fresh weight
Dicot	Cotton	NA	NA	Results invalid (soil medium detrimental to plant growth)
	Cabbage	0.50	0.78	Shoot fresh weight
	Radish	1.0	2.4	Shoot fresh weight
	Cucumber	0.13	0.19	Shoot fresh weight
	Soybean	1.0	1.3	Shoot fresh weight
	Sunflower	2.0	4.0	Shoot fresh weight
* For this study, an incorporated application was simulated by spraying the material into a rotating cement mixer filled with soil. This soil was used to provide the top two 2 inches of soil in each treatment pot.				

Table 4.7 Non-target Terrestrial Plant Vegetative Vigor Toxicity (Tier II) Data for Trifluralin (Treflan 95.7% purity, MRID 419345-03)				
Crop	**Species**	**NOAEC (lbs/acre)**	**IC_{25} (lbs/acre)**	**Most Sensitive Endpoint**
Monocot	Corn	0.50	1.09	Fresh Shoot weight
	Sorghum	ND [d]	2.648 [a]	Height
	Onion	0.25	1.45	Height
	Wheat	ND	>2 [c]	Height and fresh shoot weight
Dicot	Cabbage	ND	2.644 [a]	Height
	Cotton	ND	2.267 [a]	Height
	Cucumber	0.25 [b]	0.796	Fresh Shoot weight
	Sunflower	ND	2.476 [a]	Height
	Soybean	ND	>2 [c]	Height and fresh shoot weight
	Radish	0.25	0.939	Height
[a] values reported by the study author, not verified by EFED reviewer [b] NOAEC=0.125 lbs/acre was used in the RED, correct value is 0.25 lbs/acre [c] Endpoints determined visually by study author, no statistical analysis conducted. [d] Not determined; NOAEC values not reported by the study author or the EFED reviewer.				

Generally, EFED prefers terrestrial plant toxicity tests to be conducted using end-use products and dry shoot weights as this better represents the exposure of non-target plants in the environment. For this risk assessment, EFED is using toxicity testing results that were conducted with technical trifluralin (for vegetative vigor) and with measured fresh weights for both seedling emergence and vegetative vigor. Since the results for seedling emergence and vegetative vigor were not both conducted on end-use products for the most sensitive parameters, results between the two studies should be compared with caution.

4.2.4.2 Terrestrial Plants: Open Literature Studies

No studies evaluating toxicity of trifluralin to terrestrial plants suitable for inclusion in this risk assessment were identified in the open literature. Available open literature studies focused on efficacy evaluation; IC_{25}s were not available from those studies.

4.4 Incident Database Review

A review of the EIIS database for ecological incidents involving trifluralin was completed on 7/10/2009. The results of this review for terrestrial, plant, and aquatic incidents are discussed below in **Sections 4.4.1** through **4.4.3**, respectively. A complete list of the incidents involving trifluralin including associated uncertainties is included as **Appendix I**. This table is divided into incidents involving aquatic organisms and terrestrial organisms.

Incidents listed in EIIS are categorized by the likelihood that a particular pesticide is associated with that particular incident. These classifications include highly probable, probable, possible, unlikely or unrelated. “Highly probable” incidents usually require carcass residues or clear circumstances regarding the exposure. “Probable” incidents include those where residue information was not available or circumstances were less clear than those for “highly probable.” “Possible” incidents occur when multiple chemicals may have been involved and the contribution of an individual chemical is not obvious. An “unlikely” incident classification is given when a given chemical is considered nontoxic to the type of organism involved or the chemical was analyzed and not detected in samples. The “unrelated” category is used for incidents confirmed not to involve pesticides. No unrelated incidents were listed for trifluralin.

The number of reports listed in the EIIS database is believed to be only a fraction of the total incidents involving organismal mortality and damage caused by pesticides. Few resources are assigned to incident reporting. Reporting by states is only voluntary, and individuals discovering incidents may not be informed on the procedure of reporting these occurrences. Additionally, much of the database is generated from registrant-submitted incident reports. Registrants are legally required to provide detailed reports of only “major” ecological incidents involving pesticides, while “minor” incidents are reported aggregately. Because of these logistical difficulties, EIIS is most likely a minimal representation of all pesticide-related ecological incidents.

The EIIS database contained 83 incident reports involving trifluralin. Most of the incidents involve terrestrial ecosystems involving effects to terrestrial plants (78 or 94% of the total incidents). Five incident reports involve aquatic ecosystems. North Dakota was most represented among all 50 states (20 reports) followed by Iowa (16), Minnesota (7), California (6) and Texas (5). Additional characterization of these incidents is provided below.

Of the 83 incidents reported, 1 (1.2%) are categorized as ‘highly probable’ and 58 (69.8%) are categorized as ‘probable.’ Collectively the ‘highly probable’ and ‘probable’ categories represent

71% of the reported incidents. Regarding the legal status, the 'registered use' represents the largest legality category with 81.9% of the incidents reported.

Approximately 9.6% of the reports consist of 'misuse accidental' and 8.4 % undetermined uses.

4.4.1 Terrestrial Animal Incidents

No terrestrial animal incidents were reported.

4.4.2 Terrestrial Plant Incidents

For trifluralin, 78 incidents were reported for plant damage to a wide variety of terrestrial plants particularly from direct treatment or spray drift (e.g., Alfalfa, barley, bean, birch, blue spruce, corn, cotton, dry bean, ornamentals, peanut, percifia shrubs, pinto bean, potato, raspberry, rose, soybean, soybean seed, spreading yew, sudan grass, sugarcane, sunflower, tomato and wheat (spring and other varieties). For trifluralin, 66 of the 78 incidents reported were registered uses, five were accidental misuses and seven were of unknown legality. Of the 78 incidents involving terrestrial plants, 55 (70.5%) are classified as 'probable' in the context of trifluralin use. Other reported incident exposures included spills, stunted growth, discoloration, reduced yield, incapacitation, mortality, runoff, and carryover.

4.4.3 Aquatic Animal Incidents

All five of the aquatic incident reports involved mortality to aquatic organisms. One of the five incidents reportedly involved an eel, but the likelihood of observing impacts to eels are low compared to fish based on a reported trifluralin spill. Of the five aquatic incidents involving fish, (80%) are classified as either 'highly probable' or 'probable' in the context of trifluralin use. A wide variety of fresh and estuarine fish species were reportedly affected due to runoff and accidental spills (e.g., catfish, minnow, bass, shad, bluegill sunfish, gar and crappie).

All of the probable incidents were accidental misuses except incident # I002215-001 which occurred in Texas. This report failed to give the name of the county where the incident occurred. The incident date, which was not reported in the memo of 06/12/95, was estimated to be 06/01/95. According to the report, an area was treated with pesticide prior to asphalt paving. Light rain followed and there was a heavy thunderstorm a day later which allegedly resulted in a runoff into a three acre pond causing a fishkill of an unspecified number of bass, bluegill and crappie. Catfish and carp were not affected. It was believed that trifluralin, as Treflan, was responsible for the observed mortality.

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 DS, SFGS and SJKF or for modification to their designated critical habitat (CRLF and DS only) from the use of trifluralin 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 is estimated by calculating the ratio of exposure to toxicity. This ratio is the risk quotient (RQ), which is then compared to the pre-established acute and chronic levels of concern (LOCs) for each category evaluated (**Appendix J**). For acute exposures to the aquatic animals, as well as terrestrial invertebrates, the acute LOC is 0.05. For acute exposures to the birds (and, thus, reptiles and terrestrial-phase amphibians) and mammals, the LOC is 0.1. The LOC for chronic exposures to animals, as well as acute exposures to plants is 1.0.

Acute and chronic risks to aquatic organisms are estimated by calculating the ratio of exposure to toxicity using 1-in-10 year EECs based on the label-recommended trifluralin usage scenarios summarized in **Table 3.3** and the appropriate aquatic toxicity endpoint from **Table 4.2**. Acute and chronic risks to terrestrial animals are estimated based on exposures resulting from applications of trifluralin (**Tables 3.9** and **3.10**) and the appropriate toxicity endpoint from **Table 4.4**. Exposures are also derived for terrestrial plants, as summarized in **Table 3.11**, and toxicity endpoints are listed in **Table 4.4**.

5.1.1 Exposures in the Aquatic Habitat

5.1.1.1 Freshwater Fish and Aquatic-phase Amphibians

Acute risk to freshwater fish and aquatic-phase amphibians and the potential for direct effects to CRLF and DS specifically is based on peak EECs in the standard PRZM/EXAMS pond and the lowest acute toxicity value for freshwater fish. Based on freshwater fish toxicity data (LC_{50} value of 18.5 µg/L for bluegill sunfish) and modeled aquatic peak EECs for various use scenarios used to represent uses of trifluralin in CA (**Table 3.3**), acute RQs for freshwater fish resulted in exceedances of the Listed Species LOC (RQ>0.05) for 18 out of 25 modeled crop scenarios with exceedances occurring with at least one of the application methods for each scenario (**Table 5.1**). The RQs exceeding the Listed Species LOC ranged from 0.05 to 0.35.

The probability of acute individual effects for freshwater fish and aquatic-phase amphibians is <u><</u>1 in 3280 (calculated using a probit slope = 4.16, MRID 400980-01) for those scenarios in which the Listed Species LOC was exceeded (6.55 ppb/43.6= RQ of 0.15 for rainbow trout). At the Listed Species LOC, the probability of acute individual effects for freshwater fish and aquatic-phase amphibians also is <u><</u> 1 in 32100000. The probit slope of 23.94 (MRID 400980-01) was not used to calculate probabilities due to high uncertainty of the probit analysis derived from this value.

Chronic risk to freshwater fish and aquatic-phase amphibians and the potential for direct effects to aquatic-phase CRLF and DS specifically is based on 60-day EECs and the lowest chronic toxicity value for freshwater fish. Based on 60-day EECs for various use scenarios used to represent uses of trifluralin in CA and the NOAEC of 0.93 µg/L (estimated via ACR), no chronic RQs resulted in a Chronic LOC exceedance (all RQ < 1.0) (**Table 5.1**).

Based on exceedances of the Agency's acute listed species LOC (RQ>0.05) for 18 modeled scenarios trifluralin does have the potential to directly affect the aquatic phase CRLF and DS. Additionally, since acute RQs are exceeded, there is a potential for indirect effects to those listed species that rely on freshwater fish (and/or aquatic-phase amphibians) during at least some portion of their life-cycle (*i.e*., CRLF and SFGS).

Table 5.1 Acute and Chronic RQs for Aquatic-phase CRLF and DS (freshwater fish surrogate) Resulting from Trifluralin Application

Crop Scenario/ Formulation	Application Method	Soil Incorporation	Peak EEC µg/L	60 day EEC µg/L	Acute RQs[1]	Chronic RQs[2]
Orchard Uses						
CA Almond (including tree nuts)						
Granular	Ground	N	2.23	0.72	**0.12**	0.77
EC	Aerial	Y	5.65	0.6	**0.31**	0.65
	Ground	Y	1.23	0.18	**0.07**	0.19
CA Avocado						
Granular	Ground	N	5.81	0.73	**0.31**	0.78
CA Citrus						
Granular	Ground	N	0.74	0.14	0.04	0.15
EC	Aerial	Y	5.48	0.54	**0.30**	0.58
	Ground	Y	1.10	0.11	**0.06**	0.12
CA Fruit						
Granular	Ground	N	2.52	0.4	**0.14**	0.43
EC	Aerial	Y	5.51	0.55	**0.30**	0.59
	Ground	Y	1.12	0.13	**0.06**	0.14
CA Grape (including berries)						
Granular	Ground	N	4.0	0.62	**0.22**	0.67
CA Grape						
EC	Aerial	Y	5.5	0.54	**0.30**	0.58
	Ground	Y	1.18	0.12	**0.06**	0.13
CA Olive						
Granular	Ground	N	4.45	0.77	**0.24**	0.83
Agricultural Uses						
CA Alfalfa						
EC	Aerial	N	5.14	0.58	**0.28**	0.62
	Ground	N	1.12	0.17	**0.06**	0.18
Granular	Aerial	Y	0.32	0.07	0.02	0.08
	Ground	Y	0.32	0.07	0.02	0.08
CA Cole						
EC	Aerial	Y	2.79	0.29	**0.15**	0.31
	Ground	Y	0.59	0.1	0.03	0.11
CA Corn						
EC	Aerial	Y	2.74	0.26	**0.15**	0.28
	Ground	Y	0.55	0.07	0.03	0.08
CA Cotton						
EC	Aerial	Y	5.55	0.65	**0.30**	0.70
	Ground	Y	1.48	0.24	**0.08**	0.26
CA Melon						
EC	Ground	Y	0.73	0.13	0.04	0.14

Table 5.1 Acute and Chronic RQs for Aquatic-phase CRLF and DS (freshwater fish surrogate) Resulting from Trifluralin Application						
Crop Scenario/ Formulation	**Application Method**	**Soil Incorporation**	**Peak EEC µg/L**	**60 day EEC µg/L**	**Acute RQs[1]**	**Chronic RQs[2]**
CA Lettuce						
EC	Aerial	Y	2.77	0.27	**0.15**	0.29
	Ground	Y	0.58	0.08	0.03	0.09
CA Onion						
EC	Aerial	Y	1.96	0.21	**0.11**	0.23
	Ground	Y	0.39	0.05	0.02	0.05
CA Potato						
EC	Ground	Y	0.55	0.05	0.03	0.05
CA Rangeland Hay						
Granular	Ground	N	0.68	0.12	0.04	0.13
CA Tomato						
EC	Ground	Y	0.56	0.06	0.03	0.06
CA Row Crop						
EC	Aerial	Y	5.49	0.45	**0.30**	0.48
	Ground	Y	1.01	0.09	**0.05**	0.10
CA Sugar Beet						
EC	Aerial	Y	1.96	0.17	**0.11**	0.18
	Ground	Y	0.39	0.03	0.02	0.03
CA Wheat						
EC	Aerial	Y	2.77	0.29	**0.15**	0.31
	Ground	Y	0.57	0.11	0.03	0.12
Non-agricultural Uses						
CA Nursery						
Granular	Ground	N	6.55	0.86	**0.35**	0.92
EC	Ground	N	6.53	0.87	**0.35**	0.94
CA Residential						
Granular	Ground	N	0.0002	0.00005	<0.01	<0.01
CA Right-of-way						
Granular	Ground	N	0.002	0.0005	<0.01	<0.01
CA Turf						
Granular	Ground	N	0.16	0.02	<0.01	0.02
CA Forestry						
Granular	Ground	N	2.25	0.39	**0.12**	0.42
EC	Aerial	Y	5.5	0.63	**0.30**	0.68
	Ground	Y	1.17	0.19	**0.06**	0.20

[1, 2] LOC exceedences (acute RQ $\geq$ 0.05; chronic RQ $\geq$ 1.0) are bolded. Acute RQ = use-specific peak EEC / 96-h LC_{50} = 18.5µg/L for bluegill sunfish. Chronic RQ = use-specific 60-day EEC / NOAEC = 0.93 µg/L ACR value (rainbow trout LC50 43.6 µg/L/rainbow trout NOAEC 2.18 µg/L = 20; bluegill sunfish LC 50 18.5 µg/L / 20= ACR 0.93 µg/L)

5.1.1.2 Freshwater Invertebrates

Acute risk to freshwater invertebrates is based on peak EECs in the standard PRZM/EXAMS pond and the lowest acute toxicity value for freshwater invertebrates. Based on freshwater invertebrate toxicity data (EC_{50} value of 251 µg/L for daphnid) and modeled aquatic peak EECs for various use scenarios used to represent all of the agricultural uses of trifluralin in CA, no acute RQs resulted in a Listed Species LOC exceedance (all RQ < 0.05) (**Appendix O**).

Chronic risk is based on 21-day EECs and the lowest chronic toxicity value for freshwater invertebrates. Based on 21-day EECs for various use scenarios used to represent all of the agricultural uses of trifluralin in CA, and the NOAEC of 2.4 µg/L, no RQs resulted in a Chronic LOC exceedance (all RQ < 1.0) (**Appendix O**).

Since the RQs do not exceed the Listed Species LOC (all RQ < 0.05) or the Chronic LOC (all RQ < 1.0), trifluralin is determined to have no indirect effects to those listed species that rely on freshwater invertebrates during at least some portion of their life-history (i.e., aquatic-phase CRLF, DS, and SFGS).

5.1.1.3 Non-vascular Aquatic Plants

Acute risk to aquatic non-vascular plants is based on peak EECs in the standard pond and the lowest acute toxicity value. Based on the aquatic non-vascular plant toxicity datum (IC_{50} = 21.9 µg/L for the marine diatom (*Skeletonema costatum*) and the maximum aquatic peak EEC of all trifluralin scenarios, all RQs for aquatic non-vascular plants are ≤ 0.30 **(Appendix O)**. Since the RQs do not exceed the Acute LOC (all RQ < 1.0), trifluralin is determined to have no indirect effects to those listed species that rely on non-vascular aquatic plants during at least some portion of their life-history (i.e., aquatic-phase CRLF, DS, and SFGS).

5.1.1.4 Aquatic Vascular Plants

Acute risk to aquatic vascular plants is based on peak EECs in the standard pond and the lowest acute toxicity value. Based on the aquatic vascular plant toxicity datum (IC_{50} = 49.7 µg/L for the *Lemna gibba* and the maximum aquatic peak EEC of all trifluralin scenarios, all RQs for aquatic vascular plants are ≤ 0.13 **(Appendix O)**. Since the RQs do not exceed the Acute LOC (all RQ < 1.0), trifluralin is determined to have no indirect effects to those listed species that rely on vascular aquatic plants during at least some portion of their life-history (i.e., aquatic-phase CRLF, DS, and SFGS).

5.1.1.5 Bioaccumulation of Trifluralin in Aquatic Prey Items

As discussed in **Section 3.4**, trifluralin has the potential to accumulate in tissues of aquatic organisms. Since the CRLF and the SFGS consume algae, aquatic invertebrates and fish, they could be exposed to trifluralin accumulated in the tissues of these prey. In order to define the risks of the CRLF and the SFGS consuming benthic invertebrates and fish, KABAM was used to derive RQ values for small (1.4 g), medium (37 g) and large (238 g) CRLF as well as juvenile (2 g), adult male (113 g), and adult female (227 g) SFGS. Diet assumptions assigned to each of these species and size classes are provided in **Table 5.2**. The resulting RQ values for the CRLF and SFGS are provided in **Table 5.3**. RQ values for all size classes did not exceed the LOC for chronic dietary-based exposures to the CRLF or the SFGS through consumption of contaminated aquatic prey that have accumulated trifluralin. Acute RQs were not calculated as the avian acute toxicity studies resulted in no mortalities or sublethal effects at the highest dose tested. This highest dose was higher than exposure levels expected in the environment.

Table 5.2. Diet Assumptions of CRLF and SFGS used in KABAM.					
	Diet for:				
Trophic level in diet	**small CRLF (0.0014 kg)**	**med CRLF 1 (0.037 kg)**	**med CRLF 2 (0.037 kg)**	**large CRLF 1 (0.238 kg)**	**large CRLF 2 (0.238 kg)**
phytoplankton	0.0%	0.0%	0.0%	0.0%	0.0%
zooplankton	0.0%	0.0%	0.0%	0.0%	0.0%
benthic invertebrates	100.0%	100.0%	0.0%	100.0%	0.0%
filter feeders	0.0%	0.0%	0.0%	0.0%	0.0%
small fish	0.0%	0.0%	100.0%	0.0%	0.0%
medium fish	0.0%	0.0%	0.0%	0.0%	100.0%
large fish	0.0%	0.0%	0.0%	0.0%	0.0%
Total	100.0%	100.0%	100.0%	100.0%	100.0%
Trophic Level in Diet	**Juv SFGS (0.002 kg)**	**Adult Male SFGS 1 (0.113 kg)**	**Adult Male SFGS 2 (0.113 kg)**	**Adult Female SFGS 1 (0.227 kg)**	**Adult Female SFGS 2 (0.227 kg)**
Phytoplankton	0.0%	0.0%	0.0%	0.0%	0.0%
Zooplankton	0.0%	0.0%	0.0%	0.0%	0.0%
Benthic Invertebrate	100.0%	100.0%	0.0%	100.0%	0.0%
Filter feeders	0.0%	0.0%	0.0%	0.0%	0.0%
Small fish	0.0%	0.0%	0.0%	0.0%	0.0%
Medium Fish	0.0%	0.0%	100.0%	0.0%	100.0%
Large Fish	0.0%	0.0%	0.0%	0.0%	0.0%
Total	100.0%	100.0%	100.0%	100.0%	100.0%

Table 5.3. Chronic RQs for CRLF and SFGS Through Consumption of Aquatic Organisms which have Accumulated Trifluralin.	
Species, Size Class, and Diet[1]	**Chronic, Dietary Based RQ[2]**
small CRLF	0.013
med CRLF 1	0.013
med CRLF 2	0.021
large CRLF 1	0.013
large CRLF 2	0.028
juv SFGS	0.013
SFGS male 1	0.013
SFGS male 2	0.030
SFGS female 1	0.013
SFGS female 2	0.030

[1] See **Table 5.2** for definitions of size class and diet.
[2] Based on NOAEC = 500 mg/kg-diet (for mallard duck), bolded values exceed chronic risk LOC (RQ>1.0)

5.1.2 Exposures in the Terrestrial Habitat

5.1.2.1 Birds (Surrogate for Reptiles and Terrestrial-phase Amphibians)

As previously discussed in **Section 3.6**, potential direct effects to terrestrial species are based on spray and granular applications of trifluralin. Potential risks to birds (and, thus, reptiles and terrestrial-phase amphibians) are derived using T-REX, acute and chronic toxicity data for the most sensitive bird species for which data are available, and a variety of body-size and dietary categories.

Spray applications (no soil incorporation)

Potential direct acute effects to the terrestrial-phase CRLF and SFGS are derived by considering dose- and dietary-based EECs modeled in T-REX for a small bird (20 g) consuming small invertebrates (**Table 3.5**) and acute oral and subacute dietary toxicity endpoints for avian species. RQs for the non-definitive acute oral and subacute dietary toxicity endpoints for avian species were not calculated because the acute avian effects data shows no mortality and no sublethal effects at any of the test concentrations (*i.e.*, LD_{50} > 2000 mg/kg-bwt and LC_{50} > 5000 mg/kg-diet). Potential risks are qualitatively discussed in **Section 5.2**.

Potential direct chronic effects of trifluralin to the terrestrial-phase CRLF and SFGS are derived by considering dietary-based exposures modeled in T-REX for birds consuming small insects. EECs are divided by toxicity values to estimate chronic dietary-based RQs (NOAEC = 500 mg/kg-diet for mallard duck). The small insect RQ for the nursery scenario was 1.51, which exceeded the chronic LOC (RQ>1) (**Table 5.4**).

Based on exceedances of the Agency's chronic risk LOC (RQ > 1), trifluralin does have the potential to directly affect the CRLF and SFGS. Additionally, since relevant chronic RQs are exceeded, there is a potential for indirect effects to those listed species that rely on birds (and, thus, reptiles and/or terrestrial-phase amphibians) during at least some portion of their life-cycle (*i.e.*, CRLF, SFGS and SJKF).

Table 5.4 Summary of Chronic Dietary-based RQs for Birds for Spray Applications (not soil incorporated)[1]

Use Category	Application Rate (Interval days)	Dietary Category	**Avian Chronic Dietary based RQs** *Direct Effects for CRLF and SFGS (Small insects - avian dietary category)* *Indirect Effects for CRLF, SFGS and SJKF (all avian dietary categories listed below)*
Alfalfa	1 app @ 2 lbs/acre	Short Grass	0.96
		Tall Grass	0.44
		Broadleaf Plants/ Small Insects	0.54
		Fruits/Pods/ Seeds/ Large Insects	0.06
Nursery	3 app @ 4 lbs/acre (60 days)	Short Grass	**2.68**
		Tall Grass	**1.23**
		Broadleaf Plants/ Small Insects	**1.51**
		Fruits/Pods/ Seeds/ Large Insects	0.17

[1]Chronic RQ ≥ 1.0 exceeds chronic level of concern (LOC) for birds are bolded.

Granular applications (not soil incorporated)

Potential direct acute effects to the terrestrial-phase CRLF and SFGS are also evaluated by considering granule consumption (LD_{50}/sq-ft). LD_{50}s/sq-ft were calculated at single application rates of 1.5, 2, and 4 lbs/acre for three weight classes (20, 100, and 1000g individuals). RQs for the non-definitive acute oral endpoint for avian species were not calculated because the acute avian effects data shows no mortality and no sublethal effects at any of the test concentrations (*i.e.*, LD_{50} > 2000 mg/kg-bwt). Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**.

Spray and granular applications (soil incorporated)

For acute risk estimation, if the granular material is soil incorporated immediately after application, it is assumed that 1% of the material is available to terrestrial organisms (USEPA 1992). It was assumed that the availability of soil-incorporated spray material to terrestrial organisms would be similar to that of soil-incorporated granular pesticides. These EECs are derived by calculating 1% of the EECs based on the LD_{50}/sq ft exposure calculations. Therefore, the EEC for incorporated trifluralin at the highest single application rate of 2 lbs/acre is 0.21 mg/sq ft. Comparisons of exposure levels with highest acute dose tested and any potential risks are qualitatively discussed in **Section 5.2**.

5.1.2.2 Mammals

Potential risks to mammals are derived using T-REX, acute and chronic rat toxicity data, and a variety of body-size and dietary categories.

Spray applications (no soil incorporation)

Potential direct acute effects specifically to the SJKF are derived by considering dose- and dietary-based EECs modeled in T-REX for a large mammal (1,000 g) consuming a variety of dietary items (**Table 3.5**) and acute oral toxicity endpoints for rats. RQs for the non-definitive acute oral endpoint for mammalian species were not calculated because the acute mammalian effects data shows no mortality at the highest test concentration (*i.e.*, LD_{50} > 5000 mg/kg-bwt).). Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**.

Potential indirect acute effects to the terrestrial-phase CRLF, SFGS, and SJKF are derived by considering dose- and dietary-based EECs modeled in T-REX for all mammal sizes (15, 35, and 1000 g) consuming a variety of dietary items (**Table 3.5**) and acute oral toxicity endpoints for rats. RQs for the non-definitive acute oral endpoint for mammalian species were not calculated because the acute mammalian effects data shows no mortality at the highest test concentration (*i.e.*, LD_{50} > 5000 mg/kg-bwt). Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**.

Chronic RQs (dose-based) for mammals are derived with T-REX using a NOAEL of 10 mg/kg-bwt for rats. Exceeding RQs ranged from 1.30 to 58.20 for the small mammal (15 g), 1.11 to 49.72 for the medium mammal (35 g) and from 1.67 to 26.65 for the large mammal (1000 g) (**Table 5.5**). Two modeled scenarios for liquid applications exceeded the Agency's chronic risk LOC (RQ>1) for mammals for at least some of the food categories.

Chronic RQs (dietary-based) for mammals are derived with T-REX using a calculated NOAEC (200 mg/kg-diet) which is based on the chronic dose-based NOAEL of 10 mg/kg-bwt for rats.

Exceeding RQs (>1.0) ranged from 1.10 to 6.71 for the alfalfa and nursery scenarios (liquid applications) for mammals.

Based on exceedances of the Agency's chronic risk LOC (RQ>1), trifluralin has the potential to directly affect the SJKF. Additionally, since chronic RQs are exceeded, there is a potential for indirect effects to those listed species that rely on mammals during at least some portion of their life-cycle (*i.e.*, CRLF, SFGS, and SJKF).

Table 5.5 Summary of Chronic Dietary- and Dose-based RQs for Mammals for Spray Applications[1]

Use Category	Application Rate (Interval days)	Dietary Category	Direct Effects for SJKF (1000 g mammal only) Indirect Effects for SJKF (mammalian prey, all weight classes) Indirect Effects for CRLF and SFGS (mammalian prey, 15g only) Mammalian Chronic Dosed based RQs: 15 g	35 g	1000 g	Direct Effects for SJKF Indirect Effects for CRLF, SFGS and SJKF Mammalian Chronic Dietary based RQs
Alfalfa	1 app @ 2 lbs/acre	Short Grass	**20.82**	**17.79**	**9.53**	**2.40**
		Tall Grass	**9.54**	**8.15**	**4.37**	**1.10**
		Broadleaf Plants/ Small Insects	**11.71**	**10.00**	**5.36**	**1.35**
		Fruits/Pods/ Seeds/ Large Insects	**1.30**	**1.11**	0.60	0.15
		Granivore[2]	0.29	0.25	0.13	NA
Nursery	3 app @ 4 lbs/acre (60 days)	Short Grass	**58.20**	**49.72**	**26.65**	**6.71**
		Tall Grass	**26.68**	**22.79**	**12.21**	**3.07**
		Broadleaf Plants/ Small Insects	**32.74**	**27.97**	**14.99**	**3.77**
		Fruits/Pods/ Seeds/ Large Insects	**3.64**	**3.11**	**1.67**	0.42
		Granivore[2]	0.81	0.69	0.37	NA

[1]LOC exceedances (RQ $\geq$ 1) are **bolded**
[2] Indirect effects only occur for granivore dietary category all other dietary categories include both direct/indirect effects

Granular applications (not soil incorporated)

Potential direct acute effects to mammals are also evaluated by considering granule consumption (LD_{50}/sq-ft). LD_{50}s/sq-ft were calculated at single application rates of 1.5, 2, and 4 lbs/acre for three weight classes (15, 35, and 1000g individuals). RQs for the non-definitive acute oral endpoint for mammalian species were not calculated because the acute mammalian effects data shows no mortality and no sublethal effects at any of the test concentrations (*i.e*., LD_{50} > 5000 mg/kg-bwt). Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**.

Spray and granular applications (soil incorporated)

For acute risk estimation, if the granular material is soil incorporated immediately after application, it is assumed that 1% of the material is available to terrestrial organisms (USEPA 1992). It was assumed that the availability of soil-incorporated spray material to terrestrial organisms would be similar to that of soil-incorporated granular pesticides. These EECs are derived by calculating 1% of the EECs based on the LD_{50}/sq ft exposure calculations. Therefore, the EEC for incorporated trifluralin at the highest single application rate of 2 lbs/acre is 0.21 mg/sq ft. Comparisons of exposure levels with highest acute dose tested and any potential risks are qualitatively discussed in **Section 5.2**.

5.1.2.3 Terrestrial Invertebrates

In order to assess the risks of spray applications of trifluralin to terrestrial invertebrates, the honey bee is used as a surrogate for terrestrial invertebrates. The toxicity value for terrestrial invertebrates is calculated by multiplying the lowest available acute contact LD_{50} > 24.17 µg/bee by 1 bee/0.128g, which is based on the weight of an adult honey bee. EECs (µg/g of bee) calculated by T-REX for small and large insects are divided by the calculated toxicity value for terrestrial invertebrates, which is 188 µg/g of bee. It is important to note that the calculated RQs may overestimate risk as the LD_{50} values from all submitted bee studies were non-definitive (50% mortality was not reached at the highest dose). RQs for the non-definitive acute contact endpoint for terrestrial invertebrates were not calculated because the acute bee effects data did not allow for calculation of a definitive LD_{50} although mortality was observed at the highest test dose. Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**.

5.1.2.4 Terrestrial Plants

Generally, for indirect effects, potential effects on terrestrial vegetation are assessed using RQs from terrestrial plant seedling emergence and vegetative vigor EC_{25} data as a screen. All RQs are provided in **Appendix O**, and example output from TerrPlant v.1.2.2 is provided in **Appendix M**.

Based on the TerrPlant modeling results, there are LOC exceedances for 23 out of 25 uses for at least one of the application methods for risks to non-listed monocot plants. Fourteen out of 25 uses for at least one of the application methods resulted in LOC exceedances for non-listed dicot plants. RQs that exceed the LOC range from 1.11 to 4.89 for monocots and 1.05 to 2.32 for dicots **(Tables 5.6 and 5.7)**. Since the non-listed plant RQs are exceeded, there is a potential for indirect effects to those listed species that rely on terrestrial plants during at least some portion of their life-cycle (*i.e*., CRLF, SFGS, SJKF and DS).

Table 5.6 TerrPlant RQs for Monocots Inhabiting Dry and Semi-aquatic Areas Exposed to Trifluralin via Runoff and Drift from Liquid and Granular Applications (single application only); only scenarios with RQs exceeding the LOC are reported.

Scenario	Formulation[1]	Method[1*]	Application Rate (lb/acre)	Drift Value (%)	Dry Area RQ	Semi-aquatic Area RQ	Spray Drift RQ
Avocado Citrus Grapes and berries Olive Stone and Pome fruit Tree nuts Nursery Right of way Forestry	G	Ground	4	0	0.44	**4.44**	N/A
Alfalfa	G	Aerial * Ground *	2	0	0.11	**1.11**	N/A
Citrus Grapes Stone and Pome fruit Tree nuts Cotton Row crop Forestry	EC	Aerial* Ground*	2	5 1	**1.22** 0.33	**2.22** **1.33**	**1.11** 0.22
Alfalfa	EC	Aerial Ground	2	5 1	**1.33** 0.44	**3.33** **2.44**	**1.11** 0.22
Corn Cole Lettuce Wheat	EC	Aerial *	1	5	0.61	**1.11**	0.56
Rangeland Hay	G	Ground	2	0	0.22	**2.22**	N/A
Turf Residential	G	Ground	1.5	0	0.17	**1.67**	N/A
Nursery	EC	Ground	4	1	0.89	**4.89**	0.44

* 2 inch incorporation
[1]G = Granular application. EC = Emulsifiable Concentrate
LOC exceedances (RQ $\geq$ 1) are **bolded**

Table 5.7 TerrPlant RQs for Dicots Inhabiting Dry and Semi-aquatic Areas Exposed to Trifluralin via Runoff and Drift from Liquid and Granular Applications (single application only); only scenarios with RQs exceeding the LOC are reported							
Scenario	**Formulation[1]**	**Method[1*]**	**Application Rate (lb/acre)**	**Drift Value (%)**	**Dry Area RQ**	**Semi-aquatic Area RQ**	**Spray Drift RQ**
Avocado Citrus Grapes and berries Olive Stone and Pome fruit Tree nuts Nursery Right of way Forestry	G	Ground	4	0	0.21	**2.11**	N/A
Citrus Grapes Stone and Pome fruit Tree nuts Cotton Row crop Forestry	EC	Aerial* Ground*	2	5 1	0.58 0.16	**1.05** 0.63	0.53 0.11
Alfalfa	EC	Aerial Ground	2	5 1	0.63 0.21	**1.58** **1.16**	0.53 0.11
Rangeland Hay	G	Ground	2	0	0.11	**1.05**	N/A
Nursery	EC	Ground	4	1	0.42	**2.32**	0.21
* 2 inch incorporation [1]G = Granular application. EC = Emulsifiable Concentrate **Bolded** numbers exceed LOC=1							

5.1.3 Primary Constituent Elements of Designated Critical Habitat

For trifluralin 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 are used as the basis of the effects determination for potential modification to designated critical habitat.

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 trifluralin'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 trifluralin. A summary of the risk estimation results are provided in **Table 5.8** for direct and indirect effects to the listed species assessed here and in **Table 5.9** for the PCEs of their designated critical habitat.

Table 5.8 Risk Estimation Summary for Trifluralin - Direct and Indirect Effects

Taxa[(1)]	*LOC Exceedances (Y/N)*	*Description of Results of Risk Estimation*	*Assessed Species Potentially Affected*
Freshwater Fish and Aquatic-phase Amphibians	Y	Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. There are no exceedances of the Agency's chronic LOC (1.0) for freshwater fish.	Direct Effects: Aquatic-phase CRLF, DS Indirect Effects: Aquatic-phase CRLF, SFGS
Freshwater Invertebrates	N	There are no exceedances of the Agency's acute listed species LOC (0.05) for freshwater invertebrates. There are no exceedances of the Agency's chronic LOC (1.0) for freshwater invertebrates.	Indirect Effects: Aquatic-phase CRLF, SFGS, DS
Vascular Aquatic Plants	N	There are no exceedances of the Agency's LOC for aquatic vascular plants (1.0)	Indirect Effects: Aquatic-phase CRLF, SFGS, DS
Non-Vascular Aquatic Plants	N	There are no exceedances of the Agency's LOC for aquatic non-vascular plants (1.0)	Indirect Effects: Aquatic-phase CRLF, SFGS, DS
Birds, Reptiles, and Terrestrial-Phase Amphibians	Acute (not determined) Chronic (yes)	RQs for the non-definitive acute endpoints for avian species were not calculated because the acute avian effects data shows no mortality and no sublethal effects at any of the test concentrations. Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**. The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category (liquid application). Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e.*, short grass, tall grass and broadleaf plants and insects).	Direct Effects: Terrestrial-phase CRLF, SFGS Indirect Effects: Terrestrial-phase CRLF, SFGS, SJKF
Mammals	Acute (not determined) Chronic	RQs for the non-definitive acute endpoints for mammalian species were not calculated because the mammalian effects data shows no mortality and no sublethal effects at any of the test concentrations. Comparisons of exposure levels	Direct Effects: SJKF Indirect Effects:

Table 5.8 Risk Estimation Summary for Trifluralin - Direct and Indirect Effects			
Taxa[(1)]	*LOC Exceedances (Y/N)*	*Description of Results of Risk Estimation*	*Assessed Species Potentially Affected*
	(yes)	with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**. For liquid application, no soil incorporation,chronic dose-based RQs that exceed the chronic LOC all weight classes of mammals range from 9.53 to 58.20 for short grass; 4.37 to 26.68 for tall grass; 5.36 to 32.74 broadleaf plants/small insects; 1.11 to 3.64 for fruits/pods/seeds/large insect. RQs for all weight classes for granivores (nursery and alfalfa) and large mammals consuming fruits/pods/seeds/large insects (alfalfa) did not exceed the Chronic LOC. Chronic dietary-based RQs (liquid application, no soil incorporation) for mammals exceed the Agency's chronic LOC (1.0) for the nursery and alfalfa use categories with RQs ranging from 2.40 to 6.71 for short grass; 1.10 to 3.07 for tall grass and 1.35 to 3.77 for broadleaf plants/small insects. RQs for the fruits/pods/seeds/large insects feeding guild did not exceed the Chronic LOC.	Terrestrial-phase CRLF, SFGS, SJKF
Terrestrial Invertebrates	Not determined	RQs for the non-definitive acute endpoints for terrestrial invertebrates were not calculated because the acute avian effects data did not allow estimation of an LD_{50} (mortality <50% at all test concentrations). Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**.	Indirect Effects: Terrestrial phase CRLF, SFGS, SJKF
Terrestrial Plants - Monocots	Y	LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to non-listed monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots.	Indirect Effects: CRLF, SFGS, SJKF, DS
Terrestrial Plants - Dicots	Y	LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for non-listed dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots.	Indirect Effects: CRLF, SFGS, SJKF, DS
(1) Risks to estuarine/marine fish and estuarine/marine invertebrates were not assessed because the most sensitive freshwater fish and freshwater fish invertebrates toxicity data was use to estimate direct and indirect risks to the DS.			

Table 5.9 Risk Estimation Summary for Trifluralin– Effects to Designated Critical Habitat (PCEs) for CRLF and DS			
Taxa[1]	**May Affect Habitat (Y/N)**	**Description of Results of Risk Estimation**	**Species Associated with Designated Critical Habitat that May be Modified**
Freshwater Fish and Aquatic-phase Amphibians	Y	Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. There are no exceedances of the Agency's chronic LOC (1.0) for freshwater fish.	CRLF, DS
Freshwater Invertebrates	N	There are no exceedances of the Agency's acute listed species LOC (0.05) for freshwater invertebrates. There are no exceedances of the Agency's chronic LOC (1.0) for freshwater invertebrates.	CRLF, DS
Vascular Aquatic Plants	N	There are no exceedances of the Agency's LOC for vascular aquatic plants (1.0)	CRLF, DS
Non-Vascular Aquatic Plants	N	There are no exceedances of the Agency's LOC for non-vascular aquatic plants (1.0)	CRLF, DS
Birds, Reptiles, and Terrestrial-Phase Amphibians	Acute (not determined) Chronic (yes)	RQs for the non-definitive acute endpoints for avian species were not calculated because the acute avian effects data shows no mortality and no sublethal effects at any of the test concentrations. Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**. The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category (liquid application, no soil incorporation). Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e.*, short grass, tall grass and broadleaf plants and insects).	CRLF
Mammals	Acute (not determined) Chronic (yes)	RQs for the non-definitive acute endpoints for mammalian species were not calculated because the acute avian effects data shows no mortality and no sublethal effects at any of the test concentrations. Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**. For liquid application, no soil incorporation,chronic dose-based RQs that exceed the chronic LOC all weight classes of mammals range from 9.53 to 58.20 for short grass; 4.37 to 26.68 for tall grass; 5.36 to 32.74 broadleaf plants/small insects; 1.11 to 3.64 for fruits/pods/seeds/large insect. RQs for all weight classes for granivores (nursery and alfalfa) and large mammals	CRLF

Table 5.9 Risk Estimation Summary for Trifluralin– Effects to Designated Critical Habitat (PCEs) for CRLF and DS			
Taxa[(1)]	**May Affect Habitat (Y/N)**	**Description of Results of Risk Estimation**	**Species Associated with Designated Critical Habitat that May be Modified**
		consuming fruits/pods/seeds/large insects (alfalfa) did not exceed the Chronic LOC. Chronic dietary-based RQs (liquid application, no soil incorporation) for mammals exceed the Agency's chronic LOC (1.0) for the nursery and alfalfa use categories (liquid application) with RQs ranging from 2.40 to 6.71 for short grass; 1.10 to 3.07 for tall grass and 1.35 to 3.77 for broadleaf plants/small insects;.	
Terrestrial Invertebrates	Not determined	RQs for the non-definitive acute endpoints for terrestrial invertebrates were not calculated because the acute avian effects data did not allow estimation of an LD_{50} (mortality <50% at all test concentrations). Comparisons of exposure levels with highest dose tested and any potential risks are qualitatively discussed in **Section 5.2**.	CRLF
Terrestrial Plants - Monocots	Y	LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to non-listed monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots.	CRLF, DS
Terrestrial Plants - Dicots	Y	LOCs exceeded for 10 out of 25 modeled uses for at least one of the application methods for non-listed dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots.	CRLF, DS

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 **Sections 5.2.1 through 5.2.4**. 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 trifluralin.

5.2.1 California Red-legged Frog

5.2.1.1 Direct Effects (aquatic-phase)

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

Based on the evidence presented below, there is a potential for direct impact to the aquatic-phase CRLF.

LOC exceedances:
Of the 25 scenarios modeled, for at least one of the application methods the listed species LOC was exceeded in almond, avocado, citrus, fruit, grape (including berries), grape, olive, alfalfa, corn, cotton, lettuce, onion, cole, row crop, sugar beet, wheat, nursery and forestry (exceeding RQs ranged from 0.05 to 0.35). The Chronic LOC was not exceeded for any of the modeled scenarios.

Probability of individual effect:
For all modeled scenarios, the likelihood of an individual effect (mortality) was $\leq$1 in 3280 at the calculated RQ and at the Listed Species LOC.

Species sensitivity analysis:
A formal species sensitivity analysis was not conducted, since data from only two species of fish were available for this assessment. For the bluegill sunfish and rainbow trout, the LD_{50}'s ranged from 18.5 to 43.6 µg/L. Nominal concentrations were reported for these two studies; therefore, both LD_{50}s may overestimate the actual exposure concentration due to trifluralin's tendency to volatilize and sorb to surfaces.

Analysis of aquatic-phase amphibian data:
As previously discussed in the aquatic toxicity portion of this assessment (**Section 4.1**), aquatic amphibian data were submitted by the registrant and available in the open literature. However, since these data were less sensitive than the freshwater fish data and it is unknown where the CRLF falls on sensitivity distribution for aquatic-phase amphibians or on a sensitivity distribution for aquatic vertebrates, EFED determined that using the most sensitive aquatic vertebrate toxicity data would be appropriate. In order to characterize risk, the most sensitive amphibian toxicity value for trifluralin (Fowler's toad with LC_{50} = 100 µg/L, Sanders (1970, E2891) was used to calculate RQs (**Appendix O**). The Listed Species LOC was exceeded for at least one application method and rate for ten of the modeled scenarios with exceeding RQs ranging from 0.05 to 0.07. It is important to note that this analysis may not reflect the sensitivity of the CRLF to trifluralin as the relative sensitivity of the CRLF to Fowler's toads is unknown. In addition, the LD_{50} and, thus, the RQs are based nominal concentrations of trifluralin. Since trifluralin is volatile and sorbs to surfaces, these concentrations from the toxicity study and, thus, the calculated LD_{50} are likely higher than the actual exposure concentration experienced by the tested animals. If the actual exposure concentrations in the toxicity study were less than the nominal concentrations used for calculating the LD_{50}, the calculated frog RQs would be higher if actual exposure concentrations were used.

Sublethal chronic effects:
Fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. In a study conducted by Couch (1979, E6425), sheepshead minnows exposed to 5.5 to 31 µg/L of trifluralin during the first 28 days of life developed a vertebral dysplasia. Effects of the abnormal vertebral development were dorsal vertebral growth into the neural canal, ventral compression of renal ducts, and longitudinal fusion of vertebrae. In a second study (Couch 1984, E48406), pituitaries of sheepshead minnows, exposed for 19 months to 1-5 µg/L trifluralin in the laboratory exhibited enlargement, pseudocysts, congestion of blood vessels and oedema. Most of the fish with an enlarged pituitary also had induced diffuse and/or focal vertebral hyperostosis and other dysplastic vertebral changes. Koyama (1996, E17085) also observed vertebral deformities when exposing 10 species of fish to trifluralin with lesions observed in concentrations as low as 5 to 30 µg/L, depending on species. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS.

Comparison of modeled to observed water concentrations:
Data from the CDPR surface water monitoring database website for trifluralin occurrences were obtained on Jul 15, 2009 (http://www.cdpr.ca.gov/docs/emon/surfwtr/surfcont.htm). A total of 3,915 surface water samples were analyzed for trifluralin from 1992 to 2006. Trifluralin was detected in 600 samples (15% detection rate). Of the positive samples, the average concentration was 0.06 µg/L and the highest reported concentration was 1.74 µg/L. Because this database consists of data from a multitude of studies conducted in California, the LOQ is not consistent across studies. The variation in the LOQ increases the uncertainty in the trifluralin detection rate of 15% and adds uncertainty to the calculated average trifluralin concentration. These measured concentrations are roughly comparable to the PRZM/EXAMS peak concentrations which range

from 0.0002 to 6.55 µg/L. Monitoring data may not reflect the actual maximum exposure concentrations for several reasons:

- Monitoring sites were not necessarily targeted to areas of high trifluralin usage
- Sampling times were not designed to capture the high peak values and were not frequent enough to assure the peak values were captured
- Water samples were filtered prior to analysis, removing trifluralin that had sorbed to suspended particles in the water.

Even though these measured concentrations may not capture true peak values that occur in California waters, if the observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09).

Bioaccumulation in aquatic prey items:
As discussed in **Section 3.4**, trifluralin has the potential to accumulate in tissues of aquatic organisms. Since the CRLF consume algae, aquatic invertebrates and fish, they could be exposed to trifluralin accumulated in the tissues of these prey. Acute RQs were not calculated as the avian acute toxicity studies resulted in no mortalities or sublethal effects at levels higher than those expected in the environment. Using KABAM, the chronic RQ values for the CRLF for all size classes did not exceed the LOC for chronic dietary-based exposures through consumption of contaminated aquatic prey that have accumulated trifluralin (**Table 5.3**, all RQs < 0.05).

In a 28-day laboratory BCF study with bluegill sunfish (*Lepomis macrochirus*) exposed to trifluralin, a BCF of 5,674 was observed in whole fish. The estimated steady-state BCF in KABAM for large fish was 26,226 which is a factor of 4.6 of the laboratory BCF for fish. The accumulation and depuration rates of trifluralin in fish cannot be fully assessed because radioactive residues in fish tissues were incompletely characterized. In addition, other chemical properties of trifluralin, such as its volatility and short aqueous photolysis half-life, may mediate the bioaccumulation in the environment.

Spray drift buffers:
The buffer distance needed to get below the acute aquatic fish Listed Species LOC was estimated using AgDRIFT. This distance identifies those locations where water bodies can be impacted by spray drift deposition alone (no runoff considered) resulting in concentrations above the LOC. The most sensitive freshwater fish was the bluegill with an LC_{50} value of 18.5µg/L. Several labels were used to represent the range of application methods (ground and aerial), application rates (0.8 to 4.0 lbs/acre), and drop size distribution specifications. Required buffer distance under all evaluated labels ranged from 45.9 to 2359 feet (**Table 5.10**). These required buffer distances only consider the exposure due to spray drift. Runoff (an important exposure component for trifluralin) is not assessed in this spray drift analysis conducted with AgDRIFT. This spray drift analysis does not include reproductive or growth effects or cumulative exposure due to multiple applications.

Table 5.10. Estimation of Buffer Distance Required to Eliminate LOC Exceedances (only spray drift exposure considered) for Freshwater Fish Based on AgDRIFT					
Pesticide Label[1]	**Application Rate (lb/acre)**	**Method (Boom Height for Ground)**	**Tier**	**Parameters**	**Required Buffer Distance (ft)**
Treflan EC 062719-00097	4.0	Ground (high)	I	DSD = ASAE very fine to fine	387.13
Treflan HFP 062719-00250	2.0	Ground (high)	I	DSD = ASAE very fine to fine	187.01
Treflan HFP 062719-00250	1.0	Ground (high)	I	DSD = ASAE very fine to fine	68.9
Treflan HFP 062719-00250	0.8	Ground (high)	I	DSD = ASAE very fine to fine	45.9
Treflan HFP 062719-00250	2.0	Aerial	III[2]	DSD = ASAE Very fine to fine Nonvol rate = 4.65 lb/ac Active rate = 2.0 lb/ac Spray vol rate = 5 gal/ac Specific gravity(nonvol) = 1.116 Max downwind dist = 3000 ft All else = defaults	2264
Treflan HFP 062719-00250	1.0	Aerial	III[2]	DSD = ASAE Very fine to fine Nonvol rate = 2.325 lb/ac Active rate = 1.0 lb/ac Spray vol rate = 5 gal/ac Specific gravity(nonvol) = 1.116 Max downwind dist = 3000 ft All else = defaults	2185
Treflan HFP 062719-00250	0.8	Aerial	III[2]	DSD = ASAE Very fine to fine Nonvol rate = 1.86 lb/ac Active rate = 0.8 lb/ac Spray vol rate = 5 gal/ac Specific gravity(nonvol) = 1.116 Max downwind dist = 3000 ft All else = defaults	2106

[1]Label did not specify droplet sizes; the most conservative size available was chosen: ASAE very fine to fine. Input values were calculated as follows: (LC_{50} value of 18.5µg/L =18500 ng) (initial average concentration =18500 X aquatic LOC of 0.05= 925 ng/l)
[2] Tier I and Tier II resulted in distance estimates that were 'out of range'; Tier III estimates were used for the aerial scenarios.

5.2.1.2 Direct Effects (Terrestrial-Phase)

The terrestrial-phase of the CRLF considers juvenile and adult life stages during which much time is spent in a terrestrial habitat. Submerged terrestrial-phase CRLFs are not considered here; their exposure is addressed as an aquatic-phase CRLF. Since no toxicity data were available for terrestrial-phase amphibians or reptiles, toxicity data for birds were used as a surrogate.

Acute risks from spray applications:
RQs for the non-definitive acute oral and subacute dietary toxicity endpoints for avian species were not calculated because the acute avian effects data shows no mortality and no sublethal effects at any of the test levels (*i.e.*, LD_{50} > 2000 mg/kg-bwt and LC_{50} > 5000 mg/kg-diet). With no soil incorporation, the dose-based EECs range from 1.94 to 1528.09 mg/kg-bwt, and dietary-based EECs range from 30 to 1342 mg/kg-diet. These values are lower than the LD_{50} and LC_{50}, respectively; therefore, acute dose-based and acute dietary-based risks from spray applications are unlikely. Since exposure from soil incorporated applications would be less than from non-incorporated applications, risks from incorporated applications are also unlikely. The probability of individual effect (mortality) for avian species was not calculated because the acute avian effects data shows no mortality and no sublethal effects were observed at any of the test concentrations (*i.e.*, LD_{50} > 2000 mg/kg-bwt and LC_{50} >5000 mg/kg-diet).

Acute risks from granular applications:
Potential direct acute effects to the terrestrial-phase CRLF are also evaluated by considering granule consumption (LD_{50}/sq-ft). Toxicity values were calculated for three weight classes (20, 100, and 1000g individuals) based on the highest dose tested (2000 mg/kg-bwt in mallard ducks, see TREX 1.4.1 Users Guide for details). The calculated toxicity values are >21, >130, and >1560 mg/bird. The 20 and 100 g results are relevant for evaluating direct effects and indirect effects (effects to prey base) of the terrestrial-phase CRLF. The EECs at single non-incorporated application rates of 1.5, 2, and 4 lbs/acre were 15.6, 20.8, and 41.7 mg/sq-ft, respectively, and the EEC at a single soil-incorporated application rate of 2 lbs/acre is 0.21mg/sq-ft. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded.

Chronic risks - LOC exceedances:
Of the two scenarios modeled for liquid applications, one scenario (nursery use, exceeding RQ = 1.51) resulted in an RQ that exceeded the Chronic LOC.

Chronic risks - T-HERPS refinements:
A refinement of the risks posed to the terrestrial-phase CRLF from ingestion of residues on prey items (based on liquid applications of trifluralin) was performed using the T-HERPS v. 1.0 model. T-HERPS was used to refine chronic risks to the terrestrial-phase CRLF via consumption of large insects, small herbivorous mammals, small insectivorous mammals, and small terrestrial-phase amphibians exposed to liquid applications already identified by T-REX. In T-HERPS, the nursery scenario with three applications of 4 lbs/acre exceeded the chronic LOC (1.0) with RQs of 1.77 and 1.51 for herptiles that consume small herbivore mammals and small insects, respectively. Dietary-based chronic RQ values that exceed the LOC (1.0) are provided in **Table 5.11**. All RQ values are reported in **Appendix O**.

T-HERPS can also be used to refine acute risks to herptiles; however, for this assessment, no acute risks were identified. Therefore, refinements to acute risks were not assessed.

Table 5.11 Summary of dietary-based RQs for herpetofauna estimated based on the maximum trifluralin spray application using T-HERPS version 1.0.				
Use Category	**Application Rate (Interval)**	**Dietary Category**	**EEC**	**Chronic Dietary- Based RQ[1]**
Nursery	3 app @ 4 lbs/acre (60 days)	Broadleaf Plants/Small Insects[2]	754.72	**1.51***
		Fruits/Pods/Seeds/Large Insects[2]	83.86	0.17
		Small Herbivore Mammals[3]	884.12	**1.77***
		Small insectivorous mammals[4]	55.26	0.11
		Small terrestrial-phase amphibians[5]	26.20	0.05

[1] RQ values in bold indicate exceedance the chronic LOC (1.0). Chronic dietary-base RQs were based on dietary-based EECs divided by avian NOAEC value of 500 mg/kg-diet for mallard duck

[2] Dietary-based EECs for Small and Large Insects calculated by T-HERPS are the same as those calculated by T-REX. Resulting RQs are also identical (see **Table 5.4**).

[3] Dietary-based EECs for Small Herbivore Mammals are derived by assuming that a small mammal (35 g) consumes contaminated Short Grass prior to being consumed by the assessed herptile.

[4] Dietary-based EECs for Small Insectivorous Mammals are derived by assuming that a small mammal (35 g) consumes contaminated Large Insects prior to being consumed by the assessed herptile.

[5] Dietary-based EECs for Small Terrestrial-phase Amphibians are derived by assuming that a small herptile (2.3 g) consumes contaminated Small Insects prior to being consumed by the assessed herptile.

Chronic risks - Soil invertebrate consumption:
Chronic direct effects to terrestrial-phase CRLF that may consume soil invertebrates that have accumulated trifluralin residues in their tissues were modeled; however, this pathway is not included in T-REX. In order to explore the potential exposures of mammals and birds to total residues of trifluralin that have accumulated in soil invertebrates inhabiting trifluralin treatment sites, a simple fugacity approach was employed to estimate trifluralin concentrations in earthworms and subsequent exposures to mammals and birds consuming earthworms. Earthworms were chosen to represent all soil invertebrates that may be consumed by the terrestrial organisms under evaluation. This approach is explained in detail in **Appendix P**.

Using a single application rate of 4 lbs/acre, PRZM estimated trifluralin concentration are 6.41 g/m^3 in soil, and the estimated concentration of trifluralin in earthworms is 147 mg/kg-earthworm. If it is assumed that a bird or herptile consumes only contaminated soil invertebrates, its dietary-based exposures would be approximately 30% of the NOAEC in the avian reproduction study (NOAEC=500 mg/kg-diet). Therefore, chronic risks to terrestrial-phase CRLF from consumption of trifluralin-contaminated soil invertebrates are unlikely.

5.2.1.3 Indirect Effects (via Reductions in Prey Base)

5.2.1.3.1 Algae (non-vascular plants)

As discussed in **Section 2.5.3**, the diet of CRLF tadpoles is composed primarily of unicellular aquatic plants (*i.e.*, algae and diatoms) and detritus.

LOC exceedances:
All RQs for aquatic non-vascular plants are ≤ 0.30 (no LOC exceedances).

Comparison of modeled to observed water concentrations:
As the EECs estimated for aquatic plants are the same as for freshwater fish, the discussion presented in **Section 5.2.1.1** is also applicable here. In summary, of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates.

Spray drift buffers:
Spray drift buffers were not determined as there were no exceedances of the LOC at maximum label rates for all scenarios.

5.2.1.3.2 Aquatic Invertebrates

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

Based on the evidence presented below, indirect impacts to aquatic-phase of the CRLF through reductions in the prey base (specifically aquatic invertebrates) are not expected from acute or chronic exposure to trifluralin.

LOC exceedances:
No acute or chronic LOC exceedances occurred for freshwater invertebrates with acute RQs <0.05 and chronic RQs < 1.0 for all 25 application scenarios.

Percent effect analysis:
A percent effect analysis was conducted by determining an expected percent effect on the prey item (aquatic invertebrates) at the Listed Species LOC of 0.05, implying effect at the calculated EEC. The Listed Species LOC was used since there were no RQs exceeding 0.05, and a default slope of 4.5 was used as one was not available from the submitted daphnid study. The percent effect to aquatic invertebrates at the Listed Species LOC was calculated as 0.0000002%.

Comparison of modeled to observed water concentrations:
As the EECs estimated for freshwater invertebrates are the same as for freshwater fish, the discussion presented in **Section 5.2.1.1** is also applicable here. In summary, of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates.

5.2.1.3.3 Fish and Aquatic-phase Frogs

Indirect effects to fish and frogs as food items are based on the direct effects analysis for aquatic-phase CRLFs (**Section 5.2.1.1**). There is a potential for indirect impact to the aquatic-phase of the CRLF due to reduction of fish and aquatic-phase frogs as a prey base based on the summarized lines of evidence below.

- There were Listed Species LOC exceedances in 18 of the 25 modeled scenarios.
- For all modeled scenarios, the likelihood of an individual effect (mortality) was $\leq$1 in 3280 at the calculated RQ.
- A formal species sensitivity analysis was not conducted; available data provided a range of LD_{50}'s from 18.5 to 43.6 µg/L for two species.
- Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates.
- Available aquatic-phase amphibian data suggests trifluralin is less toxic to aquatic-phase amphibians than to fish; however, there are Listed Species LOC exceedances for the most sensitive aquatic-phase amphibian data.
- Fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS.
- Trifluralin has the potential to accumulate in tissues of aquatic organisms. The resulting RQ values for the three size classes of frogs did not exceed the LOC for chronic dietary-based exposures through consumption of aquatic prey that have accumulated trifluralin. Acute RQs were not calculated as the avian acute toxicity studies resulted in no mortalities at levels higher than those expected in the environment.
- The spray drift buffer distance needed to get below the acute aquatic fish Listed Species LOC ranged from 45.9 to 2359 feet depending on application rate and method. These required buffer distances only considers the exposure due to spray drift; based on EFED's modeling, spray drift exposure alone does not cause the LOC to be exceeded. Runoff is not assessed in this spray drift analysis.

5.2.1.3.4 Terrestrial Invertebrates

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

For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortality at this dose, MRID 05001991). The toxicity value for terrestrial invertebrates is calculated by multiplying the LD_{50} > 24.17 µg/bee by 1 bee/0.128g, which is based on the weight of an adult honey bee, resulting in a toxicity estimate of 188 µg/g-bee (equivalent to 188 mg/kg-bee). Since definitive LD_{50} values were not available, RQs were not calculated. However, expected exposure levels were compared to the highest dose tested to evaluate the likelihood of risk. Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Therefore, EECs must be less than 9.4 mg/kg-insect (188 ÷ 20) in for risks to be considered unlikely.

EECs for small insects ranged from 135 mg/kg-insect at 1 lb/acre application to 755 mg/kg-insect at 4 lbs/acre with three (60-day interval) applications, and EECs for large insects ranged from 15 mg/kg-insect at 1 lb/acre application to 84 mg/kg-insect at 4 lbs/acre with three (60-day interval) applications (**Table 3.10**). Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios.

5.2.1.3.5 Mammals

Life history data for terrestrial-phase CRLFs indicate that large adult frogs consume terrestrial vertebrates, including mice.

Acute risks from spray applications:
RQs for the non-definitive acute oral toxicity endpoints for mammalian species were not calculated because the acute effects data shows no mortality and no sublethal effects at any of the test levels (*i.e.*, LD_{50} > 5000 mg/kg-bwt). With no soil incorporation, the dose-based EECs range from 1.02 to 1279.2 mg/kg-bwt. These values are lower than the LD_{50}; therefore, acute dose-based risks from spray applications are unlikely. Since exposure from soil incorporated applications would be less than from non-incorporated applications, risks from incorporated applications are also unlikely. The probability of individual effect (mortality) for mammalian species was not calculated because the acute mammalian effects data shows no mortality and no sublethal effects were observed at any of the test concentrations (*i.e.*, LD_{50} > 5000 mg/kg-bwt).

Acute risks from granular applications:
Potential direct acute effects to mammals are also evaluated by considering granule consumption (LD_{50}/sq-ft). Toxicity values were calculated for three weight classes (15, 35, and 1000g individuals) based on the highest dose tested (5000 mg/kg-bwt, see TREX 1.4.1 Users Guide for details). The calculated toxicity values are >167, >321, and >4170 mg/mammal. The 15 and 35 g results are relevant for evaluating indirect effects (effects to prey base) of the terrestrial-phase CRLF. The EECs at single non-incorporated application rates of 1.5, 2, and 4 lbs/acre were 15.6, 20.8, and 41.7 mg/sq-ft, respectively, and the EEC at a single soil-incorporated application rate of 2 lbs/acre is 0.21 mg/sq-ft. Risks to mammals from granular applications of trifluralin are unlikely as the calculated EECs do not exceed the toxicity values.

Chronic risks from spray applications (non-incorporated):
Chronic dose-based RQ values representing trifluralin exposures to small mammals indicate risks resulting from both modeled scenarios (**Table 5.5**) for all feeding guilds except granivores. Chronic diet-based RQ values representing trifluralin exposures to mammals indicate risks resulting from both modeled scenarios for all feeding guilds except granivores and fruits/pods/seeds/large insects.

5.2.1.3.6 Terrestrial-phase Frogs

Terrestrial-phase adult CRLFs also consume small frogs. RQ values, estimated using T-REX, representing direct exposures of trifluralin to terrestrial-phase CRLFs are used to represent exposures of trifluralin to small frogs in terrestrial habitats. The indirect effects to frogs as food items are based on the direct effects analysis for the terrestrial-phase CRLF (**Section 5.2.1.2**). Indirect impacts to CRLFs through reductions in the prey base (specifically terrestrial-phase frogs) are expected from chronic exposure to trifluralin based on the summarized lines of evidence below:

- Acute risks from spray applications are unlikely since the EECs are lower than the LD_{50} and LC_{50} and no mortality or sublethal effects were noted in the acute bird studies.
- Acute risks from granular applications cannot be precluded at the highest application rate (4 lbs/acre with no soil incorporation) for 20 g individuals.
- One scenario (nursery use, exceeding RQ = 1.51) resulted in an RQ that exceeded the Chronic LOC.
- Chronic risks were refined with T-HERPS, resulting in LOC exceedances for the nursery scenario with three applications at 4 lbs/acre. RQs exceeded the chronic LOC (1.0) with values of 1.77 and 1.51 for herptiles that consume small herbivore mammals and small insects, respectively.
- Based on the fugacity model, chronic exposure of herptiles to trifluralin through consumption of contaminated soil invertebrates from fields treated with trifluralin is unlikely.

5.2.1.4 Indirect Effects (via Habitat Effects)

5.2.1.4.1 Aquatic Plants (Vascular and Non-vascular)

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

Potential indirect effects to the CRLF based on impacts to habitat and/or primary production were assessed using RQs from freshwater aquatic vascular and non-vascular plant data. Based on the evidence presented in **Section 5.2.2.1**, indirect impacts to aquatic-phase of the CRLF due to reduction effects on its habitats through impact on vascular and non-vascular aquatic plants are not expected from acute exposure to trifluralin.

5.2.1.4.2 Terrestrial Plants

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

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

Trifluralin is a synthetic fluorinated dinitroaniline preemergent herbicide that enters plants through developing roots preventing the alignment and separation of chromosomes during mitosis (mitosis disruptor) which typically results in the swelling of root tips as cells in this region fail to divide or elongate. Trifluralin is readily absorbed by young roots. Signs of toxicity to post-emergent broadleaf weeds include localized chlorosis and stunting.

Based on the available toxicity data for terrestrial plants (using Treflan HFP formulation, MRID 439844-01, 419345-03), it appears that monocots and dicots in the seedling emergence studies are typically more sensitive to trifluralin via soil or root uptake than monocots and dicots in the vegetative vigor studies via foliar exposure.

Riparian vegetation typically consists of three tiers of vegetation, which includes a groundcover of grasses and forbs, an understory of shrubs and young trees, and an overstory of mature trees. Frogs spend a considerable amount of time resting and feeding in riparian vegetation; the moisture and cover of the riparian plant community provides good foraging habitat and may facilitate dispersal in addition to providing pools and backwater aquatic areas for breeding (USFWS 2002). According to Hayes and Jennings (1988), the CRLF tends to occupy water bodies with dense riparian vegetation including willows (*Salix* sp.). Upland habitat includes grassland and woodlands, as well as scrub/shrub habitat. No guideline data or open literature studies are available regarding the toxicity of trifluralin to woody plants. Because trifluralin may be applied near woody species, toxicity to the woody part of the plant, excluding green bark, is an uncertainty.

As shown in **Tables 5.4** and **5.5**, RQ values exceeded LOCs for monocots and dicots inhabiting dry and semi-aquatic areas exposed to liquid and granular formulations of trifluralin via runoff and drift. Spray drift RQ values exceeded LOCs for liquid aerial applications at a single application rate of 2.0 lbs/acre.

Based on exceedances of the terrestrial plant LOCs for 23 out of 25 trifluralin modeled uses (for at least one of the application methods) following runoff and spray drift to dry and semi-aquatic areas, the following general conclusions can be made with respect to potential harm to riparian habitat:

- Trifluralin may enter riparian areas via runoff and/or spray drift where it may contact foliar surfaces of emerged seedlings or form a chemical barrier on soil, which would affect pre-emergent plants.
- Based on trifluralin's mode of action and a comparison of seedling emergence and vegetative vigor EC_{25} values to EECs estimated using TerrPlant, emerging or developing seedlings may be affected in areas receiving both runoff and drift and in areas receiving drift alone at applications rates greater than a single application of 0.8 lbs/acre. Furthermore, based on the residual nature of trifluralin, it is possible that impacts to germinating seedlings and emerging plants would occur for several months after application. If inhibition of new growth occurs, it 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.
- Because nine out of ten of the species tested in the seedling emergence studies and all ten species tested in the vegetative vigor studies were affected, it is likely that many species of herbaceous plants may be potentially affected by exposure to trifluralin via runoff and spray drift.

A review of trifluralin incidents for terrestrial plants that were reported in the EIIS database indicated that there were 78 incidents involving terrestrial plants. However, none of these incidents reported effects to wild plants. Most incidents involved spray drift or direct treatment(e.g., Alfalfa, barley, bean, birch, blue spruce, corn, cotton, dry bean, ornamentals, peanut, percifia shrubs, pinto bean, potato, raspberry, rose, soybean, soybean seed, spreading yew, sudan grass, sugarcane, sunflower, tomato and wheat (spring and other varieties). The absence of reports of adverse effects on wild terrestrial plants does not provide evidence of an absence of incidents and, consequently, risk.

In summary, terrestrial plant RQs exceed LOCs, which indicates risk to upland and riparian vegetation. However, while it is not expected that woody plants with mature bark are sensitive to environmentally relevant trifluralin concentrations, the lack of a guideline study on established woody plants precludes estimation of effects. In addition, several incidents reported damage to woody plants including shrubs, raspberries, rose, and spreading yew. Therefore, the potential of effects to woody plants cannot be precluded. Because upland and riparian areas are comprised of a mixture of both woody plants and herbaceous vegetation, terrestrial-phase CRLFs may be indirectly affected by adverse effects solely to herbaceous vegetation, which provides habitat and cover for the CRLF and its prey. Therefore, the indirect effect to the terrestrial-phase CRLF via reduction in terrestrial plants is neither insignificant nor discountable.

Spray drift RQ values did not exceed LOCs for ground (liquid) and granular formulations for all application scenarios. Only one aerial (liquid) application scenario for monocots exceeded the LOC of 1.

In order to estimate buffer distances that are protective of plant species that the terrestrial-phase CRLF, SFGS and SJKF or its prey may depend on for food and cover, AgDRIFT was used to model the dissipation distance to the EC_{25} levels for terrestrial plants.

Because trifluralin is used as a pre-emergent and post-emergent herbicide, buffer distances were calculated for the most sensitive endpoints for both monocots and dicots in the seedling emergence and vegetative vigor studies Spray drift RQ values did not exceed LOCs for ground (liquid) and granular formulations for all application scenarios. Only one aerial application scenario for monocots exceeded the LOC for a liquid application scenario (**Table 5.12**). For established monocots, the calculated buffer distances for seedling emergence and vegetative vigor were 55.7 feet and 6.56 feet, respectively.

Table 5.12 Estimation of Buffer Distance Required to Eliminate LOC Exceedances (only spray drift exposure considered) for Terrestrial Plants Based on AgDRIFT

Pesticide Label	App. Rate (lb/acre)	Method (Boom Height for Ground)	Tier	Parameters*	Required Buffer Distance (ft)	
					Seedling Emergence	Vegetative Vigor
					Monocot	Monocot
Treflan HFP 062719-00250	2.0	Aerial	I	DSD = ASAE very fine to fine	55.7	6.56

*Fraction of applied input values for AGDRIFT:
For seedling emergence EC_{25} = 0.09 lbs/acre; Fraction of the applied = EC_{25} ÷ Rate = 0.045
For vegetative vigor EC_{25} = 1.09 lbs/acre; Fraction of the applied = EC_{25} ÷ Rate = 0.545

5.2.1.5 Modification to Designated Critical Habitat

5.2.1.5.1 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. There is a potential for habitat modification via impacts to terrestrial plants (**Section 5.2.3.2**)

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

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

As discussed above, there is potential for habitat modification of the terrestrial-phase CRLF via impacts to terrestrial plants as indicated by potential impacts to herbaceous vegetation, which provides habitat, cover, and a means of dispersal for the terrestrial-phase CRLF and its prey. This habitat modification could be caused by all modeled uses of triflualin at the maximum labeled rate.

The third terrestrial-phase PCE is "reduction and/or modification of food sources for terrestrial phase juveniles and adults." To assess the impact of trifluralin on this PCE, acute toxicity endpoints for terrestrial invertebrates and acute and chronic toxicity endpoints for mammals and terrestrial-phase frogs are used as measures of effects. Based on the characterization of indirect effects to the terrestrial-phase CRLF via reduction in prey base (**Section 5.2.2.4** for terrestrial invertebrates, **Section 5.2.2.5** for mammals, and **Section 5.2.2.6** for frogs), there is potential for critical habitat modification via a reduction of terrestrial invertebrates, small mammals, and frogs as food items.

The fourth terrestrial-phase PCE is based on alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source. As discussed in

Section 5.2.1.2, direct acute effects to the terrestrial-phase CRLF are unlikely. However, direct chronic effects to the terrestrial-phase CRLF are likely for liquid applications of trifluralin at rate of three applications of 4.0 lbs/acre. Indirect effects to the terrestrial-phase CRLF via reduction in prey base are likely. Therefore, there is potential for habitat modification via direct and indirect effects to the terrestrial-phase CRLF.

5.2.2 Delta Smelt

To assess potential direct effects of trifluralin to the DS, toxicity data for both the freshwater fish and estuarine/marine fish were evaluated since the DS lives in brackish waters. Since freshwater fish were more sensitive than estuarine/marine fish, the freshwater fish toxicity data were used to assess potential direct effects of trifluralin to the DS.

To assess potential indirect effects (prey) of trifluralin to the DS, toxicity data for both the freshwater invertebrates and estuarine/marine invertebrates were evaluated since the DS lives in brackish waters. Since freshwater invertebrates were more sensitive than estuarine/marine invertebrates, the freshwater invertebrate toxicity data were used to assess potential indirect effects of trifluralin to the DS.

5.2.2.1 Direct Effects

RQ values representing direct effects of trifluralin to aquatic-phase CRLF (**Section 5.2.1.1**) are also used to represent potential effects of trifluralin to the DS. There is a potential for direct impact to the DS based on the summarized lines of evidence below:

- There were Listed Species LOC exceedances in 18 of the 25 modeled scenarios.
- For all modeled scenarios, the likelihood of an individual effect (mortality) was $\leq$1 in 3280 at the calculated RQ and at the Listed Species LOC.
- A formal species sensitivity analysis was not conducted; available data provided a range of LD_{50}'s from 18.5 to 43.6 µg/L for two species.
- Of the 3,915 non-targeted water samples tested for trifluralin, 15% had detectible concentrations of trifluralin; those concentrations where trifluralin was detected were comparable to the PRZM/EXAMS estimates. If the observed maximum concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09).
- The spray drift buffer distance needed to get below the acute aquatic fish Listed Species LOC ranged from 45.9 to 2359 feet depending on application rate and method. These required buffer distances only considers the exposure due to spray drift; based on EFED's modeling, spray drift exposure alone does not cause the LOC to be exceeded.
- The Chronic LOC was not exceeded for any scenario.
- Fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS.

5.2.2.2 Indirect Effects (*via Reductions in Prey Base*)

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).

RQ values representing exposures of trifluralin to freshwater invertebrates that may serve as prey for the aquatic-phase CRLF are also used to represent exposures of trifluralin to freshwater invertebrates that may serve as prey for the DS. No acute or chronic LOC exceedances occurred for freshwater invertebrates with acute RQs <0.05 and chronic RQs < 1.0 for all 25 application scenarios.

Based on the multiple lines of evidence as described in **Section 5.2.1.3.2**, impacts to potential invertebrate prey are not expected from acute or chronic exposure to trifluralin.

5.2.2.3 Indirect Effects (*via Habitat Effects*)

5.2.2.3.1 Aquatic Plants (Vascular and Non-vascular)

Aquatic plants serve several important functions in aquatic ecosystems. Non-vascular aquatic plants are primary producers and provide the autochthonous energy base for aquatic ecosystems. Vascular plants provide structure, rather than energy, to the system, as attachment sites for many aquatic invertebrates, and refugia for juvenile organisms, such as fish and frogs. Emergent plants help reduce sediment loading and provide stability to nearshore areas and lower streambanks. In addition, vascular aquatic plants are important as attachment sites for egg masses of aquatic species. Results of the indirect effects assessment are used as the basis for the habitat modification analysis.

No acute LOC exceedances occurred for aquatic plants (vascular and non-vascular) with RQs < 1.0 for all application scenarios. Based on the multiple lines of evidence as described in **Section 5.2.1.4.1**, impacts to aquatic plants found near trifluralin use sites are not expected.

5.2.2.3.2 Terrestrial Plants

Terrestrial plants serve several important habitat-related functions for the DS. Among other things, 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.

Based on the results of the submitted terrestrial plant toxicity studies and the reported terrestrial plant incidents, the herbicide trifluralin is phytotoxic to many plant species (seedling emergence endpoints are more sensitive than vegetative vigor endpoints). Additionally, monocots are more sensitive to trifluralin than are dicots, based on available data. However, for adjacent upland and wetland plants, terrestrial plant RQs for both monocots and dicots exceed the Agency's risk to non-listed species LOC for 23 out of 25 trifluralin uses for at least one of the application methods. For the drift only RQs, one of the RQs exceed the Agency's LOC

5.2.2.4 Modification to Designated Critical Habitat for 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 potential for direct effects to the DS from trifluralin use could not be precluded based on incident data and RQ exceedances. Furthermore, it was concluded that trifluralin is likely to affect the DS by potentially affecting its habitat (terrestrial plants). Therefore, trifluralin may also affect critical habitat of the DS that is located in close proximity to triflualin use sites.

5.2.3 San Francisco Garter Snake

5.2.3.1 Direct Effects

Direct acute and chronic exposures of the SFGS were evaluated using the same approaches employed for estimating direct exposures to the terrestrial-phase CRLF (**Section 5.2.1.2**). In addition, toxicity estimates for both listed species, the terrestrial-phase CRLF and the SFGS, are based on the same surrogate avian toxicity data. Therefore, RQ values representing the potential for direct exposures and effects of trifluralin to the terrestrial-phase CRLF, are also used to represent the potential for direct exposures and effects of trifluralin to the SFGS.

Direct effects to SFGS are expected from exposure to trifluralin based on the summarized lines of evidence below:

- Acute risks from spray applications are unlikely since the EECs are lower than the LD_{50} and LC_{50} and no mortality or sublethal effects were noted in the acute bird studies.
- Acute risks from granular applications cannot be precluded at the highest application rate (4 lbs/acre with no soil incorporation) for 20 g individuals.
- One scenario (nursery use, exceeding RQ = 1.51) resulted in an RQ that exceeded the Chronic LOC.
- Chronic risks were refined with T-HERPS (**Table 5.11)**, resulting in LOC exceedances for the nursery scenario with three applications at 4 lbs/acre. RQs exceeded the chronic LOC (1.0) with values of 1.77 and 1.51 for herptiles that consume small herbivore mammals and small insects, respectively.
- Based on the fugacity model, chronic exposure of herptiles (specifically the SFGS) to trifluralin through consumption of contaminated soil invertebrates from fields treated with trifluralin is unlikely.

5.2.3.2 Indirect Effects (*via Reductions in Prey Base*)

Newborn and juvenile SFGS prey almost exclusively on Pacific tree frogs in temporary pools during the spring and early summer to the point that the SFGS may be so dependent on their anuran prey that they are not able to switch to other available prey sources if necessary to survive. SFGS under 500 mm snout-to-vent length (SVL) require Pacific tree frogs in various stages of metamorphosis, whereas individuals over 500 mm SVL can consume Pacific tree frog, CRLF, and bullfrog tadpoles and adults.

The main diet of adult SFGS consists of CRLFs. Adult SFGSs may also feed on smaller juvenile non-native bullfrogs (*Rana catesbeiana*). Immature California newts (*Taricha torosa*), California toads (*Bufo boreas halophilus*), recently metamorphosed western toads (*Bufo boreas*), threespine stickleback (*Gasterosteus aculeatus*), and non-native mosquito fish (*Gambusia affinis*) are also known to be consumed by SFGS. Small mammals, reptiles, amphibians, possibly invertebrates, and some fish species may also be consumed by the SFGS.

5.2.3.2.1 Freshwater Fish and Aquatic-phase Amphibians

RQ values representing exposures of trifluralin to freshwater fish and aquatic-phase amphibians that may serve as prey for the aquatic-phase CRLF are also used to represent exposures of trifluralin to freshwater fish and aquatic-phase amphibians that may serve as prey for the SFGS.

Based on the multiple lines of evidence presented in **Section 5.2.1.1**, there is a potential for indirect impact to the SFGS due to reduction of fish and aquatic-phase frogs as a prey base:

- There were Listed Species LOC exceedances in 18 of the 25 modeled scenarios.

- Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates.
- Fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS.
- Trifluralin has the potential to accumulate in tissues of aquatic organisms. The resulting RQ values for the three size classes of SFGS (**Table 5.3**) did not exceed the LOC for chronic dietary-based exposures through consumption of aquatic prey that have accumulated trifluralin. Acute RQs were not calculated as the avian acute toxicity studies resulted in no mortalities at levels higher than those expected in the environment.
- The spray drift buffer distance needed to get below the acute aquatic fish Listed Species LOC ranged from 45.9 to 2359 feet depending on application rate and method. These required buffer distances only considers the exposure due to spray drift; based on EFED's modeling, spray drift exposure alone does not cause the LOC to be exceeded.

5.2.3.2.2 Freshwater Invertebrates

RQ values representing exposures of trifluralin to freshwater invertebrates that may serve as prey for the aquatic-phase CRLF are also used to represent exposures of trifluralin to freshwater invertebrates that may serve as prey for the SFGS. No acute or chronic LOC exceedances occurred for freshwater invertebrates with RQs <0.05 for all 25 application scenarios for at least one of the application methods.

Based on the multiple lines of evidence as described in **Section 5.2.1.3.2** for the CRLF, indirect impacts to SFGS through reductions in the prey base (specifically aquatic invertebrates) are not expected from acute or chronic exposure to trifluralin.

5.2.3.2.3 Terrestrial Vertebrates

RQ values representing exposures of trifluralin to small and large terrestrial vertebrates that may serve as prey for the terrestrial-phase CRLF are also used to represent exposures of trifluralin that may serve as prey for the SFGS. Similarly, the Agency determined a reasonable potential exists for direct effects on birds (surrogate for terrestrial phase amphibians) based on exposure to food items receiving direct deposition from trifluralin application (*i.e.,* T-REX, T-HERPS modeling).

Indirect impacts to SFGS through reductions in the prey base (specifically terrestrial-phase herptiles) are expected from chronic exposure to trifluralin based on the summarized lines of evidence below:

- Acute risks from spray applications are unlikely since the EECs are lower than the LD_{50} and LC_{50} and no mortality or sublethal effects were noted in the acute bird studies.
- Acute risks from granular applications cannot be precluded at the highest application rate (4 lbs/acre with no soil incorporation) for 20 g individuals.

- One scenario (nursery use, exceeding RQ = 1.51) resulted in an RQ that exceeded the Chronic LOC.
- Chronic risks were refined with T-HERPS, resulting in LOC exceedances for the nursery scenario with three applications of 4 lbs/acre. RQs exceeded the chronic LOC (1.0) with values of 1.77 and 1.51 for herptiles that consume small herbivore mammals and small insects, respectively.
- Based on the fugacity model, chronic exposure of herptiles to trifluralin through consumption of contaminated soil invertebrates from fields treated with trifluralin is unlikely

Indirect impacts to SGFS through reductions in the prey base (specifically mammals) are expected from chronic exposure to trifluralin based on the summarized lines of evidence below (see **Section 5.2.1.3.4** for details):

- RQs for the non-definitive acute oral toxicity endpoints for mammalian species were not calculated because the acute effects data shows no mortality and no sublethal effects at any of the test levels (*i.e*., LD_{50} > 5000 mg/kg-bwt). With no soil incorporation, the dose-based EECs range from 1.02 to 1279.2 mg/kg-bwt, much lower that the highest dose tested (5000 mg/kg-bwt); therefore, acute dose-based risks from spray applications are unlikely. Since exposure from soil incorporated applications would be less than from non-incorporated applications, risks from incorporated applications are also unlikely.
- Risks to mammals from granular applications of trifluralin are unlikely as the calculated EECs do not exceed the toxicity values.
- Chronic RQ values representing trifluralin exposures to small mammals indicate risks resulting from all application scenarios (**Table 5.3**). In addition, the chronic dietary-based RQ values exceeded LOCs for all liquid application scenarios. The NOAEL yielded dose-based LOC exceedances for all liquid application scenarios.

5.2.3.2.4 Terrestrial Invertebrates

For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortaliy at this dose, MRID 05001991). The toxicity value for terrestrial invertebrates is calculated by multiplying the LD_{50} > 24.17 µg/bee by 1 bee/0.128g, which is based on the weight of an adult honey bee, resulting in a toxicity estimate of 188 µg/g-bee (equivalent to 188 mg/kg-bee). Since a definitive LD_{50} value was not available, RQs were not calculated. However, expected exposure levels were compared to the highest dose tested to evaluate the likelihood of risk. Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Therefore, EECs must be less than 9.4 mg/kg-insect in for risks to be considered unlikely.

EECs for small insects ranged from 270 mg/kg-insect at 2 lb/acre application to 755 mg/kg-insect at 4 lbs/acre with three (60-day interval) applications, and EECs for large insects ranged from 30mg/kg-insect at 2 lb/acre application to 84 mg/kg-insect at 4 lbs/acre with three (60-day interval) applications (**Table 3.11**). Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios.

5.2.3.3 Indirect Effects (*via Habitat Effects*)

SFGS inhabit areas near densely vegetated ponds and in open hillsides where it can sun, feed, and find cover in rodent burrows. It forages extensively in aquatic habitats. Fresh-water habitats including natural and manmade (e.g. stock) ponds, slow moving streams, vernal pools and other ephemeral or permanent water bodies which typically support inundation during winter rains and hold water for a minimum of 12 weeks in a year of average rainfall and upland habitats within 200 ft of the mean high water mark of such aquatic habitat.

5.2.3.3.1 Aquatic Plants (Vascular and Non-vascular)

Aquatic plants serve several important functions in aquatic ecosystems such as primary production (non-vascular, vascular) and refugia structure (vascular plants). Rooted plants help reduce sediment loading and provide stability to nearshore areas and lower streambanks. In addition, vascular aquatic plants are important as attachment sites for egg masses of aquatic species.

Based on the evidence presented in **Section 5.2.1.4.1**, indirect impacts to the SFGS due to reduction effects on its habitats through impact on vascular and non-vascular aquatic plants are not expected from acute exposure to trifluralin.

5.2.3.3.2 Terrestrial Plants

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

LOC exceedances:
As noted in **Section 5.2.1.4.2**, terrestrial plant RQs exceed LOCs, which indicates risk to upland and riparian vegetation. Because upland and riparian areas are comprised of a mixture of both woody plants and herbaceous vegetation, SFGS may be indirectly affected by adverse effects solely to herbaceous vegetation, which provides habitat and cover for the SFGS and its prey. Therefore, the indirect effect to the SFGS via reduction in terrestrial plants is neither insignificant nor discountable.

5.2.4 San Joaquin Kit Fox

SJKF occupies a variety of habitats, including grasslands, scrublands (*e.g.*, chenopod scrub and sub-shrub scrub), vernal pool areas, oak woodland, alkali meadows and playas, and an agricultural matrix of row crops, irrigated pastures, orchards, vineyards, and grazed annual grasslands. Kit foxes dig their own dens, modify and use those already constructed by other animals (ground squirrels, badgers, and coyotes), or use human-made structures (culverts, abandoned pipelines, or banks in sumps or roadbeds). They move to new dens within their home range often (likely to avoid predation by coyotes). The SJKF forages in California prairie and Sonoran grasslands in the vicinity of freshwater marshes and alkali sinks, where there is a dense ground cover of tall grasses and San Joaquin saltbush. Seasonal flooding in such habitats is normal. It feeds on small animals including blacktailed hares, desert cottontails, mice, kangaroo rats, squirrels, birds, lizards, insects and grass. The San Joaquin kit fox satisfies its moisture requirements from prey and does not depend on freshwater sources.

5.2.4.1 Direct Effects

Direct exposure of the SJKF and the resulting risks in terrestrial environments were evaluated based on dose- and dietary-based EECs estimated using two approaches (*i.e.*, T-REX for foliar spray applications and using fugacity-based modeling for insectivorous wildlife). All estimated EECs (*i.e.*, EECs for short grass, tall grass, broadleaf plants/small insects, fruits/pods/seeds/large insects) were considered relevant for evaluation of direct effects because the SJKF has been known to feed on insects and grasses. A mammalian body weight of 1000g was modeled based on the adult size of the kit fox. Based on these multiple lines of evidence presented below, there is potential for direct effects to the SJKF as result of labeled trifluralin use in California.

Acute risks from spray applications:
RQs for the non-definitive acute oral toxicity endpoints for mammalian species were not calculated because the acute effects data shows no mortality and no sublethal effects at any of the test levels (*i.e.*, LD_{50} > 5000 mg/kg-bwt). With no soil incorporation, the dose-based EECs range from 1.02 to 1279.2 mg/kg-bwt. These values are lower than the LD_{50}; therefore, acute dose-based risks from spray applications are unlikely. Since exposure from soil incorporated applications would be less than from non-incorporated applications, risks from incorporated applications are also unlikely. The probability of individual effect (mortality) for mammalian species was not calculated because the acute mammalian effects data shows no mortality and no sublethal effects were observed at any of the test concentrations (*i.e.*, LD_{50} > 5000 mg/kg-bwt).

Acute risks from granular applications:
Potential direct acute effects to mammals are also evaluated by considering granule consumption (LD_{50}/sq-ft). Toxicity values were calculated for three weight classes (15, 35, and 1000g individuals) based on the highest dose tested (5000 mg/kg-bwt). The calculated toxicity values are >167, >321, and >4170 mg/mammal. The 1000 g results are relevant for evaluating direct effects of the SJKF. The EECs at single non-incorporated application rates of 1.5, 2, and 4 lbs/acre were 15.6, 20.8, and 41.7 mg/sq-ft, respectively, and the EEC at a single soil-incorporated application rate of 2 lbs/acre is 0.21 mg/sq-ft. Risks to mammals from granular applications of trifluralin are unlikely as the calculated EECs do not exceed the toxicity values.

Chronic risks from unincorporated spray applications:
Chronic dose-based RQ values representing trifluralin exposures to large mammals (1000 g) indicate risks resulting from both the alfalfa and nursery application scenarios (for alfalfa, LOC exceedances for mammals consuming short grass, tall grass, and broadleaf plants/small insects; for nursery, LOC exceedences for mammals consuming short grass, tall grass, broadleaf

plants/small insects, and fruits/pods/seeds/large insects; **Table 5.5**). Chronic diet-based RQ values representing trifluralin exposures to all mammals indicate risks to mammals consuming short grass, tall grass, and broadleaf plants/small insects for both the alfalfa and nursery scenarios (**Table 5.5**).

Soil invertebrate consumption - potential chronic effects:

Chronic direct effects to SJKF that may consume soil invertebrates that have accumulated trifluralin residues in their tissues were modeled; however, this pathway is not included in T-REX. In order to explore the potential exposures of mammals to total residues of trifluralin that have accumulated in soil invertebrates inhabiting trifluralin treatment sites, a simple fugacity approach was employed to estimate trifluralin concentrations in earthworms and subsequent exposures to mammals consuming earthworms. Earthworms were chosen to represent all soil invertebrates that may be consumed by the terrestrial organisms under evaluation. This approach is explained in detail in **Appendix P**.

Using a single application rate of 4 lbs/acre, PRZM estimated trifluralin concentrations are 6.41 g/m^3 in soil, and the estimated concentration of trifluralin in earthworms is 147 mg/kg-earthworm. If it is assumed that a mammal consumes only contaminated soil invertebrates, its dietary-based exposures would be approximately 74% of the NOAEC in the mammalian reproduction study (NOAEC=200 mg/kg-diet, MRIDs 00151901, 00151902, 00151903). Therefore, chronic risks to the SJKF from consumption of trifluralin-contaminated soil invertebrates are unlikely.

5.2.4.2 Indirect Effects (*via Reductions in Prey Base*)

Potential forage items of the SJKF include small mammals, grasses, and insects.

5.2.4.2.1 Terrestrial Invertebrates

RQ values representing exposures of trifluralin to terrestrial invertebrates that may serve as prey for the terrestrial-phase CRLF and SFGS are also used to represent exposures of trifluralin to terrestrial invertebrates that may serve as prey for the SFKF.

For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortaliy at this dose, MRID 05001991). The toxicity value for terrestrial invertebrates is calculated by multiplying the LD_{50} > 24.17 µg/bee by 1 bee/0.128g, which is based on the weight of an adult honey bee, resulting in a toxicity estimate of 188 µg/g-bee (equivalent to 188 mg/kg-bee). Since a definitive LD_{50} values was not available, RQs were not calculated. However, expected exposure levels were compared to the highest dose tested to evaluate the likelihood of risk. Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Therefore, EECs must be less than 9.4 mg/kg-insect in for risks to be considered unlikely.

EECs for small insects ranged from 135 mg/kg-insect at 1 lb/acre application to 755 mg/kg-insect at 4 lbs/acre with three (60-day interval) applications, and EECs for large insects ranged from 15 mg/kg-insect at 1 lb/acre application to 84 mg/kg-insect at 4 lbs/acre with three (60-day interval) applications (**Table 3.11**). Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios.

5.2.4.2.2 Mammals

Indirect impacts to SJKF through reductions in the prey base (specifically mammals) are expected from chronic exposure to trifluralin based on the summarized lines of evidence below:

- RQs for the non-definitive acute oral toxicity endpoints for mammalian species were not calculated because the acute effects data shows no mortality and no sublethal effects at any of the test levels (*i.e*., LD_{50} > 5000 mg/kg-bwt). With no soil incorporation, the dose-based EECs range from 1.02 to 1279.2 mg/kg-bwt, much lower that the highest dose tested (5000 mg/kg-bwt); therefore, acute dose-based risks from spray applications are unlikely. Since exposure from soil incorporated applications would be less than from non-incorporated applications, risks from incorporated applications are also unlikely.
- Acute risks to mammals from granular applications of trifluralin are unlikely as the calculated EECs do not exceed the toxicity values.
- Chronic RQ values representing trifluralin exposures to mammals indicate risks resulting from all spray application scenarios using either dose-based or dietary-based exposure estimates (**Table 5.5**).

5.2.3.2.3 Terrestrial Plants

As noted in **Section 5.2.1.4.2**, terrestrial plant RQs exceed LOCs, which indicates risk to upland and riparian vegetation. Because upland and riparian areas are comprised of a mixture of both woody plants and herbaceous vegetation, SJKF may be indirectly affected by adverse effects solely to herbaceous vegetation, which provides a food source for the SJKF. Therefore, the indirect effect to the SJKF via reduction in terrestrial plants is neither insignificant nor discountable.

5.2.4.3 Indirect Effects (*via Habitat Effects*)

5.2.3.4.1 Terrestrial Plants

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

As noted in **Section 5.2.1.4.2**, terrestrial plant RQs exceed LOCs, which indicates risk to upland and riparian vegetation. Because upland and riparian areas are comprised of a mixture of both woody plants and herbaceous vegetation, SJKF may be indirectly affected by adverse effects solely to herbaceous vegetation, which provides a food source for the SJKF. Because available lines of evidence provide compelling reason to believe that trifluralin will affect any type of plants to the extent that it would affect the habitat integrity of the SJKF, labeled trifluralin use in California appears to indirectly affect the SJKF via impacts to habitat and/or primary production.

6. Uncertainties

6.1 Exposure Assessment Uncertainties

6.1.1 Maximum Usage 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. The frequency at which actual uses approach this maximum use scenario may be dependent on pest resistance, timing of applications, cultural practices, and market forces.

6.1.2 Aquatic Exposure Modeling of Trifluralin

There are no aerobic aquatic degradation data for trifluralin. A conservative default aerobic aquatic degradation half-life of 438 days was calculated as twice the aerobic soil metabolism half-life of 219 days for PRZM/EXAMS modeling. Also, there are no anaerobic aquatic degradation data for trifluralin. A default anaerobic aquatic degradation half-life of 59 days was calculated as the 90% upper confidence bound on the mean metabolism half-lives 59, 25,and 35 days.

Numerous trifluralin product labels in the LUIS Report do not specify the maximum application rate, maximum number of applications per year and/or minimum interval between applications. When not specified, the minimum interval is assumed to be 60 days because this is typically the interval when specified.

In cases of right-of-ways and residential uses, the general label language describing the extent of the application area led EFED to use conservative assumptions regarding the post-processing techniques to obtain EECs. In this modeling approach, it is assumed that right-of-ways and residential application sites are composed of equal parts pervious and impervious surfaces (*i.e.*, the EECs of both surfaces are multiplied by 50%). However, in reality, it is likely that right-of-ways and residential uses contain different ratios of the two surfaces. In general, incorporation of impervious surfaces into the exposure assessment results in increasing runoff volume in the watershed, which tends to reduce overall pesticide exposure assuming 1% overspray to the impervious surface. Further details on how this value was derived and characterization of alternative assumptions are provided in the Barton Springs salamander endangered species risk assessment for atrazine (USEPA 2006).

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

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

6.1.3 Multiple Growing Seasons per Year

Most trifluralin product labels specify application rates on a per crop cycle basis (not on a per year basis). Information from BEAD indicates that many crops can be grown more than one time/year in California (USEPA 2007). Since standard PRZM scenarios only consist of one crop per year, applications to only one crop per year were modeled. The cropping seasons range between two and four cycles per year. If trifluralin is applied for multiple cropping cycles within a year, EECs presented in this assessment may under predict exposures. For all other labeled uses, it was assumed that a maximum seasonal application specified on the label was equivalent to a maximum annual application.

6.1.4 Usage Uncertainties

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

6.1.5 Terrestrial Exposure Modeling of Trifluralin

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 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

Although there may be multiple trifluralin applications at a single site, it is unlikely that the same organism would be exposed to the maximum amount of spray drift from every application made. In order for an organism to receive the maximum concentration of trifluralin from multiple applications, each application of trifluralin would have to occur under identical atmospheric conditions (*e.g.*, same wind speed and – for plants – same wind direction) and (if it is an animal) the animal being exposed would have to be present directly downwind at the same distance after each application.

AgDRIFT 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. In many cases, the drift estimates from AgDRIFT may overestimate exposure even from single applications, especially as the distance increases from the site of application, since the model does not account for potential obstructions (*e.g*., large hills, berms, buildings, trees).

Conservative assumptions were made regarding the droplet size distributions being modeled ("ASAE Very Fine to Medium"), the application method (*e*.g., aerial), release heights and wind speeds. Alterations in any of these inputs would change the area of potential effect. Current trifluralin labels have varying directions regarding factors affecting droplet size distributions.

6.1.7 KABAM Modeling

In KABAM it is assumed that birds are surrogates for terrestrial-phase the CRLF and SFGS. As such, listed species of amphibians and reptiles are entered into the KABAM spreadsheet as birds.

In order to define the risks of CRLF consuming specific diet items such as benthic invertebrates and fish, KABAM can be used to derive Risk Quotient (RQ) values. Body weight assumptions of post-metamorphic small (1.4 g), medium (37 g), and large (238 g) CRLF are consistent with those incorporated into T-HERPS and are derived from data from Fellars (2007). Diet assumptions assigned to each of these size classes are provided in **Table 5.2**.

The SFGS is an endangered species that feeds on aquatic organisms. KABAM can be applied to this species by using weights of juvenile (2 g), adult male (113 g), and adult female (227 g) for input values. The juvenile weight was obtained from Cover and Boyer (1988). Diet assumptions assigned to each of these size classes are provided in **Table 5.2**. Different weights to be assessed, as well as the different diets, reflect the range of both variables.

The weight of the male SFGS is assumed to be 0.113 kg and consumes either 100% benthic invertebrates or 100% medium fish. The weight of the medium size class of fish in KABAM is 0.1 kg, which is very close to the snake bodyweight. Based on the available data for the SFGS weights, the 0.113 kg is an average value. The consumption of the 0.1 kg fish may be more likely for larger male snakes in the range of weights. This diet class therefore represents the upper bound for risk to adult male SFGS through dietary bioaccumulation. Diet assumptions assigned to each of these size classes are provided in **Table 5.2**.

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 Use of Surrogate Species Effects Data

Guideline toxicity tests and open literature data on trifluralin considered suitable for quantitative use are available for aquatic-phase frogs; therefore, freshwater fish are used as surrogate species for aquatic-phase amphibians. Although data evaluating the acute toxicity to aquatic-phase amphibians were reviewed, EFED determined that the use of freshwater fish data is preferable to the use of aquatic-phase amphibian data because it is unknown where the CRLF would fall on a species sensitivity distribution. The available open literature information on trifluralin toxicity to aquatic-phase amphibians shows that acute ecotoxicity endpoints for aquatic-phase amphibians are generally about 6 times less sensitive than freshwater fish (bluegill sunfish LC_{50} = 18.5 µg/L, Fowler's tadpoles LC_{50} = 115 µg/L). Because amphibian data is not required from the registrant, it is EFED's standard approach to use freshwater fish as a surrogate for aquatic-phase amphibians. In addition, because acute amphibian data were less sensitive than acute freshwater fish data, the use of freshwater fish as a surrogate provides a more conservative estimation of risk to the aquatic-phase CRLF.

To assess potential direct and indirect effects of trifluralin to the DS, toxicity data for both the freshwater fish and estuarine/marine fish were evaluated since the DS lives in brackish waters. The most sensitive freshwater or estuarine/marine fish toxicity data was utilized in the risk estimation. Since the available data indicated freshwater fish were more sensitive than estuarine/marine fish, the freshwater fish toxicity data was used to assess potential direct effects of trifluralin to the DS. Similarly, freshwater invertebrate data was used to assess potential indirect effects (prey) to the DS, as the available data indicated freshwater invertebrates were more sensitive than estuarine/marine invertebrates. The extrapolation of the risk conclusions from the most sensitive tested species to the aquatic-phase CRLF and DS may overestimate the potential risks to those species.

Acceptable guideline toxicity tests and open literature studies for reptiles are not currently available for quantitative use to assess potential risks of trifluralin use in California to the SFGS. Therefore, toxicity data for surrogate species (*i.e.*, birds for reptiles) are used in some instances to assess risks. Efforts are made to select the organisms, which are most likely to be affected by the type of compound and usage pattern; however, there is an inherent uncertainty in extrapolating across phyla. In addition, the Agency's LOCs are intentionally set very low, and conservative estimates are made in the screening level risk assessment to account for these uncertainties.

6.2.3 Toxicity Endpoints

There are no acute estuarine/marine fish data for evaluation of toxicity to trifluralin. For acute estuarine/marine fish, data from ethafluralin (an herbicide in the same chemical class as trifluralin) are used.

There are no estuarine/marine invertebrate data for evaluation of chronic toxicity to trifluralin. For chronic estuarine/marine invertebrate toxicity, the ACR was calculated using trifluralin data for the freshwater invertebrate (acute and chronic) and the estuarine/marine invertebrate (acute).

Acute toxicities for birds and mammals are based on studies in which no individuals died and no sublethal effects were identified. The toxicity value for the terrestrial invertebrate is based on the non-definitive LD_{50} of > 24.17 µg/bee. For these taxa, RQs are not calculated; risks are discussed qualitatively.

6.2.4 Sublethal Effects

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

6.2.5 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 trifluralin to the CRLF, DS, SFGS, and SJKF and the designated critical habitat of the CRLF and the DS.

In order to conclude this risk assessment, it is necessary to address the risk hypotheses defined in **Section 2.9.1**. Based on the conclusions of this assessment, none of the hypotheses can be rejected, meaning that the stated hypotheses represent concerns in terms of direct and indirect effects of trifluralin to the CRLF, DS, SFGS, and SJKF and the designated critical habitat of the CRLF and the DS.

Based on the best available information, the Agency makes a Likely to Adversely Affect (LAA) determination for the CRLF from the use of trifluralin. Additionally, the Agency makes a Likely to Adversely Affect determination for the DS, SFGS, and SJKF from the use of trifluralin. 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, FD, DFGS, and SJKF and potential modification of designated critical habitat for the CRLF and DS, 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, DS, SFGS, and SJKF and the critical habitat for the CRLF and the DS, 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 Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination** [1]	**Basis for Determination**
CRLF	LAA	**Potential for Direct Effects** ***Aquatic-phase (Eggs, Larvae, and Adults)***: *Fish, Adult survival:* Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. The chance of individual effects (*i.e*., mortality) for freshwater fish (surrogate for aquatic-phase CRLFs) is as high as ~1 in 3240. Buffer distance required to remove all LOC exceedances under the evaluated labels ranged from 45.9 to 2359 feet. Available aquatic-phase amphibian data suggests trifluralin is less acutely toxic to aquatic-phase amphibians than to fish; however, there are Listed Species LOC exceedances for the most sensitive aquatic-phase amphibian data. *Fish, Growth and reproduction*: There were no chronic LOC exceedances. However, fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS. *Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the maximum observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09).
		Terrestrial-phase (Juveniles and Adults): *Adult survival:* There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater than expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e*., short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories.
		Potential for Indirect Effects ***Aquatic prey items, aquatic habitat, cover and/or primary productivity*** *Fish, Adult survival:* Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. The chance of individual effects (*i.e*., mortality) for freshwater fish (surrogate for aquatic-phase CRLFs) is as high as ~1 in 3240. Buffer distance required to remove all LOC

Table 7.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination** [1]	**Basis for Determination**
		exceedances under the evaluated labels ranged from 45.9 to 2359 feet. Available aquatic-phase amphibian data suggests trifluralin is less acutely toxic to aquatic-phase amphibians than to fish; however, there are Listed Species LOC exceedances for the most sensitive aquatic-phase amphibian data. *Fish, Growth and reproduction*: There were no chronic LOC exceedances. However, fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS. *Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the maximum observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09). *Freshwater invertebrates:* There were no exceedances of the Listed Species Acute or Chronic LOCs. *Vascular and non-vascular plants:* There were no exceedances of the Acute LOCs. *Terrestrial plants*: LOCs were exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs were exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
		Terrestrial prey items, riparian habitat *Birds, Adult survival:* There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater then expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Birds, Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e.*, short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary

Table 7.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination** [1]	**Basis for Determination**
		categories. *Mammals: adult survival:* There were no mortalities or sublethal effects in the acute dose. Acute risks to mammals from spray or granular applications of trifluralin are unlikely as the calculated exposures do not exceed the toxicity values. *Mammals, growth and reproduction*: Chronic dose-based RQs for 15 g mammals range from 20.82 to 58.20 for short grass; 9.54 to 26.68 for tall grass; 11.71 to 32.74 broadleaf plants/small insects; 1.30 to 3.64 for fruits/pods/seeds/large insects. Chronic RQs did not exceed LOCs for 15g granivores. Chronic dietary-based RQs exceed the Agency's chronic LOC (1.0) in both crop scenarios (liquid application) for mammals consuming short grass, tall grass, and broadleaf plants/small insects; they were not exceeded for fruits/pods/seeds/large insects. *Terrestrial invertebrates*: For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortality at this dose). Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios. *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
DS	LAA	**Potential for Direct Effects**
		Fish, Adult survival: Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. The chance of individual effects (*i.e.*, mortality) for freshwater fish (surrogate for aquatic-phase CRLFs) is as high as ~1 in 3240. Buffer distance required to remove all LOC exceedances under the evaluated labels ranged from 45.9 to 2359 feet. *Fish, Growth and reproduction*: There were no chronic LOC exceedances. However, fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS.

Table 7.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination [1]**	**Basis for Determination**
		Aquatic monitoring data: Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09).
		Potential for Indirect Effects
		Freshwater invertebrates: There were no exceedances of the Listed Species Acute or Chronic LOCs. *Vascular and non-vascular plants:* There were no exceedances of the Acute LOCs. *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
SFGS	LAA	**Potential for Direct Effects**
		Adult survival: There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater then expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e.*, short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories.
		Potential for Indirect Effects
		Fish, Adult survival: Acute RQs range from 0.05 to 0.35; exceeding the Agency's acute listed species LOC (0.05) in 18 out of 25 crop scenarios with at least one of the application methods. The chance of individual effects (*i.e.*, mortality) for freshwater fish (surrogate for aquatic-phase CRLFs) is as high as ~1 in 3240. Buffer distance required to remove all LOC exceedances under the evaluated labels ranged from 45.9 to 2359 feet.

Table 7.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination** [1]	**Basis for Determination**
		Fish, Growth and reproduction: There were no chronic LOC exceedances. However, fish exposed to trifluralin may also develop vertebral dysplasia, which could significantly impact the fishes' ability to swim, and therefore, impact its fitness. The concentrations in these toxicity studies are within an order of magnitude of the maximum concentrations observed in the monitoring data and many of the chronic EECs estimated using PRZM/EXAMS. *Freshwater invertebrates:* There were no exceedances of the Listed Species Acute or Chronic LOCs. *Vascular and non-vascular plants:* There were no exceedances of the Acute LOCs. *Birds, Adult survival:* There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater then expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Birds, Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e*., short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories. *Mammals: adult survival:* There were no mortalities or sublethal effects in the acute dose. Acute risks to mammals from spray or granular applications of trifluralin are unlikely as the calculated exposures do not exceed the toxicity values. *Mammals, growth and reproduction*: Chronic dose-based RQs for 15 g mammals range from 20.82 to 58.20 for short grass; 9.54 to 26.68 for tall grass; 11.71 to 32.74 broadleaf plants/small insects; 1.30 to 3.64 for fruits/pods/seeds/large insects. Chronic RQs did not exceed LOCs for 15g granivores. Chronic dietary-based RQs exceed the Agency's chronic LOC (1.0) in both crop scenarios (liquid application) for mammals consuming short grass, tall grass, and broadleaf plants/small insects; they were not exceeded for fruits/pods/seeds/large insects. *Terrestrial invertebrates*: For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortality at this dose). Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios. *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from

Table 7.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination [1]**	**Basis for Determination**
		1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
SJKF	LAA	**Potential for Direct Effects**
		Mammals: adult survival: There were no mortalities or sublethal effects in the acute dose. Acute risks to mammals from spray or granular applications of trifluralin are unlikely as the calculated exposures do not exceed the toxicity values. *Mammals, growth and reproduction*: Chronic dose-based RQ values representing trifluralin exposures (spray applications) to large mammals (1000 g) indicate risks resulting from both the alfalfa and nursery application scenarios (for alfalfa, LOC exceedances for mammals consuming short grass, tall grass, and broadleaf plants/small insects; for nursery, LOC exceedences for mammals consuming short grass, tall grass, broadleaf plants/small insects, and fruits/pods/seeds/large insects. Chronic diet-based RQ values representing trifluralin exposures to all mammals indicate risks to mammals consuming short grass, tall grass, and broadleaf plants/small insects for both the alfalfa and nursery scenarios. Chronic risk to mammals from trifluralin through consumption of contaminated earthworms is unlikely.
		Potential for Indirect Effects
		Birds, Adult survival: There were no mortalities or sublethal effects in the avian acute dose and diet studies. The highest tested trifluralin level was greater then expected concentrations in the environment for spray applications. For the granular non-incorporated application rates of 2 and 4 lbs/acre, the expected exposure level meets or exceeds the highest level tested for 20 g individuals; therefore, acute risks to small birds and surrogate species cannot be precluded. *Birds, Growth and reproduction*: The chronic (dietary-based) RQs for birds exceeded the chronic LOC (RQ>1) for the nursery use category. Chronic RQs ranged from 1.23 to 2.68 for various dietary categories (*i.e.*, short grass, tall grass and broadleaf plants and insects). After refinement with T-HERPS, chronic RQs still exceeded LOCs for some dietary categories. *Mammals: adult survival:* There were no mortalities or sublethal effects in the acute dose. Acute risks to mammals from spray or granular applications of trifluralin are unlikely as the calculated exposures do not exceed the toxicity values. *Mammals, growth and reproduction*: Chronic dose-based RQs for all weight classes of mammals exceeded the Chronic LOC (1.0) for all weights and feeding guilds except for large mammals consuming fruits/pods/seeds/large insects (alfalfa application) and all granivores. Exceeding RQs ranged from 1.11 to 58.20. Chronic dietary-based RQs exceed the Agency's

Table 7.1 Effects Determination Summary for Effects of Trifluralin on the CRLF, DS, SFGS, and SJKF		
Species	**Effects Determination** [1]	**Basis for Determination**
		Chronic LOC (1.0) in for alfalfa and nursery scenarios (liquid application, non-incorporated) for short grass, tall grass, and broadleaf plants/small insects. *Terrestrial invertebrates*: For honeybees, the most sensitive acute contact LD_{50} > 24.17 µg/bee (13% mortality at this dose). Since mortality was observed at the highest test dose, expected exposures must be 20x less than the highest dose to preclude risks to terrestrial invertebrates. Risks to terrestrial invertebrates cannot be precluded since the calculated EECs exceed the calculated cut-point of 9.4 mg/kg-insect for all modeled scenarios. *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses.
[1] No effect (NE); May affect, but not likely to adversely affect (NLAA); May affect, likely to adversely affect (LAA)		

Table 7.2 Effects Determination Summary for the Critical Habitat Impact Analysis (CRLF and DS)[1]		
Designated Critical Habitat for:	**Effects Determination [2]**	**Basis for Determination**
CRLF	HM	***Aquatic-phase PCEs:*** *Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09). *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses. There is a potential for direct effects to aquatic-phase CRLF and indirect effects via reduction of aquatic-phase prey items (fish and aquatic-phase amphibians) as described in **Section 5**. ***Terrestrial-phase PCEs:*** *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85 % were incidents were classified as registered uses. There is a potential for direct effects to terrestrial–phase CRLF and indirect effects via reduction of terrestrial-phased prey items (mammals, terrestrial invertebrates, and frogs) as described in **Section 5**.
DS	HM	*Aquatic monitoring data:* Of the 3,915 non-targeted water samples tested for trifluralin, only 15% had detectible concentrations of trifluralin; however, those measured concentrations were comparable to the PRZM/EXAMS estimates. If the observed measured concentration (1.74 µg/L) was used as an exposure value to calculate an acute freshwater fish RQ, there would be an exceedance of the Listed Species LOC (RQ = 0.09). *Terrestrial plants*: LOCs exceeded for 23 out of 25 modeled uses for at least one of the application methods for risks to monocot plants. RQs that exceed the acute

		LOC (1.0) range from 1.11 to 4.89 for monocots. LOCs exceeded for 14 out of 25 modeled uses for at least one of the application methods for dicot plants. RQs that exceed the acute LOC (1.0) range from 1.05 to 2.32 for dicots. *Terrestrial incident reports:* All 78 reported terrestrial incidents involved plant damage or death from direct application or drift. Seventy-one percent are classified as 'probable' in the context of trifluralin use; 85% were incidents were classified as registered uses. There is a potential for direct effects to the DS as described in **Section 5**.
[1] Critical habitat has not been designated for the SFGS or the SJKF. [2] Habitat Modification (HM) or No effect (NE)		

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

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

- Enhanced information on the density and distribution of CRLF, DS, SFGS, and SJKF 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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