

Risks of Simazine Use to Federally Listed Endangered Barton Springs Salamander (Eurycea sosorum)

September 20, 2007

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(Eurycea sosorum)

Pesticide Effects Determination

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

September 20, 2007

Table of Contents

1.	Executi	ve Summary	6
2.	Problen	n Formulation	8
2	2.1 Pu	ırpose	8
2	2.2 Sc	ope	9
2	2.3 Pro	evious Assessments	11
	2.3.1	Simazine	11
	2.3.2	Barton Springs Salamander	11
2	2.4 Stı	ressor Source and Distribution	
	2.4.1	Environmental Fate and Transport Assessment	11
	2.4.2	Mechanism of Action	
	2.4.3	Use Characterization	15
2	2.5 As	ssessed Species	19
2		ction Area	
2	2.7 As	ssessment Endpoints and Measures of Ecological Effect	24
2		onceptual Model	
	2.8.1	Risk Hypotheses	26
	2.8.2	Diagram	
2	2.9 Ar	nalysis Plan	28
3.	Exposu	re Assessment	30
3		bel Application Rates and Intervals	
3		quatic Exposure Assessment	
	3.2.1	Background	32
	3.2.2	Geology/Hydrogeology	
	3.2.3	Conceptual Model of Exposure	
	3.2.4	Existing Monitoring Data	
	3.2.5	Modeling Approach	42
	3.2.6	Model Inputs	48
	3.2.7	PRZM Scenarios	50
	3.2.7	.1 Orchard	50
	3.2.7	.2 Turf	50
	3.2.7	3 Non-cropland	51
	3.2.7	.4 Residential	56
	3.2.8	Results	57
4.	Effects	Assessment	60
4	4.1 To	exicity of Simazine to Aquatic Organisms	62
	4.1.1	Toxicity to Freshwater Fish	
	4.1.1	.1 Freshwater Fish: Acute Exposure (Mortality) Studies	64
	4.1.1		
	4.1.1		
	Infor	mation	65

	4.1.2 Toxicity to Freshwater Invertebrates	. 66
	4.1.2.1 Freshwater Invertebrates: Acute Exposure Studies	. 67
	4.1.2.2 Freshwater Invertebrates: Chronic Exposure Studies	. 67
	4.1.2.3 Freshwater Invertebrates: Open Literature Data	. 68
	4.1.3 Toxicity to Aquatic Plants	
	4.1.3.1 Aquatic Plants: Laboratory Data	. 68
	4.1.4 Freshwater Field Studies	
	4.2 Use of Probit Slope Response Relationship to Provide Information on the	
	Endangered Species Levels of Concern	. 70
	4.3 Incident Database Review	
5.	Risk Characterization	. 71
	5.1 Risk Estimation	. 72
	5.1.1 Direct Effects	. 72
	5.1.2 Indirect Effects	. 73
	5.1.2.1 Evaluation of Potential Indirect Effects via Reduction in Food Items	
	(Freshwater Fish and Invertebrates)	. 74
	5.1.2.2 Evaluation of Potential Indirect Effects via Reduction in Habitat and	or/
	Primary Productivity (Freshwater Aquatic Plants)	. 75
	5.2 Risk Description	. 75
	5.2.1 Direct Effects to the Barton Springs Salamander	. 76
	5.2.2 Indirect Effects via Reduction in Food Items (Freshwater Invertebrates)	78
	5.2.3 Indirect Effects via Reduction in Habitat and/or Primary Productivity	
	(Freshwater Aquatic Plants)	. 79
5.		
	6.1 Exposure Assessment Uncertainties	. 80
	6.1.1 Modeling Assumptions	. 80
	6.1.2 PRZM Modeling Inputs and Predicted Aquatic Concentrations	
	6.2 Effects Assessment Uncertainties	
	6.2.1 Age class and sensitivity of effects thresholds	
	6.2.2 Use of surrogate species effects data	
	6.2.3 Extrapolation of long-term environmental effects from short-term	
	laboratory tests	. 82
	6.2.4 Exposure to Pesticide Mixtures	
	6.2.5 Sublethal Effects	. 83
7.	Summary of Direct and Indirect Effects to the Barton Springs Salamander	. 84
_		
8.	References	. 84
	I : a4 - 2 T-L1	
	List of Tables	
Го	able 1.1 Effects Determination Summary for the Barton Springs Salamander	7
	able 2.1 Summary of Simazine Environmental Fate Properties	
10	ore 2.1 Summary of Simazine Environmental Pate Properties	. 13

Table 2.2 Summary of Assessment Endpoints and Measures of Ecological Effect	25
Table 3.1 Label Application Information for the Barton Springs Salamander Endangered	d
Species Assessment ¹	31
Table 3.2 Extent of Potential Simazine Use Areas in the Action Area of the Barton	
Springs Segment of the Edwards Aquifer (BSSEA)	45
Table 3.3 Summary of PRZM/EZAMS Environmental Fate Data Used for Aquatic	
Exposure Inputs for Simazine Endangered Species Assessment for the Barton Springs	
Salamander	49
Table 3.4 Summary of ROW Landcover Types within Barton Springs Action Area	52
Table 3.5 Summary of ROW Types in Action Area	53
Table 3.6 Summary of PRZM Output EECs for Varying Percentages of Treated Non-	
cropland	
Table 3.7 Summary of PRZM Output EECs for all Modeled Scenarios	
Table 3.8 Summary of PRZM Output EECs for all Modeled Scenarios Using Post-RED	
Application Information	59
Table 4.1 Comparison of Acute Freshwater Toxicity Values for Simazine and	
Degradates	
Table 4.2 Freshwater Aquatic Toxicity Profile for Simazine	62
Table 4.3 Categories of Acute Toxicity for Aquatic Organisms	
Table 5.1 Summary of Direct Effect RQs for the Barton Springs Salamander	
Table 5.2 Summary of RQs Used to Estimate Indirect Effects to the Barton Springs	
Salamander via Direct Toxicity to Dietary Items	74
Table 5.3 Summary of RQs Used to Estimate Indirect Effects to the Barton Springs	
Salamander via Direct Effects on Aquatic Plants	75
Table 5.4 Preliminary Effects Determination Summary for the Barton Springs	
Salamander Based on Risk Estimation	
Table 7.1 Effects Determination Summary for the Barton Springs Salamander	84
List of Figures	
Figure 2.1 Simazine Use in Total Pounds per County	17
Figure 2.2 Location Map of Barton Springs (dots with curved line indicate location of	
springs)	
Figure 2.3 Aerial Photo of Barton Springs	
Figure 2.4 Barton Springs Salamander Action Area	24 24
Figure 2.5 Conceptual Model for Barton Springs Salamander	
Figure 3.1 Barton Springs Segment of the Edwards Aquifer with HydroZones	
Figure 3.2 Hydrogeologic Cross Section of the Barton Springs Segment of the Edwards	
Aquifer Showing Dominant Flow Pathways Within Each Hydrozone (Taken from	
	35
Figure 3.3 Conceptual Model of Surface and Subsurface Flow Within the Barton Spring	
Segment of the Edwards Aquifer Relative to the Barton Springs Salamander	-
Figure 3.4 Flow paths within Recharge Zone of the Barton Springs Segment of the	
Edwards Aquifer (Taken from Mahler, 2005a; originally published in Hauwert et al.,	
2004)	38
Figure 3.5 Location of USGS Surface Water Sites within the Barton Springs Segment . 4	

U	cation of USGS Groundwater Sites within the Barton Springs Segment 42
	thts-of-Way within the Barton Springs Salamander Action Area 51
Figure 3.8 Tre	ated Rights-of-Way within Travis County Relative to Barton Springs
Action Area	
Figure 3.9 Res	sidential Land Cover within the Barton Springs Action Area56
	Appendices
Appendix A:	Simazine Ecological Effects Characterization
Appendix B:	Multi-ai Product Analysis
Appendix C:	Life History of the Barton Springs Salamander
Appendix D:	Simazine Aquatic Incidents
Appendix E:	RQ Method and LOCs
Appendix F:	Bibliography of ECOTOX Open Literature Not Evaluated

1. Executive Summary

The purpose of this assessment is to make an "effects determination" for the Barton Springs salamander (*Eurycea sosorum*) by evaluating the potential direct and indirect effects of currently registered uses of the herbicide simazine within the Barton Springs area (action area) on the survival, growth, and reproduction of this federally listed endangered species. This assessment was completed in accordance with the U.S. Fish and Wildlife Service (USFWS) and National Marine Fisheries Service (NMFS) *Endangered Species Consultation Handbook* (USFWS/NMFS 1998) and procedures outlined in the Agency's Overview Document (U.S. EPA, 2004).

The range of the Barton Springs salamander is restricted to four spring outlets that comprise the Barton Springs complex, which is located near downtown Austin, Texas. Subsurface flow from the Barton Springs segment of the Edwards Aquifer and its contributing zone supply all of the water in the springs that make up the Barton Springs complex. Therefore, the simazine action area as it relates to the Barton Springs salamander is defined by those areas within the hydrogeologic watershed that discharge to Barton Springs.

Simazine is used as a selective herbicide to control most annual grasses and broadleaf weeds (before they emerge or after removal of weed growth). Simazine may be applied as a liquid via ground sprayer, banded application, or broadcast, or as a granular formulation. Simazine is registered for use on a variety of food and feed crops including fruits, nuts, citrus, and corn, as well as non-agricultural uses including turf, non-cropland, rights-of-way, tree plantations and nurseries, shelterbelts, and residential use. Although simazine is registered for many agricultural and non-agricultural uses, its use within the action area is limited to a single peach orchard and non-agricultural turf, rights-of-way (i.e., non-cropland), and residential uses, based on use estimates provided by the Agency's Biological and Economic Assessment Division (BEAD) and from discussions with U.S. Department of Agriculture extension agents in the Austin, TX area.

Environmental fate and transport models were used to estimate high-end exposure values that could occur at the edge of use sites and in water in the Barton Springs action area as a result of potential agricultural and non-agricultural simazine use in accordance with label directions. Modeled concentrations in Barton Springs provide estimates of exposure that are intended to represent possible simazine concentrations originating from all potential use sites within the action area. Transport of water containing simazine could occur in surface water in the contributing zone and in the recharge zone predominantly from subsurface flow through the fractured karst limestone of the Edwards Aquifer. Estimated 1-in-10-year peak exposure values for the Barton Springs were aggregated from all potential use sites and used in risk estimation. Estimated peak modeled exposure values are approximately one to two orders of magnitude higher than simazine concentrations reported in available monitoring data from Barton Springs. Detected concentrations of simazine reported from Barton Springs monitoring data are generally well below 1 μ g/L.

The highest potential simazine exposure is predicted to occur from residential uses. However, it should be noted that the aerial non-cropland use and the granular non-residential turf use, which are considered as part of the aggregate simazine exposure in this assessment, are scheduled to be cancelled in 2010.

The assessment endpoints for the Barton Springs salamander include direct toxic effects on the survival, reproduction, and growth of the salamander itself, as well as indirect effects, such as reduction of the prey base and/or modification of its habitat. Direct effects to the Barton Springs salamander are based on toxicity information for freshwater fish, which are generally used as a surrogate for aquatic-phase amphibians. Given that the salamander's prey items and habitat requirements are dependant on the availability of freshwater invertebrates and aquatic plants, respectively, toxicity information for these taxonomic groups is also discussed.

Comparison of available toxicity information for the degradates of simazine indicates lesser aquatic toxicity than the parent for freshwater fish, aquatic invertebrates, and aquatic plants. Because degradates are not of greater toxicological concern than simazine, concentrations of the simazine degradates are not assessed further, and the focus of this assessment is parent simazine.

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 if simazine use within the action area has any direct or indirect effect on the Barton Springs salamander. Based on estimated environmental concentrations (EECs) for the currently registered uses of simazine, RQ values are below the Agency's LOCs for direct and indirect effects to the Barton Springs salamander; therefore, the effects determination is "no effect". A summary of the risk conclusions and effects determination for the Barton Springs salamander is presented in Table 1.1.

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

2.1 Purpose

The purpose of this ecological risk assessment is to make an "effects determination" as directed in Section 7(a) (2) of the Endangered Species Act (ESA), for the Barton Springs salamander (Eurycea sosorum) by evaluating the potential direct and indirect effects resulting from use of the herbicide simazine (6-chloro-N, N'-diethyl-1,3,5-triazine-2,4-diamine) on the survival, growth, and/or reproduction of this Federally endangered species. The Barton Springs salamander was federally listed as an endangered species on May 30, 1997 (62 FR 23377-23392) by the U.S. Fish and Wildlife Service (USFWS or the Service). No critical habitat has been designated for this species. This ecological risk assessment is a component of the settlements for *Center for Biological Diversity and Save Our Springs Alliance v. Leavitt, No. 1:04CV00126-CKK (filed January 26, 2004)* and *Natural Resources Defense Council, Civ. No: 03-CV-02444 RDB (filed March 28, 2006)*.

In this assessment, direct and indirect effects to the survival, growth, and reproduction of the Barton Springs salamander are evaluated in accordance with the screening-level methodology described in the Agency's Overview Document (U.S. EPA 2004).

As part of the "effects determination", the Agency reaches one of the following three conclusions regarding the potential for simazine to affect the Barton Springs salamander in accordance with current labels:

"No effect";

"May affect, but not likely to adversely affect"; or

"May affect and likely to adversely affect".

If the results of the screening-level assessment show no indirect effects and levels of concern (LOCs) for the Barton Springs salamander are not exceeded for direct effects, a "no effect" determination is made, based on simazine's use within the action area. If, however, indirect effects are anticipated and/or exposure exceeds the LOCs for direct effects, the Agency concludes a preliminary "may affect" determination for the Barton Springs salamander.

If a determination is made that use of simazine within the action area "may affect" the Barton Springs salamander, 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 Barton Springs salamander. Based on the refined

information, the Agency will use the best available information to distinguish those actions that "may affect, but are not likely to adversely affect" from those actions that are "likely to adversely affect" the Barton Springs salamander. This information is presented as part of the Risk Characterization in Section 5.

2.2 Scope

Simazine is widely used as a selective herbicide to control most annual grasses and broadleaf weeds (before they emerge or after removal of weed growth). Simazine is registered for pre-plant use or use in established fields of a variety of food and feed crops including fruit and nut crops such as apples, oranges, and almonds, in addition to corn. Simazine can also be applied on Christmas trees and on turfgrass grown commercially for sod. Nonagricultural uses for simazine include nonselective weed control in industrial sites, highway medians and shoulders, railroad rights-of-way, lumberyards, petroleum tank farms, and in noncrop areas on farms, such as around buildings, equipment and fuel storage areas, along fences, road-sides, and lanes. Simazine is also registered for residential use on turfgrass including both commercial use on recreational lawns such as golf courses and commercial or homeowner use on home lawns. There is an additional registration for simazine as an algaecide in ornamental ponds and aquariums of 1,000 gallons or less. Simazine can be applied as a liquid via ground sprayer, banded application, or broadcast, or as granular formulation.

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

This ecological risk assessment is for currently registered uses of simazine in the action area associated with the Barton Springs salamander. Further discussion of the action area for the Barton Springs salamander is provided in Section 2.6.

Degradates of simazine include deisopropyl-atrazine (DIA), diamino-chlorotriazine (DACT), and hydroxysimazine (HS). Comparison of available toxicity information for the degradates of simazine indicates lesser toxicity than the parent for fish, aquatic invertebrates, and aquatic plants. Acute toxicity values for DIA are approximately 2.6-fold less sensitive than acute toxicity values for simazine in freshwater fish. In addition, no adverse effects were observed at concentrations up to 100 mg/L in fish and daphnids for DACT and in daphnids for DIA. Available aquatic plant degradate toxicity data for DIA and DACT report EC_{50} values at concentrations that are at least 69 times higher than the lowest reported aquatic plant EC_{50} value for parent simazine. Although toxicity information is not available for HS, this degradate is also likely to be less toxic than parent simazine, given that the more toxic chloro group is replaced by a less toxic hydroxyl group during its formation. Therefore, given the lesser toxicity of the degradates, as compared to the parent, and the relatively small proportion of the degradates expected to be in the environment and available for exposure relative to

simazine, the focus of this assessment is parent simazine. Additional details on available toxicity data for the degradates are provided in Section 4 and Appendix A.

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).

Simazine 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 B. The results of this analysis show that an assessment based on the toxicity of the single active ingredient of simazine is appropriate.

The results of available toxicity data for mixtures of simazine with other pesticides are presented in Section A.6 of Appendix A. Other triazine herbicides may combine with simazine to produce additive toxic effects on aquatic plants. The variety of chemical interactions presented in the available data set suggest that the toxic effect of simazine, in combination with other pesticides used in the environment, can be a function of many factors including but not necessarily limited to: (1) the exposed species, (2) the cocontaminants in the mixture, (3) the ratio of simazine and co-contaminant concentrations, (4) differences in the pattern and duration of exposure among contaminants, and (5) the differential effects of other physical/chemical characteristics of the receiving waters (e.g. organic matter present in sediment and suspended water). Quantitatively predicting the combined effects of all these variables on mixture toxicity to any given taxa with confidence is beyond the capabilities of the available data. However, a qualitative discussion of implications of the available pesticide mixture effects data involving simazine on the confidence of risk assessment conclusions for the Barton Springs salamander is addressed as part of the uncertainty analysis for this effects determination.

Although current registrations for simazine allow for use nationwide, this ecological risk assessment and effects determination addresses currently registered uses of simazine in portions of the action area that are reasonably assumed to be biologically relevant to the Barton Springs salamander.

2.3 Previous Assessments

2.3.1 Simazine

A Reregistration Eligibility Decision (RED) was completed for simazine on April 6, 2006 (U.S. EPA, 2006a)¹. The results of the Agency's ecological risk assessment for simazine, which was completed as part of the RED, suggest the potential for adverse acute effects to non-target terrestrial and aquatic plants, and direct chronic effects to birds and mammals. In addition, a number of the granular uses resulted in potential direct adverse effects to freshwater invertebrates and fish, although there was a high degree of uncertainty associated with the freshwater fish data set because exposure concentrations were not verified in the available acute toxicity tests. Simazine is not likely to be acutely toxic to estuarine/marine fish and invertebrates, and it is unlikely to cause acute mortality to birds and mammals, although acute sublethal effects to birds are possible.

2.3.2 Barton Springs Salamander

The Agency has also completed effects determinations for the Barton Springs salamander for three pesticides including atrazine (U.S. EPA 2006b), metolachlor (U.S. EPA, 2007a), and diazinon (U.S. EPA, 2007b) as part of the settlement for the court case *Center for Biological Diversity and Save Our Springs Alliance v. Leavitt, No. 1:04CV00126-CKK (filed January 26, 2004).* The results of these three endangered species risk assessments show that atrazine, metolachlor, and diazinon have either no effect or are not likely to adversely affect the Barton Springs salamander by direct toxic effects and/or indirect effects resulting from effects to aquatic invertebrates and plants.

2.4 Stressor Source and Distribution

2.4.1 Environmental Fate and Transport Assessment

Simazine is moderately soluble in water at 20°C with a solubility of 3.5 mg/L. Based on laboratory studies, simazine could persist for several months ($t_{1/2} = 91$ days; aerobic soil metabolism) in the environment and maybe for years in oxygen deprived aquatic systems ($t_{1/2} = 664$ days; anaerobic aquatic metabolism), as it is not easily degraded by soil microbial organisms. If released on soil surface and under direct sunlight, it will undergo relatively faster degradation ($t \frac{1}{2} \approx 22$ days). Simazine is also quite resistant to aqueous abiotic reactions (stable to hydrolysis at pH 5, 7, and 9 and to photolysis in buffered solution at pH 7), thus increasing its likelihood to runoff and contaminate surface water. However, it must be noted that a supplemental aqueous photolysis study showed simazine degrading with a half-life of 16 hours in the presence of acetone as a sensitizer.

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¹ Available via the internet at: http://www.epa.gov/oppsrrd1/reregistration/REDs/simazine_red.pdf

Laboratory adsorption data show low water/soil partitioning for simazine. The Freundlich $K_{d\text{-}ads}$ constants for the adsorption phase were below 5 for all soils tested. Organic matter (OM) seems to affect the sorption efficiency of simazine as the adsorption coefficient was shown to be strongest in clay soil ($K_{d\text{-}ads}$ 4.31, OM 4.8%) and weakest loam soil ($K_{d\text{-}ads}$ 0.48, OM 0.8%). These data indicate that simazine is highly mobile, thus having strong potential to leach to ground water systems, especially in OM poor soil systems, such as loam and sand soils.

Based on its low vapor pressure (6.1 x 10^{-9} mm Hg at 20° C) and Henry's Law Constant (3.2 x 10^{-10} atm·m³/mol at 25° C), volatilization loss of simazine from soil and water systems is expected to be insignificant compared to dissipation by chemical degradation and metabolism. Based on laboratory bioaccumulation in rainbow trout, simazine is not expected to bioaccumulate in fish, which is in concurrence with simazine's low K_{ow} value of 122. The BCF in all tissue tested ranged from 0.9 (viscera) to 2.3 (muscle). Elimination of accumulated residues by day 28 of depuration was 52% in viscera and 98% in muscle.

Based on its persistence and mobility, as demonstrated by the laboratory data, simazine is expected to reach surface water via transport from soil surfaces during runoff events and ground water via vertical movement through soil (leaching). Aside from monitoring data, terrestrial field and aquatic dissipation studies were also submitted for simazine. Unfortunately, most of the terrestrial field studies did not follow the Subdivision N Guidelines and were deemed not acceptable to provide information on the behavior of simazine under actual terrestrial field conditions. Two supplemental studies, however, indicated that simazine could persist in the fields for over one month to several years depending on soil texture and soil temperature. In addition, a non-guideline study on simazine persistence in soil as a function of temperature and soil moisture (MRID 00027881) also indicated that although decreasing soil moisture slows simazine's metabolism rate in soil, soil temperature exerted the greatest influence in the breakdown of simazine by microbes: a decrease in soil temperature from 25 to 15°C (with other factors remaining constant) could increase simazine's half-life by up to 250 to 300%. As for aquatic field studies, dissipation of simazine is variable, with half lives ranging from 12 days in swimming pool water, to 53 days in surface water man-made ponds, and up to 700 days in a lake in Missouri. The fast degradation of simazine in the swimming pool water study could be attributed partially to photodegradation, which was seen in laboratory studies to accelerate in the presence of photosensitizers or chemical species (such as hydroxyl radicals) capable of inducing photoreactions.

There are three types of degradates/metabolites for simazine. The first type of degradate is formed via dealkylation of the amino groups, for which mono- and fully dealkylated degradates are known (G-28279 or DIA and G-28273 or DACT). The second type is formed by substitution of the chloro group by a hydroxyl group (G-30414 or hydroxysimazine, HA). The third type is formed by substitution of the chloro group by a hydroxyl group together with partial or complete dealkylation (GS-17791 and GS-17792). From limited laboratory data, the relative concentrations of the degradates in soil were generally DIA>DACT~Hydroxysimazine, except for one aerobic soil metabolism

study and one aerobic aquatic metabolism study, where the concentration of hydroxysimazine was higher than that of DIA towards the end of the studies. The highest detected concentration of DIA in the laboratory studies was approximately 10% of total applied radioactivity (aerobic soil metabolism study) and less than 5% on soil surfaces of two supplemental terrestrial field studies, which indicates that DIA, and subsequently DACT and hydroxysimazine, may not form and persist in the environment at any substantial levels.

Like parent simazine, the dealkylated degradates are very mobile in the sand soil and the loam soil, as shown by their low (<2) adsorption coefficients (K_{ads}). Mobility for these dealkylated degradates, however, appears to decrease in soil with higher clay content (K_{ads} in clay soil range from 1.56 to 4.3). Therefore, although laboratory studies indicate that the dealkylated degradates are as likely (or even more likely) to leach to ground water as parent simazine, as with simazine, soil characteristics must be taken into account when assessing the leaching potential of these degradates in a specific region. Hydroxysimazine, on the other hand, shows the strongest adsorption to soil, with K_{ads} values of 8 in sand to 480 in clay soil, thus possessing lower leaching potential than its parent. Acceptable field dissipation studies are not currently available to confirm the laboratory findings on the mobility of these degradates.

In summary, simazine is somewhat persistent and mobile in soils and has the potential to reach surface water and ground water via run off and leaching, respectively. When present in ground water and in surface water, simazine will further persist, especially in systems with relatively long hydrologic residence times (such as in some reservoirs), mostly due to its resistance to abiotic hydrolysis and to direct aqueous photolysis, its susceptibility to biodegradation, and its limited volatilization potential. For simazine degradates such as DIA and DACT, laboratory and field studies indicate that their concentrations in the environment are likely to be insignificant compared to parent simazine.

The relatively low soil/water partitioning of simazine and its chloro degradates indicates that the concentrations of the degradates in/on suspended and bottom sediment in equilibrium with the water column will be somewhat comparable to their parent. In contrast, hydroxysimazine concentration would be much higher. Table 2.1 lists the environmental fate properties of simazine, along with the major and minor degradates detected in the submitted environmental fate and transport studies.

Hydrolysis	stable at pH 4, 7, and 9 @ 20C	none	00027856	Acceptable
Direct Aqueous Photolysis	stable (t ½ >30 days - duration of study) in sterile, unbuffered water irradiated with a mercury vapor lamp.	G-28273 (max 11% TAT at study end)	00143171	Supplemental

Table 2.1 Summary of Simazine Environmental Fate Properties							
Study	Half-lives, Days	Major Degradates Minor Degradates	MRID#	Study Status			
	t ½ ~ 16 hrs in sterile, unbuffered 1% aqueous acetone solution irradiated with artificial light stable (t ½ ~ 382 days) in sterile buffered solution, irradiated with xenon lamp	G-28279 (max 82 % after 98 hr) G-28273, G-30414 and GS-17792 none	42503708	Acceptable			
Soil Photolysis	22 days (corrected for dark control, 12-hr irradiation)	none G-30414, G-28279, G- 28273, and GS-17792.	40614410	Supplemental/ Unacceptable			
Metabolism		G-28279 (max 10% at day 60) G-30414, G-28273, GS-17792, G-28516, GS-17791, and CO ₂	00158638	Supplemental			
		GS-30414 (max 62% at study end) <i>GS-17792 and GS-28279</i>	43004501	Supplemental			
Anaerobic Soil Metabolism	56 days (sandy loam)	none G-28279, G-30414, G- 28273, and GS-17792	00027857	Supplemental			
Anaerobic Aquatic Metabolism	664 days (sandy clay)	none G-30414, G-28279, and G- 28273	40614411	Acceptable			
Aerobic 61 (sediment), 109 (water), and 71 Aquatic days (total system) Metabolism		G-30414 (max 12% day 30) G-28279, G-30044, and G- 31709	43004502	Acceptable			
K _{d-ads} / K _{d-des} (mL/g) 4.3/9.3 (clay), 0.7/2.3 (sand), 1.3/6.2 (sandy loam), and 0.5/0.8 (loam)		NA	41442903 41257903	Acceptable			
$ \begin{array}{c c} K_{\text{oc- ads}} / K_{\text{oc-des}} \\ (mL/g) & 153 / 331 (\text{clay}), 123/426 (\text{sand}), \\ 114/555 (\text{sandy loam}), \text{and} \\ 103/167 (\text{loam}) & \end{array} $							
Terrestrial Field Dissipation	186 days (bareground, MN) max 0.13 ppm in 12-18" at day 270	G-28279 and G-30414 (6- 12")	40614417	Unacceptable			
Dissipation	149 days (bareground, CA) 0.56 ppm in 6-12" at day 564	G-28279 (max 0.16 ppm 6- 12" day 269) and G-30414 (max 0.57ppm 18-24" day 564)	40614418	Unacceptable			
	33 days (citrus crop, FL) 0-8"	G-28279 (max 0.28 ppm 0-	40634201	Unacceptable			

Table 2.1 Summary of Simazine Environmental Fate Properties							
Study Half-lives, Days		Major Degradates Minor Degradates	MRID#	Study Status			
	44 days (bareground, FL) 0-8"	8" day 19) and G-30414 (max 0.01ppm 0-8" day 91) G-28279 (max 0.39 ppm 0- 8" day 18) and G-30414 (max 0.52ppm 0-8" day 30)					
	26 days (citrus crop, FL) 0-8"	G-28279 (max 0.24ppm 0- 8" day 15) and G-30414 (max 1.4ppm 0-8" day 15)	40634202	Unacceptable			
	15 days (bareground, FL) 0-8"	G-28279 (max 0.31ppm 0- 8" day 15) and G-30414 (max 0.83ppm 0-8" day 31)		Supplemental			
	119 days (raspberries, OR) 0-8" 125 days (bareground, OR) 0-8"	G-28279 (max 1.1ppm 0-8") and G-30414 (max <0.09ppm 0-8")	40614413 40614414	Unacceptable			
	110 days (corn plot, MO) 0-8" 101 days (bareground, MO) 0-8"	G-28279 (max <0.2ppm 0- 8") and G-30414 (max <0.24ppm 0-8")	40614415 40614416	Unacceptable			
480 days (Nebraska) 12-24"		Not analyzed	00027863	Supplemental			
Aquatic Field	60 to 700 days in lakes	G-28279	00027829	Supplemental			
Dissipation	12 days in GA swimming pool water	G-28279 and G-30414	40614420	Supplemental			
	53 days in IA man-made pond water	G-28279 and G-30414	40614422	Supplemental			

G-28279 = DIA/CEAT; G-28273 = DACT; G-30414 = Hydroxysimazine

2.4.2 Mechanism of Action

Simazine is part of the triazine herbicide family (including atrazine, cyanazine, propazine) and is very effective at stopping the photosynthetic process in susceptible plants by binding to specific sites within the plant's chloroplasts. Specifically, simazine inhibits photosynthesis via competition with plastoquinone II at its binding site in the process of electron transport in photosystem II.

2.4.3 Use Characterization

Currently, Syngenta Crop Protection, Inc. is the primary manufacturer of simazine; however, there are about 13 registrants with active registrations. Syngenta Crop Protection, Inc. is supporting the majority of the uses (Princep Caliber 90®, Princep®). Other registrants and products include Atanor S.A. (Simazina Atanor), Chem-Real Investment Corp., Ciba, Ltd. (Gesastop®, Princep®), Drexel Chemical Co. (Drexel®)

Simazine), Helm AG, Makhteshim-Agan (Simanex®), Micro-Flo Co., OXON Italia S.P.A., Platte Chemical Co., Sanachem (Pty) Ltd., Sanonda Co. Ltd., Sostram Corp. (Sim-Trol®), Terra International, Inc., and Tecomag (Nezitec®).

An analysis of available use and land cover information, including extensive discussions with local experts (Davis, 2006; Garcia, 2006; Perez, 2006; see Appendix C of the atrazine Barton Springs salamander assessment: U.S. EPA, 2006b for more detail) in the fields of agriculture and soil science, was completed to determine which simazine uses are likely to be present in the action area. This evaluation is intended to place priority on those simazine use areas likely to be in closest proximity to the salamander's habitat. The analysis indicates that, of all registered uses for simazine, the non-agricultural uses are likely to result in the highest exposures to the salamander. This is due to the lack of simazine use sites present in the area and the fact that very little agricultural crops other than a single orchard operation are actually grown in the action area. However, all uses determined to be present in the action area defined by the hydro-geologic framework of the karst aquifer system have been assessed including orchard, golf course/recreational turf, non-cropland, and residential turf uses.

Nationally, simazine is widely used as a selective herbicide to control most annual grasses and broadleaf weeds (before they emerge or after removal of weed growth) in agricultural crops, such as corn, citrus, fruits, and nuts. The highest single application rate for these uses is 9.6 lb ai/A on Florida citrus, although this use is not relevant to this site-specific assessment in Texas. Application can be made via ground sprayer, banded application, broadcast, or via granular formulations. In addition, simazine can also be aerially applied to non-cropland at a rate of 5.0 lb ai/A

Based the available national usage data, the Agency's Biological and Economic Analysis Division (BEAD) estimated that approximately 4.5 -5 million pounds of simazine was used on agricultural crops in the US during 1997 - 2001, and approximately 1.2 million pounds on non-agricultural crops. The Corn Belt of the Midwest (Illinois, Indiana, Kentucky, Missouri, and Ohio) comprised the majority (1.5 million pounds) of the estimated crop usage of simazine. Approximately 637,000 pounds was used on Florida citrus (85% on oranges and 15% on grapefruit) and 606,000 pounds on California fruits and nuts (90% on almonds, grapes, oranges, and walnuts). Non-agricultural use accounted for about 1.2 million pounds of simazine, of which roughly 10% (or 3% of total annual simazine use) represents the 8 lb ai/A granular use on rights-of-way, industrial sites, highways, etc. It should be noted, however, that the 8 lb ai/A granular use of simazine has been cancelled.

A national map (Figure 2.1) showing the estimated poundage of simazine uses across the United States is provided below. On the county level, simazine use is heaviest in the Central valley of CA where mostly almonds, nuts, fruits, and citrus are grown and in Florida with turf and citrus.

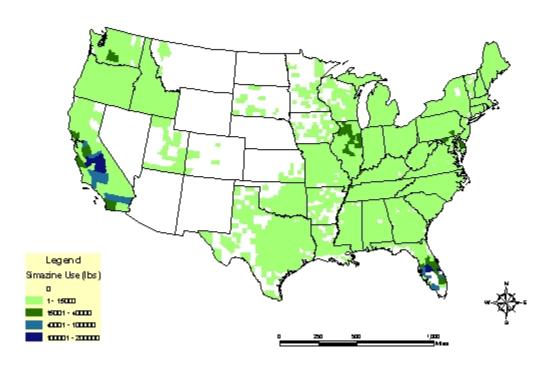


Figure 2.1 Simazine Use in Total Pounds per County

Critical to the development of appropriate modeling scenarios and to the characterization of application rates used is an assessment of local usage information. The Agency's Biological and Economic Analysis Division (BEAD) provided an analysis of both national and local use information for simazine (Kaul et al., 2005; Kaul and Carter, 2005; Zinn and Jones, 2006; Kaul, et al., 2006). State level usage data were used to calculate county-level usage because no reliable county level data are available for Texas. State usage data were obtained from USDA-NASS² and EPA proprietary data³ sources. Data from both sources were averaged together over the years 2000 to 2004 to calculate average annual usage statistics by state and crop for simazine, including pounds of active ingredient applied and percent of crop treated.

Because no reliable county level usage data are available for Texas, average annual pounds applied and acres treated by county were calculated by apportioning the estimated

17

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 US EPA proprietary usage databases provide estimates of pesticide usage for select agricultural use sties by chemical, crop and state.

state level usage to counties based on the proportion of total state acres grown of each crop in each county. The most recently available acreage data were obtained from USDA's 2002 Census of Agriculture.

In this analysis, the Agency gathered information on the agricultural uses of simazine in the three counties (Hays, Travis, and Blanco) located within or adjacent to the action area for simazine in the context of the Barton Springs salamander. Typically, information is derived for crops for which simazine is registered, including amounts of simazine used by county, application rates, methods of application, application timing, and intervals between applications. However, given the low reported usage in this part of Texas and the limited agricultural and non-agricultural uses, only a limited amount of information was available to estimate the total pounds of simazine applied. Given the limited nature of the data, estimates of typical application rates, timing, or percent treated could be not be determined.

Subsequent information based on land cover data (City of Austin, 2003a and b; USGS, 2003) and discussions with local experts (Davis, 2006; Garcia, 2006; Perez, 2006) indicates that most of the agricultural commodities listed above are actually grown to the east of the action area and thus are not included in this assessment. However, additional analysis, completed as part of the diazinon Barton Springs salamander assessment (U.S. EPA, 2007b), indicates that a single orchard operation is located within the action area.

Land cover data also suggest that many of the currently registered non-agricultural simazine uses could not be excluded from the assessment. However, the non-agricultural forestry use of simazine on conifers was not evaluated as part of this assessment because forest land cover data from the U.S. Geological Survey and the U.S. Forest Service indicate that pine plantations are not present within the action area for the Barton Springs salamander (http://nationalatlas.gov/atlasftp.html). Based on this analysis, a suite of scenarios was developed relevant to this assessment, including a single agricultural scenario (orchard for the lone peach orchard in the action area) and three non-agricultural scenarios (rights-of-way representing non-cropland, turf, and residential uses) using local land cover, soils, and agronomic and climatic data specific to Travis and Hays counties in Texas.

Locally, county level estimates of simazine were derived using state level estimates from USDA-NASS and EPA proprietary data. State level data from 1998 to 2004 were averaged together and extrapolated down to the county level based on apportioned to county level crop acreage from the 2002 USDA Agriculture of Census (AgCensus) data. Statewide, the Screening Level Estimate of Agricultural Uses prepared by BEAD for simazine suggests that 60,000 pounds were applied to grapefruit, 20,000 pounds were applied to oranges, and 1,000 pounds of simazine were applied to peaches. Of these uses, both grapefruit and oranges are grown in the southern Rio Grande valley and are not associated with the action area. Specifically for the action area, the information developed by BEAD for the three county area (Hays, Travis, and Blanco) suggests that only 138 lbs of simazine were used on peaches and pecans on a total of 19 acres.

Given the low rates noted above, no typical usage information is available for simazine in the action area.

2.5 Assessed Species

The Barton Springs salamander is aquatic throughout its entire life cycle. As members of the Plethodontidae Family (lungless salamanders), they retain their gills, and become sexually mature and eventually reproduce in freshwater aquatic ecosystems. The best available information indicates the Barton Springs salamander is restricted to the four springs outlets that make up the Barton Springs complex (Figures 2.2 and 2.3), located in Zilker Park near downtown Austin, Texas. As such, this species has one of the smallest ranges of any vertebrate species in North America (Chippindale, 1993). The Barton Springs segment of the Edwards Aquifer and its contributing zone supply all of the water in the springs that make up the Barton Springs complex. Flows of clean spring water are essential to maintaining well-oxygenated water necessary for salamander respiration and survival.

The subterranean component of the Barton Spring salamander's habitat may provide a location for reproduction (USFWS, 2005); however, little is known about the reproductive biology of the Barton Springs salamander in the wild. It appears that salamanders can reproduce year-round, based on observations of gravid females, eggs, and larvae throughout the year in Barton Springs (USFWS, 2005). Survey results indicate Barton Springs salamanders prefer areas near the spring outflows, with clean, loose substrate for cover, but they may also be associated with aquatic plants, especially moss. In addition to providing cover, moss and other aquatic plants harbor a variety and abundance of the salamander's prey, freshwater invertebrates. Based on available information, both adults and juveniles eat freshwater invertebrates (USFWS, 2005).

Further information on the status and life history of the Barton Springs salamander is provided in Appendix C.

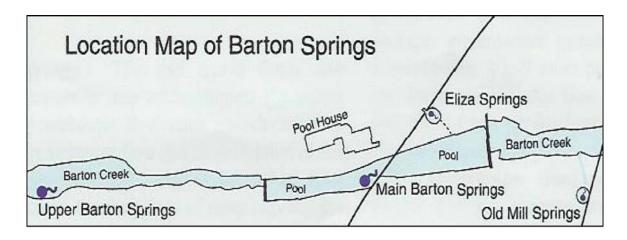


Diagram from Hauwert et al., Barton Springs Edwards Aquifer Conservation District Report

Figure 2.2 Location Map of Barton Springs (dots with curved line indicate location of springs)



Figure 1.3 Aerial Photo of Barton Springs

2.6 Action Area

For listed species assessment purposes, the action area is considered to be the area affected directly or indirectly by the federal action and not merely the immediate area involved in the action (50 CFR 402.02). It is recognized that the overall action area for the national registration of simazine uses is likely to encompass considerable portions of the United States based on the large array of both agricultural and non-agricultural uses. Based on the available simazine monitoring data (discussed further in Section 3.2.4) and the toxicity data for the most sensitive non-vascular aquatic plant, the Agency's LOCs are likely to be exceeded in many watersheds that are in proximity or downstream of simazine use sites. Therefore, the overall action area for simazine is likely to include many watersheds of the United States that co-occur and/or are in proximity to agricultural and non-agricultural simazine use sites. However, in order to focus this assessment, the scope limits consideration of the overall action area to those geographic portions that may be applicable to the protection of the Barton Springs salamander as they occur within hydrogeologic framework of Barton Springs. Deriving the geographical extent of this portion of the action area is the product of consideration of the types of effects simazine may be expected to have on the environment, the exposure levels to simazine that are associated with those effects, and the best available information concerning the use of simazine and its fate and transport within Barton Springs.

Unlike exposure pathways for most aquatic organisms, where stressors are transported via surface water to the receptor within a defined watershed, the Barton Springs salamander resides in a unique environment in which the source of the water, hence the stressor, reaches the salamander via subsurface flow. Thus, the fate and transport of simazine is an important factor in defining the action area for the Barton Springs salamander. The fate profile (see Section 2.4.1) describes why runoff from treated fields, transported through the fractured limestone of the Edwards Aquifer, is considered the principal route of exposure for the salamander. Thus, the action area for this assessment is defined by those areas within the hydrogeologic "watershed" that drain to the springs. In this case, the area draining to the springs is defined by the subsurface geologic framework as opposed to surface hydrology. Figure 2.4 depicts the extent of the action area based on this hydrogeologic framework. More detail on the definition of the action area follows.

The Barton Springs salamander is known to inhabit only 4 springs (Main Barton Springs, Eliza Springs, Old Mill Springs, and Upper Barton Springs; see Figures 2.2 and 2.3), located in the Barton Springs Segment of the Edwards Aquifer (BSSEA), and associated subterranean areas in the aquifer itself (USFWS, 2005). Barton Springs, located in Zilker Park near downtown Austin, Texas is an aquifer-fed system consisting of four hydrologically connected springs: (1) Main Springs (also known as Parthenia Springs or Barton Springs Pool); (2) Eliza Springs (also known as the Elks Pit); (3) Old Mill Springs (also known as Sunken Garden or Walsh Springs); and (4) Upper Barton Springs (Pipkin and Frech, 1993). Collective flow from this group of springs represents the fourth largest

spring system in Texas (Brune, 1981). The springs themselves are fed by the BSSEA, and thus groundwater input is the primary determinant of water quality for the salamander. Main Springs supply the water for Barton Springs Pool, and during high groundwater flow conditions, the surface water flow from Barton Creek may enter the pool if it overtops the dam at the upper end of the pool. Thus, any pesticide used in the land areas contributing to the groundwater in the Barton Springs segment of the aquifer or the surface water in Barton Creek could potentially be transported to these areas.

Flow to the Barton Springs is controlled by the geology and hydrogeology of the BSSEA. Numerous geological and groundwater studies (Slade et al., 1986, Hauwert et al., 2004) have been conducted that define the extent of the area contributing to the Barton Springs. The BSSEA represents an approximately 150 square mile portion of the Edwards Aquifer system in central Texas. Within the BSSEA, both surface water and groundwater flow are controlled by the subsurface geology via the fractured nature of the limestone within of the BSSEA. This is particularly relevant for Barton Springs because surface water flow from Barton Creek into the pool system is diverted via a bypass channel upstream from the main pool to limit the input of surface water from Barton Creek. Thus, the dominant source of water to the pool system is subsurface flow.

Subsurface flow in the BSSEA as it relates to Barton Springs is well defined and includes the Barton Creek watershed upstream of the springs accounting for potential surface water inputs into Barton creek. The BSSEA is characterized as a karst system, which permits relatively rapid transit of groundwater, with velocities along the dominant flow path of 1-5 miles/day, depending on groundwater flow conditions (USFWS, 2005) particularly within the fracture portions. Based on dye tracer studies, pesticides applied within the recharge and contributing zones could potentially be present in the water of the springs on a time scale of days to weeks (Hauwert et al., 2004).

Four hydrogeologic zones characterize the BSSEA. These are, from west to east, the Contributing Zone, the Recharge Zone, the Transition Zone, and the Artesian Zone. Of these zones, the Contributing and Recharge Zones have the greatest and most direct influence on Barton Springs. There is evidence that the Transition Zone has some limited input into the Barton Springs, while the Artesian Zone contributes no subsurface flow to the springs (Slade et al., 1985, Hauwert et al., 2004). Further description of the geology and hydrogeology of the BSSEA is provided in Section 3.2.2.

In addition, an evaluation of usage information was completed to determine whether any or all of the area defined by the BSSEA should be included in the action area. Current labels and local use information were reviewed to determine which simazine uses could possibly be present within the defined area. These data suggest that limited agricultural uses are present within the defined area and that non-agricultural uses cannot be precluded. Finally, local land cover data (City of Austin, 2003a and b; USGS, 2003) were analyzed and interviews with the local agricultural sector (Davis, 2006; Garcia, 2006; Perez, 2006; see Appendix C of the atrazine Barton Springs salamander assessment: U.S. EPA, 2006b for more detail) were conducted to refine the characterization of potential simazine use in the areas defined by Hays, Travis, and

Blanco counties. The overall conclusion of this analysis was that while certain agricultural uses could be excluded, and some non-agricultural uses of simazine were unlikely, no areas could be excluded from the final action area based on usage and land cover data. This analysis was a critical piece of developing the overall exposure assessment approach.

Finally, the environmental fate properties of simazine were evaluated to determine which routes of transport are likely to have an impact on Barton Springs. Review of the environmental fate data as well as physico-chemical properties suggests that transport via overland and subsurface flow are likely to be dominant routes. Spray drift and/or long-range atmospheric transport of pesticides could also potentially contribute to concentrations in the aquatic habitat used by the salamander. Given the physico-chemical profile for simazine and the fact that simazine has been detected in both air and rainfall samples, the potential for long range transport from outside the area defined by the BSSEA cannot be precluded, but is not expected to approach concentrations predicted by modeling (see Section 3.2.5). However, because areas where the atmospheric component of simazine loading is considered to be high are typically high use areas (Midwest corn belt), and the area surrounding Barton Springs is not a high use simazine area, the expected loadings from atmospheric transport of simazine are not expected to approach concentrations predicted by modeling (see Section 3.2.5).

Simazine has been documented to be transported away from the site of application by both spray drift and volatilization (Majewski et al., 2000; Majewski and Capel, 1995; Capel et al., 1994, McConnell, et al, 2004, Kuang, et al, 2003, Foreman, et al, 1999, Dubus, et al, 2000). The Agency typically addresses spray drift as a localized route of transport off of the application site in exposure assessments. In the case of the Barton Springs salamander assessment, spray drift is not considered to be a major route of exposure because the source area for simazine reaching the springs is generally removed from the spring system where the salamander resides, and the simazine exposures that reach the springs do so via subsurface flow. Therefore, there is no direct pathway between the application site and receptor for drift to occur (no applications of simazine are reportedly made within the immediate vicinity of the springs). The Agency does not currently have quantitative models to address the long range transport of pesticides from application sites. The environmental fate profile of simazine, coupled with the available monitoring data, suggest that long range transport of volatilized simazine is a possible route of exposure to non-target organisms. The full extent of the action area could hypothetically be influenced by this route of exposure. However, given the amount of direct use of simazine within the immediate area surrounding the species (Kaul, et al., 2006), the magnitude of documented exposures in rain (Majewski et al., 2000; Majewski and Capel, 1995; Capel et al., 1994; McConnell, et al., 2004; Foreman, et al., 1999) at or below available surface water and groundwater monitoring data (as well as modeled estimates for surface water), the extent of the action area is defined by the transport processes of runoff and subsequent overland and subsurface flow for the purposes of this assessment.

Based on this analysis, the action area for simazine as it relates to the Barton Springs salamander is defined by the contributing, recharge, and transition zones within the BSSEA. Figure 2.3 presents the action area graphically.

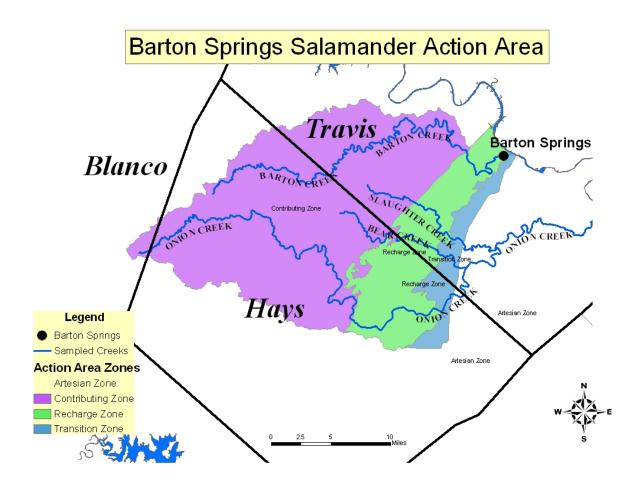


Figure 2.4 Barton Springs Salamander Action Area

2.7 Assessment Endpoints and Measures of Ecological Effect

Assessment endpoints are defined as "explicit expressions of the actual environmental value that is to be protected." Selection of the assessment endpoints is based on valued entities (*i.e.*, Barton Springs salamanders), the ecosystems potentially at risk (*i.e.*, Barton Springs), the migration pathways of simazine (*i.e.* ground water and surface water transport), and the routes by which ecological receptors are exposed to simazine-related contamination (*i.e.*, direct contact in aqueous medium).

Assessment endpoints for the Barton Springs salamander include direct toxic effects on the survival, reproduction, and growth of the salamander itself, as well as indirect effects,

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⁴ From U.S. EPA (1992). Framework for Ecological Risk Assessment. EPA/630/R-92/001.

such as reduction of the prey base and/or modification of its habitat. Each assessment endpoint requires one or more "measures of ecological effect," which are defined as changes in the attributes of an assessment endpoint itself or changes in a surrogate entity or attribute in response to exposure to a pesticide. Measures of ecological effect are evaluated based on acute and chronic toxicity information from registrant-submitted guideline tests, and data from open literature which meets specific acceptance criteria.

Guideline test are performed on a limited number of organisms, which serve as surrogates for other types of organisms expected to have similar responses. Open literature data may expand the number of organisms for which toxicity data are available; however, these studies may or may not have been conducted in accordance with standardized protocols and are often not directly comparable to the guideline tests. The Agency's guidance (U.S. EPA, 2004) specifies that, in absence of data from more closely related species, bird toxicity data is used for terrestrial-phase amphibians and freshwater fish data are used for aquatic-phase amphibians. In order to provide a conservative estimate of potential risk, the most sensitive organism in the representative phylogenic class is used. Barton Springs salamanders are neotenic (retain gills throughout their lives) and are considered aquatic-phase amphibians. No species-specific simazine toxicity data were available for salamanders or other aquatic-phase amphibians at the time of this risk assessment. Therefore, fish data are used as surrogates for the Barton Springs salamander, in accordance with guidance specified in the Agency's Overview Document (U.S. EPA, 2004).

A complete discussion of all the toxicity data available for this risk assessment, including registrant-submitted and open literature data, and the 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 Barton Springs salamander risks associated with exposure to simazine is provided in Table 2.2.

2.8 Conceptual Model

2.8.1 Risk Hypotheses

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

- Simazine in groundwater, surface water, and/or runoff from treated areas may directly affect Barton Springs salamanders by causing mortality or adversely affecting growth or fecundity;
- Simazine in groundwater, surface water, and/or runoff from treated areas may indirectly affect Barton Springs salamanders by reducing or changing the composition of prey populations; and
- Simazine in groundwater, surface water, and/or runoff from treated areas may indirectly affect Barton Springs salamanders by reducing or changing the composition of the plant community in the springs, thus affecting primary productivity and/or cover.

2.8.2 Diagram

The conceptual model is a graphic representation of the structure of the risk assessment. It specifies the stressor, release mechanisms, abiotic receiving media, biological receptor types, and effects endpoints of potential concern. The conceptual model for the potential effects of simazine on the Barton Springs salamander is shown in Figure 2.5. Exposure routes shown in dashed lines are not quantitatively considered because these exposures are expected to be sufficiently low as not to cause direct or indirect effects to the Barton Springs salamander.

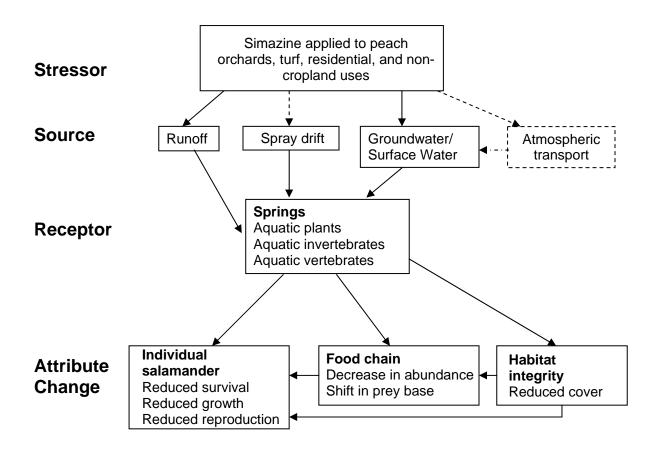


Figure 2.5 Conceptual Model for Barton Springs Salamander

The conceptual model provides an overview of the expected exposure routes for Barton Springs salamanders within the simazine action area previously described in Section 2.6. In addition to freshwater aquatic vertebrates including Barton Springs salamanders, other aquatic receptors that may be potentially exposed to simazine include freshwater invertebrates and aquatic plants. For freshwater vertebrate and invertebrate species, the major routes of exposure are considered to be via the respiratory surface (gills) or the integument. Direct uptake and adsorption are the major routes of exposure for aquatic plants. Direct effects to freshwater invertebrates and aquatic plants resulting from exposure to simazine may indirectly affect the Barton Springs salamander via reduction in food and habitat availability. The available data indicate that simazine is not likely to bioconcentrate in aquatic food items, with fish bioconcentration factors (BCFs) ranging from 0.9 in viscera and 2.3 in muscle (U.S. EPA, 2006a). Therefore, bioconcentration of simazine in salamanders via the diet was not considered as a route of exposure.

Individual Barton Springs salamanders with the greatest potential to experience direct adverse effects from simazine use are those that occur in surface water and/or groundwater with the highest concentrations of simazine. Water passing into, and through Barton Springs comes from groundwater in the Barton Springs Segment of the

Edwards Aquifer. When Barton Creek floods, some of the surface flow enters Barton Springs Pool; however, during normal flow, the water from Barton Creek enters a bypass channel upstream from the main pool and does not enter the pool itself.

Based on historical records of pesticide use in Zilker Park and the area surrounding Barton Springs dating to 1997, simazine has not been used in this area (personal communication with Elizabeth McVeety, pesticide applicator at Zilker Park, April 21, 2006). According to the City of Austin Parks and Recreation Department (PARD) Integrated Pest Management Plan (IPM) (2005), the main concern within the Park is control of fire ants, and spot treatment of Round-up (glyphosate) is the only herbicide specified for control of Johnson grass and poison ivy. Although the IPM does not specifically address simazine use within Zilker Park, it is currently being revised to specifically restrict simazine use within the Park in the future (personal communication with Elizabeth McVeety, pesticide applicator at Zilker Park, July 24, 2006). Given that simazine is not used within the Barton Springs area, it is unlikely that simazine in runoff would indirectly affect Barton Springs salamanders by reducing or changing the composition of riparian zone vegetation and increasing sedimentation of the springs in the main pool. Increased sedimentation in the main pool is more likely to result from high groundwater flow conditions, when the surface water flow from Barton Creek overtops the dam at the upper end of the pool. Therefore, potential indirect effects to Barton Springs salamanders via reduction or change in the riparian zone vegetation (i.e., terrestrial plants) and resulting sedimentation are not considered as a route of exposure and are not further addressed in this risk assessment.

The source and mechanism of release of simazine into surface and groundwater are ground application via foliar spray and coated fertilizer granules to agricultural (i.e., peach orchards) and non-agricultural sites (i.e., turf and rights-of-way, etc). Surface water runoff from the areas of simazine application is assumed to follow topography, resulting in direct runoff to Barton Creek and/or runoff to the recharge area of the Barton Springs Segment of the Edwards Aquifer, where it becomes groundwater that discharges to the surface water of Barton Springs. Additional release mechanisms include spray drift and atmospheric transport via volatilization, which may potentially transport site-related contaminants to the surrounding air. However, spray drift is not considered to be a major route of exposure because the source area for simazine is generally removed from the spring system where the salamander resides, and the simazine exposures that reach the springs do so via subsurface flow. Atmospheric transport is not considered as a route of exposure for this assessment because the magnitude of documented exposures in rainfall are at or below available surface water and monitoring data, as well as modeled estimates of exposure (U.S. EPA, 2006).

2.9 Analysis Plan

The purpose of this assessment is to make an "effects determination" for the Barton Springs salamander by evaluating the potential direct and indirect effects of the herbicide simazine on the survival, growth, and reproduction of this Federally endangered species. This assessment was completed in accordance with the procedures outlined in the

Agency's Overview Document (U.S. EPA, 2004) and the Services' Evaluation Memorandum (USFWS/NMFS, 2004b).

Simazine is used throughout the United States on a number of agricultural crops (primarily corn, citrus, fruits, and nuts) and on non-agricultural sites (including turf, golf courses, industrial sites, rights-of-way). Although the action area for simazine is likely to encompass a large area of the United States, given its use, the scope of this assessment limits consideration of the overall action area to those portions that are applicable to the protection of the Barton Springs salamander. Specifically, the action area for the Barton Springs salamander is defined by the contributing, recharge, and transition zones within the BSSEA. An analysis of simazine use data within the action area indicates that only one single orchard operation is present; therefore, non-agricultural uses of simazine are likely to result in the highest exposures to the salamander.

Screening-level estimates of aquatic exposure are based on PRZM/EXAMS modeling, which assumes a static non-flowing water body. Screening-level EECs were modeled for all aggregate uses of simazine within the action area (i.e., peach orchard, turf, non-cropland, and residential uses) in accordance with the label.

The assessment endpoints for the Barton Springs salamander include direct toxic effects on the survival, reproduction, and growth of individual salamanders, as well as indirect effects, such as reduction of the prey base, and/or modification of its habitat. Direct effects to the salamander are based on available toxicity information for freshwater fish. Given that the salamander's prey items and habitat requirements are dependant on the availability of freshwater invertebrates and aquatic plants, respectively, toxicity information for these taxonomic groups is also discussed.

Comparison of available toxicity information for the degradates of simazine indicates lesser aquatic toxicity than the parent for freshwater fish, aquatic invertebrates, and aquatic plants. Because degradates are not of greater toxicological concern than simazine, concentrations of the simazine degradates are not assessed further, and the focus of this assessment is parent simazine.

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 simazine use within the action area has the potential to adversely affect the Barton Springs salamander via direct toxicity or indirectly based on direct effects to their food supply (i.e., freshwater invertebrates) or habitat (i.e., aquatic plants). When RQs for a particular type of effect are below LOCs, the potential for adverse effects to the Barton Springs salamander is expected to be negligible, leading to a conclusion of "no effect". Where RQs exceed LOCs, a potential to cause adverse effects is identified, leading to a conclusion of "may affect". If a determination is made that use of simazine within the action area "may affect" the Barton Springs salamander, 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" from those actions that are "likely to adversely affect" the Barton Springs salamander.

3. Exposure Assessment

3.1 Label Application Rates and Intervals

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

In the April 2006 RED (U.S. EPA, 2006), EPA stipulated a number of changes to the use of simazine including label restrictions and other mitigation measures designed to reduce risk to human health and the environment. The label changes include cancellation of a number of granular and aerial uses of simazine. In addition, a number of other mitigation measures, including rate reductions, cancellations of certain uses, added spray drift language, and buffer restrictions near streams, rivers, lakes, and reservoirs are proposed. These proposed mitigation measures are expected to become final in 2010. Of the proposed mitigation measures relevant to this assessment that are expected to become final in 2010, all aerial applications and non-residential granular uses will be cancelled and spray drift and buffer restriction language will be added to the labels. The proposed spray drift language includes specific application restrictions for wind speed (< 10 mph), droplet size (coarse or coarser ASAE standard 572 spray), and release height (nozzle height no more than 4 feet above ground or crop canopy). The proposed buffer restrictions prohibit application of simazine within 66 feet of streams and rivers and 200 feet of lakes and reservoirs.

Currently registered non-agricultural uses of simazine within the Barton Springs action area include recreational turf, residential turf, and non-cropland areas defined as industrial sites, highway medians, rights-of-way, lumberyards, tank farms, fuel storage areas, and fence lines. Agricultural uses within the Barton Springs action area include orchard use on peaches. According to use data gathered by EPA, there is no agricultural use of simazine on citrus, tree fruits (other than peaches), berries, and forestry within the Barton Springs action area, although corn, citrus, and tree fruits represent the greatest use nationally. The simazine uses assessed as part of this endangered species risk assessment are summarized in Table 3.1.

Simazine is formulated as liquid, water dispersible granules, wettable powder, emulsifiable concentrate, and granular formulations. Application equipment for the agricultural uses includes ground application (the most common application method), aerial application, band treatment, incorporated treatment, various sprayers (low-volume, hand held, directed), and spreaders for granular applications. Risks from ground boom and aerial applications are considered in this assessment because they are expected to result in the highest off-target levels of simazine due to generally higher spray drift

levels. Ground boom methods of application tend to use lower volumes of application applied in finer sprays than applications coincident with sprayers and spreaders and thus have a higher potential for off-target movement via spray drift.

Orchard (Peach)	4	1	April 1	Liquid	Ground	NA
Turf	1	2	April 1	Granular	Ground	30 days
Non- cropland	5	1	April 1	Liquid	Aerial	NA
Residential	1	2	April 1	Liquid	Ground	30 days

 $¹⁻Based \ on \ 2006 \ RED$ and Label Change Summary Table memorandum dated August 27, 2007 (U.S. EPA, 2007c)

3.2 Aquatic Exposure Assessment

The exposure assessment represents an application of the standard approach outlined in the Overview Document (U.S. EPA, 2004) for the hydrogeologic conditions of Barton Springs. The Agency's PRZM model was used to provide edge of field estimates of exposure, which are assumed to be the concentrations of simazine transported with runoff water directly to Barton Springs via subsurface flow through the fractured limestone of the Edwards Aquifer. Actual conditions are likely to result in lower simazine concentrations through dilution, mixing, retention, and degradation. Available monitoring data from the spring systems were also evaluated and compared with model estimates. While of high quality and targeted to the Barton Springs system, the monitoring data are not considered to be robust in terms of capturing peak simazine concentrations (i.e., the sample frequency is likely to miss the peak concentration).

New regionally-specific PRZM scenarios representing both agricultural and non-agricultural use sites were developed following standard methodology (U.S. EPA, 2005) to capture the upper bounds of exposure. Durations of exposure were used to match available ecotoxicity thresholds. The highest overall exposures were predicted to occur from the residential turf use of simazine while the other uses (non-cropland, turf, orchard) yield exposures that are more than five times lower. None of the assessed uses are in close proximity to the spring system; therefore, direct exposure and spray drift are not anticipated. In general, the exposure assessment yields modeled peak exposure estimates

that are approximately one to two orders of magnitude higher than those seen in monitoring data, while the annual average concentrations are consistent with those seen in monitoring. Intermediate duration exposures (14-day, 21-day, 30-day, 60-day, and 90-day averages) cannot be estimated from the monitoring data due to insufficient sample frequency.

3.2.1 Background

The Barton Springs salamander resides in a geographically limited area defined by a set of spring fed pools in the outskirts of the city of Austin. These pools represent the total aerial extent of the salamander, as defined in Sections 2.5. The pools are a unique system in that they are fed via two sources of water. Surface water has historically reached the pool system via overland flow through Barton Creek. However, water from Barton Creek is currently diverted near the inflow to the pool system and provides only limited input to the pool system during high flow (flood) events. The bulk of the water reaching the pool system is fed via a series of springs. The springs consist of the Main Spring. Upper Spring, Old Mill Spring, and Eliza Spring with approximately 80% of the flow originating from the Main Spring. All of the springs are fed via subsurface flow originating in fractured limestone aquifer of the Edwards Aquifer, which trends southsouthwest away from the pool system. Groundwater from the fractured limestone (karst) is derived from perennial groundwater flow and via recharge that originates from both surface streams and infiltration of rainfall. Therefore, the basic conceptual model of exposure for this assessment focuses on the subsurface pathway delivering groundwater to the pools via the karst system.

The hydrogeology of the Barton Springs Segment of the Edwards Aquifer (BSSEA) defines the action area (see Section 2.6) of simazine use for the Barton Springs salamander. Several hydrogeologic zones define the BSSEA. From west to east, these are the Contributing Zone, the Recharge Zone, the Transition Zone, and the Artesian Zone. The relevance and route of exposure relative to the Barton Springs system is different for each zone and is defined by the geology of the system. Given the basic geology and hydrogeology of these zones within the BSSEA, the Contributing Zone and the Recharge Zone (and to a lesser extent the Transition Zone) are likely to contribute directly to the Barton Springs pool systems. Therefore, land use patterns within these zones were considered to determine the potential for simazine exposure to the Barton Springs salamander. Figure 3.1 shows the extent of the BSSEA.

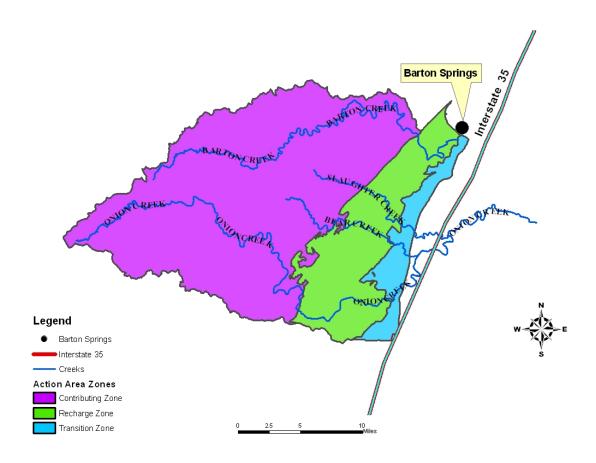


Figure 3.1 Barton Springs Segment of the Edwards Aquifer with HydroZones

Groundwater flow within the Recharge Zone is dominated by subsurface flow via fractured limestone. Numerous studies have been conducted which document the nature of the subsurface geology and the nature and extent of groundwater flow via these fractures (Slade et al., 1986; Hauwert et al., 2004; Mahler, 2005a). Flow within these fractures has been documented to travel from the point of origin to outflow at the springs within hours to days of individual precipitation events, suggesting that simazine reaching the Recharge Zone is likely to have the most immediate impact on Barton Springs.

The Contributing Zone lies due west of the Recharge Zone. In this zone, runoff from sites treated with simazine is transported via overland flow to surface water streams and ponds. Simazine may then be transported via surface water streams to the Recharge Zone, where it is available for infiltration into the network of karst fractures that ultimately feed the Barton Springs system. Unlike stressors originating within the Recharge Zone, some dilution and degradation is expected during this transport process. "Losing" streams (defined as a stream where flow is lost to groundwater recharge) within the Recharge Zone have been reported to provide as much as 85% (Slade et al., 1986) of the annual recharge to groundwater. Historically, surface water flow through Barton Creek has contributed to the loading of water, sediment, and contaminants to the Barton Springs pools. However, in the current configuration of Barton Creek relative to the Barton Springs pools, the creek has been artificially routed past the pools to ensure that the springs are providing the bulk of the recharge to the pools. Occasionally, large precipitation events may result in a bypass of this configuration overflowing of the pool system. In general, the pools are typically fed by groundwater flow through the karst fractures of the Recharge Zone that can receive stressors from both direct infiltration and "loss" from surface water streams.

The Barton Springs system consists of a series of connected pools located within the city limits of Austin, Texas. The Barton Springs salamander has been found within the fractures (springs) feeding the pool system and within the pools themselves. Each receptor location is somewhat unique from the other in how exposures are expected to interact with the salamander.

Exposures to stressors for salamanders residing within the fracture system are due to a combination of base flow with occasional runoff derived from pulses of increased flow. With the increased flow comes the potential for an increase in the magnitude of exposure that is of short duration depending on the climatic event. Base flow within the spring systems is fed by loss of volume from surface streams as they traverse the Recharge Zone of the BSSEA and from groundwater movement out of the Contributing Zone into the fractured limestone of the Recharge Zone. The short term pulsed increases in runoff-derived water through the springs are the result of increased loss through surface streams originating in the Contributing Zone and direct infiltration of precipitation and runoff from surface areas of the Recharge Zone. Thus, salamanders residing within the fracture system of the springs are likely to be exposed to longer-term base flow concentrations of simazine with occasional shorter duration pulses of higher concentrations correlated with precipitation-driven runoff events transported through the fractures.

Salamanders have also been found to reside within the pools themselves. In general, the organisms residing in the pools will be exposed to the same sources of exposure. However, it is expected that the magnitude and duration of exposure will be somewhat different given the tendency of water to move through the pools (except in the most extreme climatic events) more slowly. This suggests that exposures in the pools will be generally lower in magnitude than in the springs, but will also tend to have a longer duration of exposure than in the springs.

Figures 3.2 and 3.3 present the conceptual models of both of these potential exposure pathways. More details on the geology and hydrogeology may be found in the following section. Finally, a more complete description of the Barton Springs pool system in which the salamander resides is provided in Appendix C.

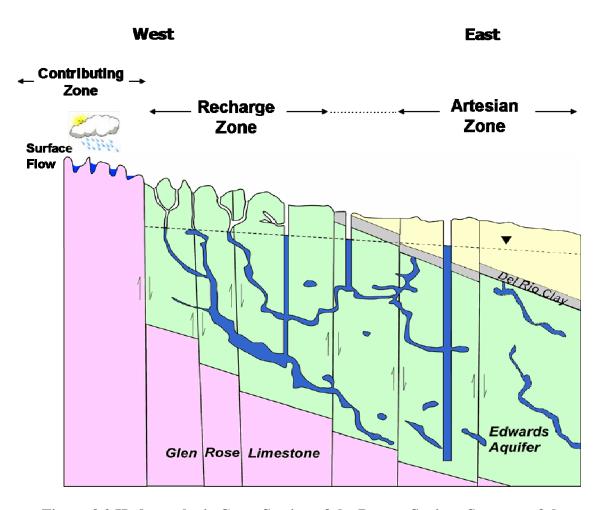


Figure 3.2 Hydrogeologic Cross Section of the Barton Springs Segment of the Edwards Aquifer Showing Dominant Flow Pathways Within Each Hydrozone (Taken from Mahler, 2005a)

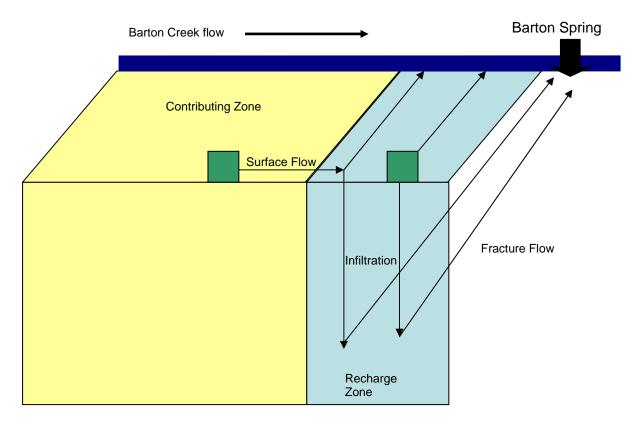


Figure 3.3 Conceptual Model of Surface and Subsurface Flow Within the Barton Springs Segment of the Edwards Aquifer Relative to the Barton Springs Salamander

3.2.2 Geology/Hydrogeology

The Barton Springs pool system lies at the extreme northern end of the BSSEA, which is a portion of a larger fractured limestone aquifer system known as the Edwards Aquifer. The Edwards Aquifer and BSSEA are major sources of groundwater used for drinking water and represent a critical source of water necessary to replenish surface water resources for both recreational and ecological uses throughout the eastern half of Texas.

The Edwards Aquifer is a karst system of limestone and dolomite of Cretaceous age (Slade et al., 1986). The aquifer covers roughly 6,000 square kilometers and stretches from north of Austin to an area southwest of San Antonio. In general, the physical trend of the Edwards Aquifer (and Barton Springs Segment) is south to north, and the carbonate rocks within the aquifer dip to the east except where broken by fractures within the Recharge Zone (Slade et al., 1986). The thickness of the aquifer generally increases from north to south and is typically 400 to 450 feet thick (Slade et al., 1986). It is a principal source of groundwater for drinking water in Texas, and where it discharges to the surface, it is critical for providing freshwater for both recreational and ecological needs.

The Barton Springs Segment extends from the Colorado River south roughly 20 miles into Hays County and covers 391 square kilometers. The Barton Springs Segment is separated from the rest of the Edwards Aquifer by a hydrogeologic divide with groundwater north of the divide flowing north-northeast towards the Colorado River and south of the divide flowing south-southwest. In general, the BSSEA discharges at a number of springs along the Colorado River and Barton Creek. Flow through the BSSEA is typically around 35 cubic feet per second (cfs) during low flow periods, but can reach above 75 cfs during high flow conditions, while the average flow is reported to range between 53 cfs (Hauwert et al., 2004) and 56 cfs (Mahler, 2005a). Slade et al. (1986) also estimated that up to 85% of the recharge reaching the BSSEA was derived from infiltration from the main creeks crossing the Recharge Zone. The remaining infiltration was derived from water coming from minor tributaries and from upland areas in the Contributing Zone and from direct infiltration of precipitation.

Hauwert et al. (2004) conducted dye trace studies of the flow systems in the BSSEA between 1996 and 2002. In these studies, the authors attempted to discern specific flow patterns within the Recharge Zone using dye tracing, mapping of the potentiometric table, water chemistry, local knowledge of geology, and cave mapping. Non-toxic dye injection into caves, sinkholes, and wells was used to define the route of groundwater flow, estimate flow velocities, and approximate travel times. The important finding of this study relative to this assessment is that travel times within the Recharge Zone range from hours up to one week in close proximity to the springs (defined by Travis County), while farther south and west, travel times can increase to approximately 4 weeks. Flow through fractures also may occur within the Transitional Zone that separates the Recharge Zone from the eastern artesian portion of the BSSEA. Figure 3.4 presents a summary of the flow paths defined by this study (Hauwert et al., 2004).

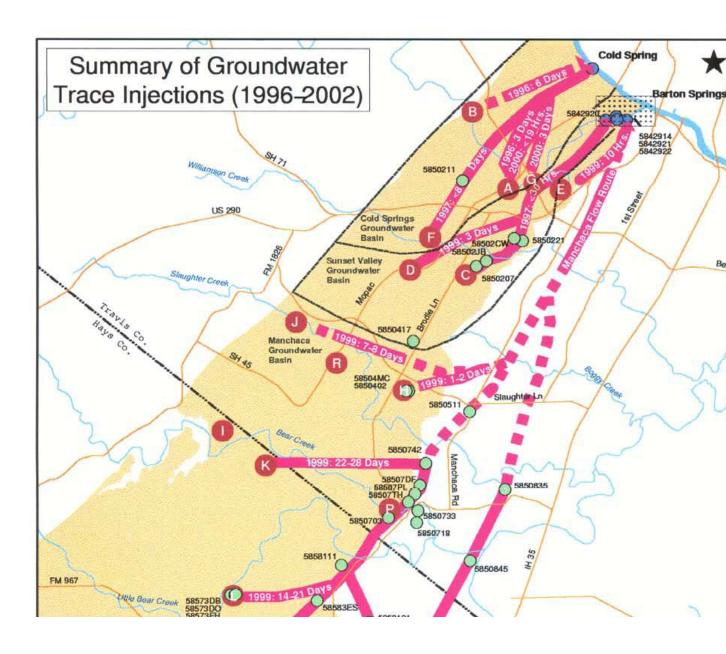


Figure 3.4 Flow paths within Recharge Zone of the Barton Springs Segment of the Edwards Aquifer (Taken from Mahler, 2005a; originally published in Hauwert et al., 2004)

3.2.3 Conceptual Model of Exposure

Given the understanding of the geology and hydrogeology described above, a combination of modeling and monitoring data is needed to assess the potential exposures from simazine to the Barton Springs salamander. Routes of exposure are dependent on

the location of registered use sites for simazine within the action area (defined in Section 2.6 as the Contributing and Recharge Zones), and locations within the pool system (fractures versus pools) where the salamander resides. For instance, uses which are predominantly within the Recharge Zone of the BSSEA result in concentrations in water that is likely to reach the springs via direct transport through the fractures within the karst zone. Uses in the Contributing Zone result in concentrations in water that are transported over longer flowpaths and are subject to both surface and sub-surface transport processes. The interconnected nature of the subsurface network in the BSSEA Recharge Zone can have a major influence on mixing, dilution, storage and degradation of flow (Field, 2004).

Because of the limited nature of the available monitoring data both within the spring network and in the surrounding groundwater and surface water, an analysis of potential use sites within the action area is needed. Available agricultural statistics, land cover data, usage information, and soils data were evaluated relative to the hydrogeologic framework described above. This information was used to determine whether agricultural use sites are present in the Recharge Zone, the Contributing Zone, or both. Analysis of land cover data and usage information suggests that limited agriculture is present in the Contributing and Recharge Zones of the Barton Springs Watersheds.

In order to address the potential for simazine exposure from use on these sites, a suite of PRZM modeling scenarios was developed for the specific agronomic, soil, and climatic data available. As noted above, the action area for the development of the Barton Springs scenarios is comprised of two primary hydrologic zones (in order of importance): 1) the Recharge Zone and 2) the Contributing Zone. Spatial data containing the hydrozone boundaries were obtained from the Barton Springs/Edwards Aquifer Conservation district (ftp://www.bseacd.org/from/HCP Shape Files/). The areas to the east of the Recharge Zone are not considered relevant to the assessment because groundwater flow to the Barton Springs system comes either directly from transport through the Recharge Zone, which occurs generally south to north, or indirectly via the Contributing Zone/Recharge Zone interaction, where flow is dominantly west to east.

This assessment assumes that the estimated environmental concentration (EEC) is derived from both ground water and surface runoff; thus, spray drift is not a factor in the exposure assessment.

3.2.4 Existing Monitoring Data

USGS provided monitoring data for surface streams, groundwater wells, and the four springs making up the Barton Springs system (Mahler, 2005a). Specifically, the data provided long-term trends within all three source types. In addition, recent data from the USGS targeted single runoff events within the spring systems that included high frequency sampling to match the hydrograph correlated with the several specific runoff events.

Four springs were included in the USGS analysis, including Main Spring, Eliza Spring, Upper Spring, and the Old Mill Spring. All four springs represent the main source of inflow into the Barton Springs pool system with the Main Spring providing roughly 80% of overall flow. Sampling and analysis of these springs indicates that the highest detection of simazine was 0.09 µg/L in the Upper Spring while the 95th centile of all samples was 0.03 ug/L. Overall, a total of 135 samples were analyzed for simazine with 56 non-detections for a frequency of detection of 59%. The average of all samples was 0.01 ug/L. The overall picture for simazine in the four springs is one of low level of detection. Given the nature of the flow regime within the springs, it is unlikely that these sampling events have captured the peak exposures, however, the overall pattern suggests that simazine exposures are generally well below 1 ug/L. The bulk of the simazine analysis was conducted after 2000; therefore, evaluation of long-term trends in this data cannot be completed.

Analysis of the stream data indicates that higher concentrations have been detected in surface water; however, the frequency of detection is less than that seen in the springs. Overall, a total of 75 samples were analyzed for simazine with a maximum detection of 1.5 ug/L in 1994. The 95th centile from all samples was 0.1 ug/L and the overall frequency of detection was 25%. More recent data suggests a decrease in exposure in surface water with no sample after 2000 above 0.11 ug/L. Figure 3.5 shows the location of USGS stream samples within the BSSEA.

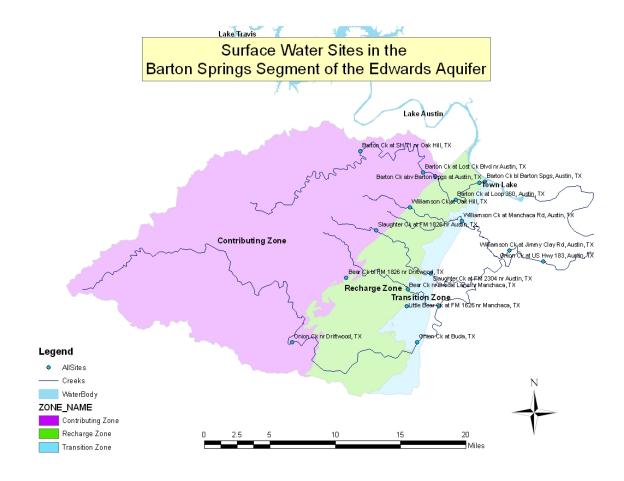


Figure 3.5 Location of USGS Surface Water Sites within the Barton Springs Segment

Statewide surface water data for simazine collected by the USGS NAWQA Program between 1993 and 2003 shows that simazine was detected in 554 out of 863 samples (51 samples were estimated outside the range of quantitation) for a frequency of detection of 64%. The highest detected concentration of simazine in all of the NAWQA samples was $4.86~\mu g/L$ in 2002 from a single site in the Trinity River Study Unit in Dallas county (station ID numbers 08057200) that is outside of the action area for this assessment. Of all detections, only 22 samples were above 1 ug/L and of these all sites were in either urban or mixed watersheds.

Analysis of the well data from the USGS collected between 2000 and 2005 suggests that simazine was detected in groundwater from five locations within the BSSEA. In general, with the exception of a single sample analyzed from well # YD-58-34-414 in 2004 at 0.0224 μ g/L, all simazine detections were below 0.1 μ g/L. Overall, the low frequency and magnitude of detection of simazine suggests that its occurrence in baseflow may be a minimal source of exposure at Barton Springs. Data are also available for two degradates of simazine (DIA and DACT) from groundwater wells within the action area. DIA was detected once out of two samples at 0.0032 ppb and DACT was detected twice out of

seven samples at 0.0122 ppb and 0.0075 ppb. It should also be noted that these degradates are common to both simazine and atrazine, and the source cannot be distinguished from these monitoring data. Figure 3.6 presents the location of the groundwater wells within the BSSEA.

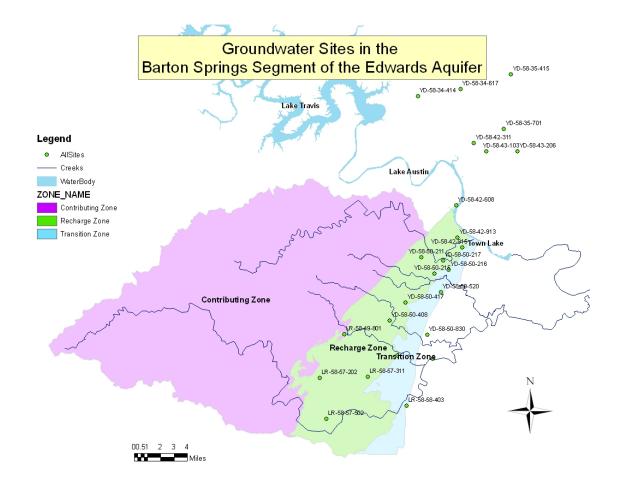


Figure 3.6 Location of USGS Groundwater Sites within the Barton Springs Segment

Overall, the monitoring data provided by USGS indicate relatively consistent low-level concentrations of simazine over time with periodic spikes related to storm-derived runoff events. Because of the limited nature of the runoff-related sampling, it is not possible to determine whether these data are representative of overall peak exposures (Mahler, personal communication, 2005b). Therefore, these data represent a lower bound on exposures and are considered to be representative of long-term baseflow exposure in the spring system.

3.2.5 Modeling Approach

The analysis of available monitoring data and usage information indicates that the exposure assessment cannot rely exclusively on monitoring data. Although of high quality and in selected instances targeted to pesticide use and single runoff events, the

unique nature of flow through the BSSEA and the relationship of the flow regime to the Barton Springs salamander indicates that the exposure assessment should rely on modeling to augment the available monitoring data.

Typically, the Agency conducts modeling using scenarios intended to represent use sites in areas that are highly vulnerable to either runoff, erosion, or spray drift. Runoff estimates predicted by the PRZM model are linked to the Exposure Analysis Modeling System (EXAMS). For ecological risk assessment, the Agency relies on a standard water body to receive the edge of field runoff estimates. The standard water body is of fixed geometry and includes processes of degradation and sorption expected to occur in ponds, canals, and low order streams (e.g. first and second order streams), but with no flow through the system.

The unique geology/hydrogeology of the BSSEA suggests that, for the use sites being evaluated, an estimate of exposure in surface water may not be suitable by itself. If the use site resides exclusively within the Recharge Zone, the principal route of exposure is expected to be either via "edge of field" runoff into an adjacent fracture or via overland flow to a stream which subsequently "loses" some of that flow to the fracture system. In order to provide the most conservative estimate for each scenario modeled, edge of field concentrations are used to represent sources originating with the Recharge Zone (instead of EXAMS concentrations) to mimic the pulsed nature of exposures moving through karst fractures.

Standard Approach for Water Body Modeling. The Agency's standard approach for conducting modeling in support of ecological risk assessment assumes that 100% of a 10-hectare field is covered by the relevant use and that a standard water body adjacent to the field receives the edge-of-field runoff and spray drift. The standard water body is of fixed geometry and includes processes of degradation and sorption expected to occur in ponds, canals, and low order streams (*e.g.* first and second order streams), but with no flow through the system. Modeling scenarios for the 10-hectare field are linked with meteorological data to represent use sites in areas that are highly vulnerable to runoff, erosion, or spray drift. Runoff and spray drift estimates predicted by PRZM (v3.12beta, May 24, 2001) are linked to the Exposure Analysis Modeling System (EXAMS v2.98.04, Jul. 18, 2002) using a graphical user interface or shell (PE4v01.pl, Aug. 13, 2003) to yield 1-in-10-year estimated environmental concentrations (EEC).

The Approach for Barton Springs Modeling. Because of the unique and location-specific nature of the Barton Springs assessment, an approach was taken that incorporated the specific hydrology of the area in an effort to make the modeling approach more relevant than the standard modeling approach that the Agency uses for more generic national-type assessments. The initial screening and refined approaches for modeling exposure are described below.

Initial Screen. In the initial screening approach, the driving assumption was that runoff from a treated field directly infiltrates into the fractured limestone and flows instantaneously into the Barton Springs. Runoff concentrations were determined by

PRZM runs along with a set of relevant Barton Springs PRZM input scenarios. This screening approximation does not consider any mitigating processes that may occur in an actual Barton Springs system that would serve to lower the concentrations such as dispersion, dilution, and degradation within the Karst system. Runoff scenarios from turf, non-cropland, and an orchard, all of which have been shown (as described later) to exist within the action area, were evaluated.

Refined Approach. Because this refined assessment considers the specific hydrologic nature of Barton Springs, a brief description of the Spring's salient features are given here. The Barton Springs are supplied predominantly with water discharging from fractures and conduits formed in the Barton Springs Segment of the Edwards Aquifer (BSSEA) as a result of dissolution of the fractured limestone aquifer over time. Approximately 85% of the water that recharges this aquifer infiltrates through the beds of six creeks that cross the recharge zone (Slade et al., 1986; Barrett and Charbeneau, 1996), with the remaining approximately 15% of the recharge derived from precipitation and recharge in interbed areas in the recharge zone. In the BSSEA, natural ground water discharge occurs primarily at Barton Springs (Lindgren et al., 2004). Recharge features in creek bottoms overlying the recharge zone allow only a limited flow of water during a storm event; therefore, water that is in excess of the flow capacities of recharge features leaves the recharge zone as creek flow. The Contributing Zone encompasses the watersheds of the upstream portions of the six major creeks that cross the Recharge Zone, and therefore provides the source for most of the water that will enter the BSSEA as recharge. These streams gain water, as they flow across the land surface in the Contributing Zone, from the lower-permeability Glen Rose limestone of the adjacent Trinity aquifer (Lindgren et al., 2004). Kuniansky (1989) estimated baseflow discharge from the Trinity aquifer to streams and creeks in this area ranging from 25% to 90% of total flow. In the portion of the Trinity aguifer nearest the contributing zone, this was loosely estimated at 30%. The remainder of water in creeks in the Contributing Zone is derived from precipitation and runoff.

The refined conceptual model attempts to capture the most important aspects of this unique hydrology. In this regard, the nature of the contributing zone and the recharge zone are distinguished and treated separately. Runoff from the recharge zone is assumed to enter the karst environment directly, whereas runoff from the contributing zone is assumed to mix with stream water prior to entering the karst environment of the recharge zone. The long-term average flow volume in the streams in the contributing zone was assumed to be due 30% to aquifer discharge and 70 % to runoff, as is consistent with Kuniansky (1989). Thus surface runoff in the contributing zone mixes with the aquifer discharge flow prior to flowing into the recharge zone.

Masses and volumes of runoff were determined for this assessment from modeling scenarios developed specifically for the orchards and other non-agricultural uses of simazine found in the Barton Springs Salamander action area (see Section 3.2.6). Use on peaches was modeled with the orchard scenario. Similar to the Agency's standard ecological risk assessment methodology described above, 30 years of meteorological data

for the Austin area were linked to these specific scenarios to estimate 1-in-10-year edge of field exposure to potential simazine uses.

A summary of the potential simazine use areas is presented in Table 3.2. Only one operational orchard was identified in the action area. Its area (7 acres) was reported online (http://barsanaorchards.com/news8article.html; Mar. 1, 2007). The use areas are shown to be much smaller than the area where no use occurs (non-use area), the latter of which accounts for roughly 100% of the action area.

Turf	324 (0.14%)	272 (0.12%)	52 (0.023%)
Orchard	7 (0.003%)	7 (0.003%)	0
Non-cropland	453 (0.2%)	390 (0.17%)	63 (0.028%)
Residential turf	8169 (3.61 %)	5381 (2.38 %)	2787 (1.23 %)
Non-use area	226,000	169,000	57,600

^{1 –} Source of data is from City of Austin Land Use Data (2003)

Determination of Runoff Concentrations and Volume. As described previously, the contributing zone and the recharge zone are treated differently. Calculations for the contributing zone are described first and these are followed by calculations for the recharge zone.

Contributing Zone. This refined assessment uses the long-term average stream flow information to calculate an approximate average daily stream flow in the contributing zone. Because the ratio of runoff flow to base stream flow was given by Kuniansky (1989) to be 70:30, knowing the long-term runoff flow enables an estimate of the long-term average streamflow. The long-term (30 years simulated) runoff volume was calculated for each of the scenarios in Table 3.6 using PRZM and the respective areas within the contributing zone. The cumulative runoff volume for the contributing zone was calculated according to

$$V_{CZ} = \sum_{t=1}^{n} \left(V_{CZorchard,t} + V_{CZturf,t} + V_{CZnoncropland,t} + V_{CZresidential,t} + V_{CZnon-use,t} \right)$$
(3.1)

where $V_{CZ} = 30$ year simulated cumulative runoff volume [volume] $V_{CZorchard,t} = orchard runoff volume on day t in the contributing zone [volume] <math>V_{CZturf,t} = turf runoff volume on day t in the contributing zone [volume]$

 $V_{CZ_{noncropland,t}}$ = non-cropland runoff volume on day t in the contributing zone [volume]

 $V_{CZresidential,t}$ = residential runoff volume on day t in the contributing zone [volume]

 $V_{CZ_{non-use,t}}$ = non-use runoff volume on day t in the contributing zone [volume] n = number of days in simulation

The estimated daily aquifer-driven base flow in the streams within the contributing zone was calculated from the 70:30 ratio as given by Kuniansky (1989):

$$V_{base} = \frac{V_{CZ}}{n} \left(\frac{0.30}{0.70} \right) \tag{3.2}$$

where V_{base} = the long-term average daily aquifer-driven stream volume [volume]

Daily runoff volume was calculated by adding the daily runoff flows as follows:

$$V_{CZ,t} = V_{CZorchard,t} + V_{CZturf,t} + V_{CZnoncropland,t} + V_{CZresidentail,t} + V_{CZnon-use,t}$$
(3.3)

where $V_{CZ,t}$ = the total runoff volume on day t in the contributing zone [volume] $V_{CZ,t}$ = the volume for scenario i on any day t in the contributing zone [volume]

Daily stream volume was calculated by adding the base stream flow to the daily runoff volume as follows:

$$V_{stream.t} = V_{CZ.t} + V_{base} \tag{3.4}$$

where $V_{\text{stream},t}$ = the total stream volume on day t in the contributing zone [volume]

The concentration in runoff in the contributing zone was calculated directly from the PRZM output and the area of the scenarios as follows:

$$C_{CZ,t} = \frac{\left(M_{CZorchard,t} + M_{CZturf,t} + M_{CZnoncropland,t} + M_{CZresidential,t}\right)}{\left(V_{CZ,t}\right)}$$
(3.5)

where $C_{CZ,t}$ = the concentration in runoff across the contributing zone on any day t [mass/volume]

 $M_{CZi,t}$ = the mass of simazine in runoff in the contributing zone for scenario i on any day t [mass]

Daily stream concentrations were calculated from the PRZM output, the area of the scenario, the stream base flow, and the average base flow concentration as follows:

$$C_{stream,t} = \frac{\left(C_{CZ,t} \times V_{CZ,t} + C_{base} \times V_{base}\right)}{V_{stream,t}}$$
(3.6)

where $C_{\text{stream},t}$ = the concentration in contributing zone streams on any day t [mass/volume]

C_{base} = the average concentration monitored in base flow [mass/volume]

Note that the background concentration in base flow was assumed to be negligible, but in order to be protective was represented by a concentration of 0.1 ug/L. This concentration is consistent with detected concentrations of simazine in both the stream and groundwater well data specific to the action area.

The above calculated stream volume ($V_{\text{stream,t}}$) in Eqn. 3.4 along with its associated concentration ($C_{\text{stream,t}}$) in Eqn. 3.6 are assumed to be delivered to the recharge zone where they will mix with recharge zone runoff as described next.

Recharge Zone. Runoff originating in the recharge zone was determined in a similar manner as for the contributing zone:

$$V_{RZ,t} = V_{RZorchard,t} + V_{RZturf,t} + V_{RZnoncropland,t} + V_{RZresidential,t} + V_{RZnon-use,t}$$
(3.7)

where V_{RZ} = runoff volume on day t in the recharge zone [volume]

 $V_{RZorchard,t}$ = orchard runoff volume on day t in the recharge zone [volume]

 V_{RZturf} = turf runoff volume on day t in the recharge zone [volume]

 $V_{RZnoncropland}$ = non-cropland runoff volume on day t in the recharge zone [volume]

 $V_{RZresidential} = residential runoff volume on day t in the recharge zone [volume]$

 $V_{RZ_{non-use}}$ = non-use runoff volume on day t in the recharge zone [volume]

The concentration of runoff in the recharge zone was determined from the PRZM mass output (output as mass/area), the area represented by the scenario, and the volume of runoff in the recharge zone as follows:

$$C_{RZ,t} = \frac{\left(M_{RZorchard,t} + M_{RZturf,t} + M_{RZnoncropland,t} + M_{RZresidential,t} + M_{RZnon-use,t}\right)}{V_{RZ,t}}$$
(3.8)

where $C_{RZ,t}$ = the concentration in runoff across the recharge zone on any day t [mass/volume]

 $M_{RZi,t} =$ the mass of simazine in runoff in the recharge zone for scenario i on any day t [mass]

Barton Springs Daily Concentrations. It is assumed that the stream flow from the contributing area and the runoff from the recharge area mix and flow through the karst and into the Barton Springs. Stream flow that does not ultimately pass through the Barton Springs is assumed not important because of the assumption of instant mixing of

simazine residues in flow volumes prior to potential diversion. The discharge in losing streams that remains and leaves the action area is expected to carry the same simazine concentrations as were lost to karst fractures and introduced to the Barton Springs. Therefore, neglecting loss processes, the total discharge produced is determined as:

$$V_{Springs,t} = V_{stream,t} + V_{RZ,t}$$
 (3.9)

where $V_{Springs,t}$ = the total flow through the Barton Springs on day t [volume]

Using these calculations, runoff from the recharge zone provides 11% of discharge through the Barton Springs, on average. This is similar to the approximation by Slade et al. (1986) and Barrett and Charbeneau (1996) that 15% of recharge to the Barton Springs originates in the recharge zone.

Finally, the concentration in the Barton Springs is determined from:

$$C_{Springs,t} = \frac{C_{RZ,t}V_{RZ,t} + C_{stream,t}V_{stream,t}}{V_{Springs,t}}$$
(3.10)

where $C_{Springs,t} =$ the daily concentration in Barton Springs [mass/volume]

Daily EECs in the Barton Springs were post-processed (see U.S. EPA 2007b for details) in order to provide durations of exposure. Peak, 14-day, 21-day, 30-day, 60-day, and 90-day average concentrations were calculated across 30 years of daily EEC values. In order to match the standard PRZM/EXAMS output, the maximum values for each of the 30 years of daily and rolling averages were ranked and the 90th percentiles from the rankings were selected as the final 1-in-10-year EECs for use in risk estimation.

3.2.6 Model Inputs

EECs from surface water sources were calculated using the Agency's Tier II PRZM model. PRZM is used to simulate pesticide transport as a result of runoff and erosion from a standardized watershed. The linkage program shell, PE4v01.pl, which incorporates the site-specific scenarios developed by the Agency, was used to run PRZM (U.S. EPA, 2005). However, new, site-specific scenarios were developed for use in this assessment. Linked site-specific use scenarios and meteorological data are used to estimate exposure for each modeling scenario. Weather and agricultural practices are simulated over 30 years to estimate the 1 in 10 year exceedence probability at the site.

Further information on these models may be found at:

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

The appropriate PRZM input parameters were selected from environmental fate data submitted by the registrant and in accordance with US EPA-OPP EFED water model

parameter selection guidelines, Guidance for Selecting Input Parameters in Modeling the Environmental Fate and Transport of Pesticides, Version 2.3, February 28, 2002. These parameters are consistent with those used in both the 2006 simazine RED (U.S. EPA, 2006a) and the cumulative triazine risk assessment (U.S. EPA, 2006c); no new environmental fate data were incorporated into this assessment. The date of first application was identified based on several sources of information including data provided by BEAD, crop profiles maintained by the USDA, and conversations with local experts. More detail on the crop profiles and the previous assessments may be found at:

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

http://www.epa.gov/pesticides/reregistration/simazine/

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

A summary of the model inputs used in this assessment are provided in Table 3.3.

Molecular Weight	202 g/mole	Product Chemistry
Henry's constant	$3.2 \times 10^{-10} \text{ Pa m}^3 / \text{ mole}$	Product Chemistry
Vapor Pressure	$6.1 \times 10^{-9} \text{ torr}$	Product Chemistry
Solubility in Water	3.5 ppm	Product Chemistry
Photolysis in Water	stable	00143171 42503708
Aerobic Soil Metabolism Half-lives	$t_{1/2} = 130 \text{ days}$ (upper 90th percentile confidence bound on mean half-life of 110 and 91 days)	00158638 43004501
Hydrolysis	stable	00027856
Aerobic Aquatic Metabolism (water column)	$t_{1/2} = 213$ days (input value is three times the single laboratory aerobic aquatic metabolism half-life of 71 days)	43004502
Anaerobic Aquatic Metabolism (benthic)	t _{1/2} = 168 days (input value is three times the single laboratory anaerobic aquatic metabolism half-life of 56 days)	40614411
Кос	123 (average of 152.5, 123.3, 114, and 102.7)	41442903 41257903
Application Efficiency	99 % for ground	default value ²

Table 3.3 Summary of PRZM/EZAMS Environmental Fate Data Used for Aquatic Exposure Inputs for Simazine Endangered Species Assessment for the Barton Springs Salamander

Fate Property	Value	MRID (or source)
Spray Drift Fraction ¹	1 % for ground	default value ²

^{1 –} Spray drift not included in final EEC due to edge-of-field estimation approach

3.2.7 PRZM Scenarios

A total of four use scenarios previously developed were used to provide exposure estimates in this assessment: orchard, turf, residential, and non-cropland (using right-of-way scenario) uses. Each scenario used meteorological data from a weather station located in Austin, Texas. No weather station closer to the action area provides the data required for exposure modeling. A discussion of each assessed exposure scenario is provided below.

3.2.7.1 Orchard

This scenario is intended to represent an orchard that may include cultivation of peaches, nectarines or pecans. USDA data for Hays and Travis counties do not include harvest data for these crops from 1990-2007 (USDA, 2007); however, the 2002 agricultural census for the two counties includes over 2000 acres of land in orchards (USDA, 2002). Discussions with extension agents in Hays and Travis counties indicate that some cultivation of peaches occurs in the BSSEA specifically in Hays County (Bryan Davis, personal communication). Crop parameters for this scenario were chosen to be reflective of a peach orchard in this area.

3.2.7.2 Turf

The turf scenario was developed consistent with the current PRZM scenarios for turf in Pennsylvania and Florida (no pre-existing turf scenario for Texas is available). This scenario is intended to represent turf areas (golf courses, parks, sod farms, and recreational fields) in the BSSEA. Because golf courses are expected to be the most likely turf areas where pesticides may be applied, much of this scenario has been parameterized to be reflective of golf course turf. NASS data for 1997 and 2002 (USDA 1997, 2002) contained no record of sod harvest in either Hays or Travis counties. Since there are several golf courses located within the BSS (City of Austin, 2003b), this scenario was parameterized to represent turf on golf courses and may be generally representative of other potential turf areas. Crop parameters are based primarily on bermudagrass (*Cynodon* spp.) because it is a primary turf grass for golf courses and athletic fields. For turf, it was assumed that 100% of the watershed feeding the fractures is represented by turf.

^{2 –} Inputs determined in accordance with EFED "Guidance for Chemistry and Management Practice Input Parameters for Use in Modeling the Environmental Fate and Transport of Pesticides" dated February 28, 2002

3.2.7.3 Non-cropland

An analysis of landcover data was completed to evaluate the extent of rights-of-way (ROW) within the action area. Available landcover data (City of Austin, 2003b) was separated into relevant classes for rights-of-way analysis including roads and utility lines, railroads, and pipelines. Each separate landcover class was added to a map of the action area previously defined. The extent of these landcover types is presented in Figure 3.7.

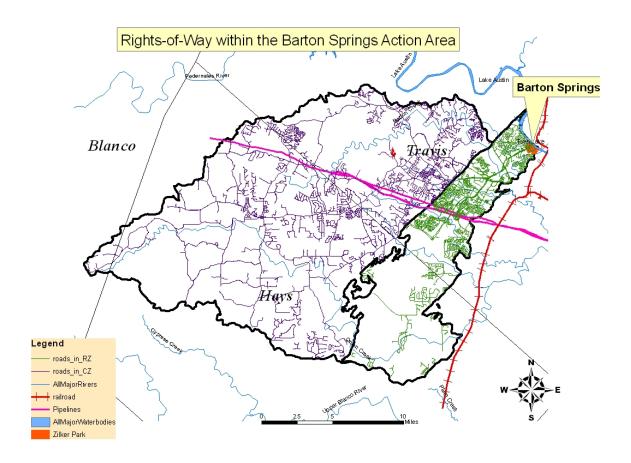


Figure 3.7 Rights-of-Way within the Barton Springs Salamander Action Area

In order to characterize the full extent of this landcover type (roads, utilities, rail, and pipelines combined) a further analysis was conducted with ArcGIS. The total area of each separate landcover type was calculated using the "Calculate Geometry" function for the entire action area and for the portion within the Recharge Zone and the Contributing Zone. Because the railroad coverage indicates that there are no rail lines within the action area, this landcover class was eliminated from any further analysis. In addition, the pipeline coverage had only length associated with the shapefile (as did the railroad shapefile) and a width of 100 feet was arbitrarily assigned to the coverage in order to estimate an area associated with this landcover. A summary of the individual and aggregated landcover class area in hectares is presented in Table 3.4. Table 3.4 also

51

shows the percentage of the total area within the action area and the Recharge and Contributing Zones.

All Classes	91600	100	23300	100	68300	100
Roads	3608	3.9	1558	6.7	2050	3
Rail	0	0	0	0	0	0
Pipelines	29.5	0.03	4.9	0.0002	24.6	0.0003
Utility (other)	84.4	0.0009	34.7	0.1	49.7	0.0007
ROW combined	3721.9	4.1	1597.6	6.9	2124.3	3.1

It is apparent from the analysis above and a visual inspection of the map that many of the roads within the action area are residential in nature and thus unlikely to have pesticides applied to them. In order to provide additional context to the estimates above, information was provided by Scott Lambert of the Travis County Department of Transportation and Natural Resources (TDNR) on areas where herbicides are typically applied to roads. This information is presented in the form of a shapefile indicating those roads within the county that are typically treated. This map is shown in Figure 3.8.

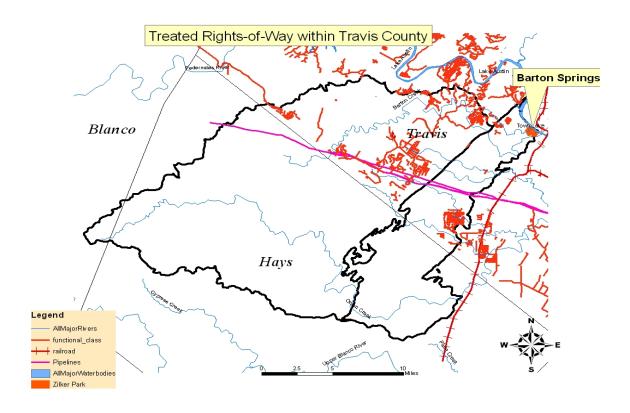


Figure 3.8 Treated Rights-of-Way within Travis County Relative to Barton Springs Action Area

This information was used to compare total area within the action area in Travis County and the subsets of this area within the Recharge Zone and the Contributing Zone in Travis County. This was completed by first intersecting the shapefile from TDNR with the action area in order to identify those portions of the road network typically treated within each zone. The "Calculate Geometry" function was used to estimate total area of these roads within the portion of Travis County within the action area and further refined to estimate the area treated within the Recharge and Contributing Zone portions of Travis County.

The previous analysis was further refined to provide estimates of total road network for the entire action area including portions of Hays and Williamson Counties lacking in similar estimates of area treated. To do this, the estimates of total roads treated within the portion of action area in the Recharge and Contributing Zones were compared to the total extent of roads (both treated and untreated) within these areas. This allows for an estimate of the percentage of roads treated within each zone in the Travis county portion of the action area. These percentage estimates were then extrapolated to the full extent of each zone in the action area including all parts of Travis, Hays, and Williamson Counties. There is some uncertainty associated with this extrapolation given that road networks may differ from Travis to Hays and Williamson Counties; however, the analysis provides an estimation of the percentage of area treated that utilizes best available data. For the Recharge Zone in Travis County, the estimate of total roads treated is 1.5%, while the estimate for the Contributing Zone is 15.4%. Using the information in Table 3.5, the total estimate of treated roadway with the Recharge Zone of the action area is 23.4 ha (1558 ha x 1.5%), and the Contributing Zone is 315.7 ha (2050 ha x 15.4%). Using the modified area estimate derived from the Travis County information on roads treated, yields a total rights-of-way area of 23.4 ha for the Recharge Zone and 315.7 ha for the Contributing Zone.

All Classes	23300 ha	68300 ha
Roads	23.4 ha	315.7 ha
Rail	0 ha	0 ha
Pipelines	4.9 ha	24.6 ha
Utility (other)	34.7 ha	49.7 ha
ROW combined	63.0 ha	390.0 ha

Finally, it is assumed that the total area of ROW within the action area would not be treated at the same time. First, it is expected that the entire width of the ROW would not be treated and that the treatments would typically be focused on a percentage of the width of the zones. Second, it is expected that the entire length of the ROW would not be treated simultaneously and that only selected portions could be treated at a single time (defined by a day of activity). For the purposes of this assessment, it is assumed, based on professional judgment, that no more than 10% of any ROW use within the action area will be treated at any one time. In order to account for this, assumptions of percentage of area treated (defined as the assumed area based on length time width of segments of the

ROW) were applied to the modeling analysis to evaluate the impact of this assumption on exposure. The values used in this assessment were 50% treated, 25% treated, 10% treated, 5% treated, and 1% treated. The results of this analysis indicate that the assumption of 10% is reasonably protective and that higher assumption does not change exposures (other than the peak) by more than a factor of two or three. The results of this analysis are presented below in Table 3.6.

Table 3.6 Summary of PRZM Output EECs for Varying Percentages of Treated Non-cropland

90th Percentile of 30 Years of Output

Use Site	Application Rate (lbs/acre)	Number of Applications (interval)	First Application Date	Peak (one-day) EEC (μg/L)	14-day EEC (μg/L)	21-day EEC (µg/L)	30-day EEC (µg/L)	60-day EEC (μg/L)	90-day EEC (μg/L)	Annual Average (µg/L)
Non-										
Cropland @ 10% Treated	5	1	April 1	1.4	0.2	0.2	0.2	0.1	0.1	0.1
Non-										
Cropland @ 50% Treated	5	1	April 1	6.6	0.7	0.5	0.4	0.2	0.2	0.1
Non-										
Cropland @ 25% Treated	5	1	April 1	3.3	0.4	0.3	0.2	0.2	0.1	0.1
Non-										
Cropland @	5	1	April 1	0.7	0.2	0.1	0.1	0.1	0.1	0.1
5% Treated										
Non-										
Cropland @ 1% Treated	5	1	April 1	0.2	0.1	0.1	0.1	0.1	0.1	0.1

3.2.7.4 Residential

This scenario is intended to be used as a surrogate for all urban/suburban home and residential uses in the Barton Springs Segment (BSS) of the Edwards Aquifer. The scenario was developed using a ¼ acre lot as a representation of a suburban residential development. For herbicide treatments to lawns, it is assumed that only a portion of the ¼ acre lot is treated. Using U.S. Census information, it is estimated that 50% of the ¼ acre lot is treated. In addition, a landcover analysis was conducted to estimate the percentage of a given residential area that is impervious surface not represented by the ¼ acre lot (e.g. parking lots and roads). The percentage of impervious surface not represented by the ¼ acre lot is estimated to be 30%. More details on the development and justification for these estimates may be found in the ecological risk assessment conducted previously for atrazine relative to the Barton Springs Salamander (U.S. EPA 2006b). Exposure estimates generated using this scenario were adjusted by a factor of 0.35 to represent the percentage of treated surface in any residential area (i.e. 50% of the 70% represented by the ¼ acre lot). Figure 3.9 presents the distribution of residential landcover mapped for the Barton Springs Action Area.

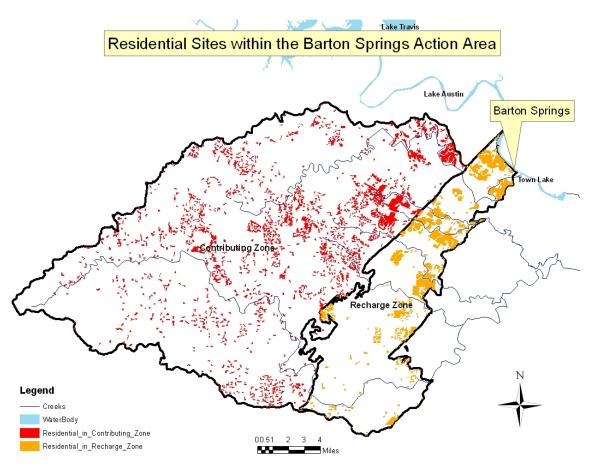


Figure 3.9 Residential Land Cover within the Barton Springs Action Area

3.2.8 Results

Table 3.7 presents the summary of all relevant time-weighted concentrations for each scenario modeled at the 90th % of exposure for the edge of field exposure. The EECs are used to derive risk quotients, which are presented as part of the Risk Characterization in Section 5. In general, exposures are driven by the residential turf modeling. When compared with the available monitoring data from the action area, these modeled concentrations are one to two orders of magnitude higher than those seen in monitoring, suggesting that the EECs in Table 3.7 may over-estimate exposure.

As previously discussed in Section 3.1, proposed mitigation measures specified in the 2006 simazine RED will result in cancellation of all aerial and non-residential granular uses in 2010. Therefore, the aerial non-cropland and granular non-residential turf uses will be completely eliminated from registered uses in 2010. However, based on the summary of modeled EECs shown in Table 3.7, it is apparent that the aggregate simazine exposure in Barton Springs is driven by the residential use. In order to provide characterization, an alternate exposure scenario was evaluated where only the residential, turf and orchard uses of simazine are present in the action area. The results of this analysis, which is presented in Table 3.8, indicate that overall exposures are expected to be similar, regardless of the cancelled non-cropland and non-residential granular simazine uses.

Table 3.7 Summary of PRZM Output EECs for all Modeled Scenarios 90th Percentile of 30 Years of Output Peak Number of First 14-day 90-day Annual 21-day 30-day 60-day Application (one-day) EEC EEC Application EEC EEC EEC **Use Site Applications** Average Rate (lbs/acre) EEC (interval) Date $(\mu g/L)$ $(\mu g \! / \! L)$ $(\mu g \! / \! L)$ $(\mu g/L)$ $(\mu g/L)$ $(\mu g/L)$ (µg/L) Orchard April 1 0.1 0.1 0.1 0.1 0.1 0.1 0.1 4 Turf 2 April 1 0.1 0.1 0.1 0.1 0.1 0.1 0.1 1 Non-0.2 0.2 0.2 0.1 0.1 5 1 April 1 1.4 0.1 Cropland Residential 0.3 2 April 1 7.5 1.0 0.7 0.6 0.4 0.1 1 Turf Aggregated 7.6 1.0 0.8 0.6 0.4 0.3 0.1 Exposure

Table 3.8 Summary of PRZM Output EECs for all Modeled Scenarios Using Post-RED Application Information¹

90th Percentile of 30 Years of Output

Use Site	Application Rate (lbs/acre)	Number of Applications (interval)	First Application Date	Peak (one-day) EEC (μg/L)	14-day EEC (μg/L)	21-day EEC (µg/L)	30-day EEC (µg/L)	60-day EEC (μg/L)	90-day EEC (µg/L)	Annual Average (μg/L)
Orchard	4	1	April 1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Residential Turf	1	2	April 1	7.5	1.0	0.7	0.6	0.4	0.3	0.1
Aggregated Exposure				7.5	1.0	0.7	0.6	0.4	0.3	0.1

¹ All non-cropland aerial uses of simazine will be cancelled by 2010.

4. Effects Assessment

This assessment evaluates the potential for simazine to directly or indirectly affect the Barton Spring salamander. As previously discussed in Section 2.7, assessment endpoints for the Barton Spring salamander include direct toxic effects on the survival, reproduction, and growth of the salamander itself, as well as indirect effects, such as reduction of the prey base and/or modification of its habitat. Direct effects to the Barton Springs salamander are based on toxicity information for freshwater vertebrates, including fish, which are generally used as a surrogate for amphibians. Given that the salamander's prey items and habitat requirements are dependent on the availability of freshwater aquatic invertebrates and aquatic plants, toxicity information for various freshwater aquatic invertebrates and plants is also discussed. Acute (short-term) and chronic (long-term) toxicity information is characterized based on registrant-submitted studies and a comprehensive review of the open literature on simazine. 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 simazine. A summary of the available freshwater ecotoxicity information, use of the probit dose response relationship, and the incident information for simazine are provided in Sections 4.1 through 4.3, respectively.

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, freshwater invertebrates, and aquatic plants.

Toxicity endpoints are established based on data generated from guideline studies submitted by the registrant, and from open literature studies that meet the criteria for inclusion into the ECOTOX database maintained by EPA/Office of Research and Development (ORD) (U.S. EPA, 2004). Open literature data presented in this assessment were obtained from 2006 simazine RED as well as ECOTOX information obtained on September 30, 2006. In order to be included in the ECOTOX database, papers must meet the following minimum criteria:

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

Data that pass the ECOTOX screen are evaluated along with the registrant-submitted data, and may be incorporated qualitatively or quantitatively into this endangered species assessment. In general, effects data in the open literature that are more conservative than the registrant-submitted data are considered. The degree to which open literature data are quantitatively or qualitatively characterized is dependent on whether the information is

relevant to the assessment endpoints (*i.e.*, maintenance of Barton Springs salamander 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.

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 and/or not appropriate for use in this assessment) are included in Appendix F. Appendix F 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.

With respect to simazine degradates, deisopropylatrazine (DIA) and diaminochloroatrazine (DACT), it is assumed that each of the degradates are less toxic than the parent compound. As shown in Table 4.1, comparison of available toxicity information for DIA and DACT indicates lesser aquatic toxicity than the parent for freshwater fish, invertebrates, and aquatic plants.

Simazine	6,400	1000	36
DACT	>100,000	>100,000	No data
DIA	17,000	126,000	2,500

Therefore, given the lesser toxicity of the degradates, as compared to the parent, concentrations of the simazine degradates are not assessed, and the focus of this assessment is limited to parent simazine. The available information also indicates that aquatic organisms are more sensitive to the technical grade (TGAI) than the formulated products of simazine; therefore, the focus of this assessment is on the TGAI. A detailed summary of the available ecotoxicity information for all simazine degradates and formulated products is presented in Appendix A.

The results of available toxicity data for mixtures of simazine with other pesticides are presented in Section A.6 of Appendix A. Based on the available information, other triazine herbicides, such as atrazine, may combine with simazine to produce additive toxic effects on aquatic plants. The variety of chemical interactions presented in the available data set suggest that the toxic effect of simazine, in combination with other pesticides used in the environment, can be a function of many factors including but not necessarily limited to: (1) the exposed species, (2) the co-contaminants in the mixture, (3) the ratio of simazine and co-contaminant concentrations, (4) differences in the pattern and duration of exposure among contaminants, and (5) the differential effects of other physical/chemical characteristics of the receiving waters (e.g. organic matter present in sediment and suspended water). Quantitatively predicting the combined effects of all

these variables on mixture toxicity to any given taxa with confidence is beyond the capabilities of the available data. However, a qualitative discussion of implications of the available pesticide mixture effects data involving simazine on the confidence of risk assessment conclusions for the Barton Springs salamander is addressed as part of the uncertainty analysis for this effects determination.

4.1 Toxicity of Simazine to Aquatic Organisms

Table 4.2 summarizes the most sensitive aquatic toxicity endpoints for the Barton Springs salamander, 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 Barton Springs salamander is presented below. Additional information is provided in Appendix A. Appendix A also includes ecotoxicity data for taxonomic groups that are not relevant to this assessment (i.e., birds, mammals, terrestrial invertebrates and terrestrial plants) because the Agency is completing endangered species risk assessments for other species concurrently with this assessment.

		calculate an EC ₀₅

Data for the TGAI are not available. Concentrations are adjusted for % a.i.

It should be noted that a considerable number of freshwater acute toxicity data and field studies are available for simazine. Reported acute toxicity values generally exceed the water solubility limit of simazine (approximately 3.5 mg/L at 20° C). While simazine concentrations in water would appear to be stable to hydrolysis and photolysis for the duration of the acute static studies, the actual exposure levels are uncertain because mean-measured concentrations are not available, and precipitation is frequently reported in the acute studies. Test concentrations are rarely measured to verify exposure levels; therefore, a high degree of uncertainty exists for the freshwater toxicity data for simazine. As such, studies with LC₅₀ values > 100 mg/L are not presented, based on the level of uncertainty associated with these values. It appears that simazine is acutely toxic to some freshwater fish and aquatic invertebrates in the range of 1 to 10 mg/L.

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

< 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

Ecotoxicity data for freshwater fish are generally used as surrogates for aquatic-phase amphibians when amphibian toxicity data are not available (U.S. EPA, 2004). A comprehensive search of the open literature provided no toxicity information on lethal or sublethal effects of simazine to amphibians. However, atrazine, a triazine herbicide in the same chemical class as simazine, has been associated with endocrine-related effects (i.e., gonadal abnormalities and laryngeal alterations) in frogs. The Agency review of the current database of published studies and registrant submitted studies on atrazine lead to the conclusion that there was sufficient evidence to formulate a hypothesis that atrazine exposure may impact gonadal development in amphibians, but there was insufficient data to confirm or refute the hypothesis (transmission of meeting minutes of the Scientific Advisory Panel (SAP) held June 17-20, 2003;

(http://www.epa.gov/oscpmont/sap/2003/June/junemeetingreport.pdf). Because atrazine and simazine share a similar mechanism of herbicidal action and similar degradates, including DIA and DACT, the current hypothesis regarding potential sublethal effects of atrazine to amphibians may be applicable to simazine depending on the outcome of future studies on atrazine. The results of these studies, as well as other recent open literature data, which focus on the potential effects of atrazine on amphibian gonadal development, are being reviewed. This information will be presented and discussed as part of a second SAP to be held in October 2007.

Given that no simazine toxicity data are available for aquatic-phase amphibians, freshwater fish data were used as a surrogate to estimate direct acute and chronic risks to the Barton Springs salamander. A summary of acute and chronic freshwater fish data, including data from the open literature, is provided below in Sections 4.1.1.1 through 4.1.1.3.

4.1.1.1 Freshwater Fish: Acute Exposure (Mortality) Studies

As shown in Table A-10, submitted acute toxicity values for technical grade simazine exceed its expected water solubility (\sim 3.5 mg/L), with values ranging from 6.4 to >32 mg ai/L. The solubility of simazine is dependant on the water temperature, with a trend toward decreasing solubility at lower temperatures. The following mathematical function describes the relationship between water solubility and temperature: Log (mg/L) = $0.021(T, K) - 5.5358 (R^2 = 0.9862, n = 5)$, where T = temperature and K = kelvin. Further examination of the test temperatures for the acute freshwater studies reveals that all submitted tests were conducted at temperatures < 18°C. Based on the mathematical relationship between solubility and temperature, the expected solubility of simazine in water at a temperature of 18°C would be approximately 3.8 mg/L. With respect to technical grade simazine, the reported acute LC₅₀ values for fathead minnow (MRID 000333-09) and bluegill sunfish (MRID 000254-38) are 6,400 and 16,000 μg/L, respectively. While both of these LC₅₀ values exceed the predicted limit of simazine's solubility in water (3,800 µg/L), a co-solvent was used to increase the limit of simazine's water solubility, and no observation of precipitate were noted in the test chambers. Therefore, the fathead minnow LC₅₀ value of 6,400 µg/L was selected as the surrogate acute freshwater fish toxicity endpoint and used to assess direct acute effects of simazine to the Barton Springs salamander. This test was categorized as supplemental because no raw data or test concentrations were provided in the study. A no effect level of 2,500 µg/L was established in the 96-hour fathead minnow study. This no effect level is consistent with the results of a 28-day subacute rainbow trout study (MRID 000436-68). Following 28-days of exposure, no mortality or other toxic symptoms were observed at the 2,500 µg/L treatment level. The subacute study was classified as supplemental because the fish were too large (25-40g) and only one treatment level (2,500 µg/L) was tested. In the acute bluegill sunfish study, which is classified as core, no mortality was observed in treatment groups $\leq 5,600 \,\mu\text{g/L}$, and 40% mortality was observed in the 10,000 µg/L treatment group.

There is additional uncertainty in all available acute freshwater studies on the TGAI regarding dissolved levels of simazine in water because mean-measured test concentrations were not analyzed. Reported nominal concentration results reflect the concentration after the application and not necessarily the concentration of simazine in water during or at the end of the 96-hour test. A number of the acute studies on both the TGAI and formulated product are classified as invalid because precipitation of the test substance in the test chambers was reported and LC₅₀ values exceed the water solubility of simazine by a large margin.

Acute effects data for freshwater fish are available for a number of simazine's formulated products including Aquazine (80% WP) and a 50% formulation. All ai-adjusted LC₅₀ values for Aquazine (>72,600 μ g/L) and the 50% formulation (13,500 to 55,000 μ g/L) exceed the lowest LC₅₀ value for the TGAI (6,400 μ g/L). The available data suggests that Aquazine and the 50% formulation are less toxic to freshwater fish than the TGAI.

Based on the available data, simazine is categorized as moderately toxic to freshwater fish on an acute basis. No additional data on the acute toxicity of simazine or its degradates to freshwater fish were located in the open literature.

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

Chronic freshwater fish toxicity studies were used to assess potential direct effects via growth and reproduction to the Barton Spring salamander. No freshwater fish early lifestage test using the TGAI was submitted for simazine. Two fish life-cycle tests with fathead minnow were submitted for Aquazine, an 80% formulation that is typically applied directly to the water (MRID 000436-76). One test was conducted with steady concentrations via continuous flow. In the second test, the chemical was applied at the beginning of the test and allowed to decrease at normal degradation rates. Both tests were conducted at the same initial test concentrations. The static test where test concentrations decrease over time is intended to be representative of typical use-pattern exposures of Aquazine. The lowest endpoint values in the continuous and usage-pattern exposures were increase in percent hatched fry (NOAEC = $130 \mu g/L$ ai) and increased fry growth (length) (NOAEC = $250 \mu g/L$ ai), respectively. However, neither of these endpoints are considered to be toxicologically relevant for the risk assessment. Therefore, a NOAEC value of 960 µg/L ai was selected, based on 12% reduction in growth (length) to 30-day old fry at a continuous exposure treatment level of 2,000 µg/L ai. The corresponding LOAEC value, based on reduction in fry growth, is 2,000 µg/L ai. Freshwater fish life-cycle studies for the 80% formulation are summarized in Table A-12 of Appendix A.

4.1.1.3 Freshwater Fish: Sublethal Effects and Additional Open Literature Information

In addition to submitted studies, data were located in the open literature that report sublethal effect levels to freshwater fish that are less than the selected measures of effect summarized in Table 4.2. Although these studies report potentially sensitive endpoints,

effects on survival, growth, or reproduction were not observed in the available full life-cycle studies at concentrations that induced the reported sublethal effects described below and in Appendix A.

No additional information is available that indicates greater acute freshwater fish sensitivity to simazine than the submitted data. In addition, no laboratory freshwater fish early life-stage or life-cycle tests using simazine and/or its formulated products were located in the open literature. However, one laboratory study on sublethal effects of simazine to male Atlantic salmon ($Salmo\ salar\ L$.) is available. In a study conducted by Moore and Lower (2001; ECOTOX# 67727), simazine inhibited *in vitro* olfactory function in male Atlantic salmon parr. The results of this study are summarized in Table A-13 of Appendix A. Following a 5 day exposure period, the reproductive priming effect of the female pheromone prostaglandin $F_{2\alpha}$ on the levels of expressible milt in males was reduced after exposure to simazine at concentrations as low as 0.1 μ g/L. Although the hypothesis was not tested, the study authors suggest that exposure of smolts to simazine during the freshwater stage may potentially affect olfactory imprinting to the natal river and subsequent homing of adults. Although this study produced a NOAEC that is lower than the fish full life-cycle test of 960 ppb, this study was not considered appropriate for RO calculation for the following reasons:

- A negative control was not used; therefore, potential solvent effects cannot be evaluated:
- The study did not determine whether the decreased response of olfactory epithelium to specific chemical stimuli would likely impair similar responses in intact fish.
- A quantitative relationship between the magnitude of reduced olfactory response of male epithelial tissue to the female priming hormone observed in the laboratory and reduction in salmon reproduction (i.e., the ability of male salmon to detect, respond to, and mate with ovulating females) in the wild is not established.

Although these studies raise questions about the potential effects of simazine on endocrine-mediated functions in anadromous fish, it is not possible to quantitatively link these sublethal effects to the selected assessment endpoints for the Barton Springs salamander (i.e., survival, growth, and reproduction of individual). Therefore, potential sublethal effects on fish are evaluated qualitatively in Section 5.2 and not used as part of the quantitative risk characterization. Further detail on sublethal effects to fish is provided in Sections A.4.3 and Table A-13 of Appendix A.

4.1.2 Toxicity to Freshwater Invertebrates

Freshwater aquatic invertebrate toxicity data were used to assess potential indirect effects of simazine to the Barton Springs salamander. Direct effects to freshwater invertebrates resulting from exposure to simazine may indirectly affect the Barton Springs salamander reduction in available food items.

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

4.1.2.1 Freshwater Invertebrates: Acute Exposure Studies

Acute toxicity data for simazine are available for the preferred test species, *Daphnia magna*, as well as seven other freshwater invertebrates including the seed shrimp (*Cypridopsis vidua*), scud (*Gammarus lacustris* and *G. fasciatus*), stonefly (*Pteronarcys californica*), sowbug (*Asellus brevicaudus*), glass shrimp (*Palaemonetes kadiakensis*), and crayfish (*Orconectes nais*). Results of acute toxicity tests with freshwater invertebrates are tabulated in Table A-14 of Appendix A.

In a comparative analysis of herbicides on six species of freshwater invertebrates, 48-hr exposure to simazine at concentrations of 1,000 and 3,700 µg/L resulted in 50 percent mortality in daphnia and seed shrimp, respectively (MRID 450882-21). In the same analysis, simazine did not appear to have any effect on the scud (G. fasciatus), sowbug, glass shrimp, or crayfish, with 48-hr TL₅₀ values exceeding 100,000 µg/L. However, as previously mentioned, toxicity values > 100,000 µg/L exceed the water solubility of simazine by a wide margin; therefore, the validity of the data is uncertain. TL₅₀ values reported in the study are median tolerance limits, representative of the concentration in water in which 50 percent of the animals exhibit a specific response (i.e., mortality, immobilization) at a given time. It should be noted that no test concentrations or raw data were provided as part of this study; therefore, it was classified as supplemental. In addition, the slope of the dose-response relationship for daphnia could not be determined due to a lack of raw data and test concentrations.

Two additional supplemental 96-hr acute toxicity studies on freshwater invertebrates are available for the technical grade of simazine. In a chemical database of acute toxicity to freshwater animals maintained by the Columbia National Fisheries Research Laboratory of the U.S. Fish and Wildlife Service, 96-hr exposure of the stonefly ($P.\ californica$) to simazine resulted in an EC50 of 1,900 µg/L (MRID 400980-01). A 96-hr EC50 value of 13,000 µg/L was reported for the scud ($G.\ lacustris$) (MRID 050092-42) in a study classified as supplemental because no mortality data were provided and test concentrations were not specified.

Based on the available data, simazine is categorized as highly to slightly toxic to freshwater invertebrates on an acute basis.

4.1.2.2 Freshwater Invertebrates: Chronic Exposure Studies

No freshwater invertebrate life-cycle test using the TGAI was submitted for simazine. A freshwater aquatic invertebrate life-cycle test using the formulated product Aquazine (80% formulation) was submitted for simazine (MRID 000436-76) using the preferred species *D. magna*. The results of this test are summarized in Table A-16 of Appendix A. No treatment-related adverse effects to parental mortality and production of offspring occurred during the 21-day study at the highest test concentration of 2,000 µg/L. The

only treatment-related effect was a significant stimulation of offspring produced at the 80 μ g/L test concentration. Therefore, the NOAEC value is 2,000 μ g/L.

4.1.2.3 Freshwater Invertebrates: Open Literature Data

Only one chronic toxicity study on freshwater invertebrates is available from the open literature. It appears that *D. pulex* fed a diet of green alga are less sensitive to the effects of simazine, as compared with those that are fed mixed bacterial cultures. Similar to the results reported in the registrant submitted studies, simazine concentrations at the highest treatment level (5,000 µg/L) were shown to enhance reproduction and growth in *D. pulex* that were fed green alga following 14 days of exposure. Conversely, reproduction was significantly reduced at simazine concentrations of 5,000 and 1,000 µg/L when mixed bacterial cultures were used as the food source. However, no significant differences in the number of offspring per adult were observed at treatment levels of 100, 200, or 2,000 µg/L; therefore, the results are erratic and not dose-dependant. Given the variability in reproductive responses in *D. pulex* fed mixed bacterial cultures and issues of comparability between chronic freshwater invertebrate guidelines (where invertebrates are not fed mixed bacterial cultures), the data from this study are addressed qualitatively. The results of this study are described in further detail in Section A.4.7 and Table A.17 of Appendix A.

4.1.3 Toxicity to Aquatic Plants

Aquatic plant toxicity studies were used to evaluate whether simazine may affect primary production. In Barton Springs, primary productivity is essential for indirectly supporting the growth and abundance of the Barton Springs salamander. In addition to providing cover, moss and other aquatic plants harbor a variety of aquatic invertebrates that salamanders eat.

Two types of studies were used to evaluate the potential of simazine to affect aquatic plants. Laboratory and field studies were used to determine whether simazine may cause direct effects to aquatic plants. A summary of the laboratory data and freshwater field studies for aquatic plants is provided in Sections 4.1.3.1 and 4.1.4.

4.1.3.1 Aquatic Plants: Laboratory Data

A summary of acute toxicity of simazine to aquatic plants is provided in Table A-21 of Appendix A. Tier II toxicity data for technical grade simazine is available for vascular duckweed (*Lemna gibba*) and the following non-vascular plants: blue-green algae (*Anabaena flos-aquae*), marine diatom (*Skeletonema costatum* and *Phaeodactylum tricornutum*), freshwater alga (*Selenastrum capricornutum*), freshwater diatom (*Navicula pelliculosa*), marine algae (*Isochrysis galbana*), and marine green algae (*Chlorococcum* sp. and *Dunaliella tertiolecta*).

One Tier II study of the freshwater aquatic vascular plant, duckweed, was completed using the TGAI of simazine (MRID 425037-04). Frond number was the most sensitive

endpoint with an EC₅₀ value of 140 μ g/L. NOAEC and LOAEC values, based on reduction in frond number and growth rate inhibition were 54 and 110 μ g/L, respectively. Growth was reduced by 9.1% in plants in the 110 μ g/L treatment group. By days 6-9 and onward, there was an increase in colony breakup, smallness of frond, and root destruction in test solutions of \geq 230 μ g/L.

The Tier II results indicate that freshwater blue-green algae (Anabaena) is the most sensitive non-vascular plant to simazine (MRID 426624-01). The EC₅₀ for Anabaena is 36 µg/L, as compared to EC₅₀ values ranging from 90 to 4,000 µg/L for other non-vascular plants. The Tier II aquatic plant study with the freshwater alga, Anabaena, was scientifically valid, but could not be classified as acceptable because a NOAEC value was not determined. In an Agency 1993 memo, dated October 18, 1993, EPA agreed that existing growth data be used to derive an EC₁₀ value for use as the NOAEC. However, current Agency policy specifies that the EC₀₅ be used to derive the NOAEC in order to protect listed species that have obligate relationships with non-vascular plants. The resulting NOAEC value based on the EC₀₅ is 5.4 µg/L. Reduction in growth rates of 36.8, 80.1, 97.6, and 107% were observed by day 5 at respective test concentrations of 78, 170, 320, and 660 µg/L. In addition, a 28% reduction in cell density was observed at the lowest test concentration of 20 µg/L.

4.1.4 Freshwater Field Studies

A number of field studies are available in the open literature that evaluate adverse effects to freshwater organisms resulting from single and multiple applications of simazine to freshwater ponds to remove noxious growths of aquatic macrophytes. Generally, direct application of simazine to ponds results in a die-off of macrophytes, which consequently results in a decrease of dissolved oxygen (DO). In many of the studies, adverse effects to freshwater fish in field studies following simazine application are attributed to indirect effects including a combination of low DO and reduced food resources, rather than direct toxicity of simazine. Available data from aquatic field studies are inadequate to determine whether simazine applications to aquatic habitats at levels of approximately 1,000 µg/L (1 ppm) result in adverse effects to non-target aquatic organisms either by direct toxicity or indirect effects such as low DO, lost food/habitat resources, and/or decreased ecosystem productivity in the absence of macrophytes. The available field data indicate that benthic macroinvertebrates are generally not adversely impacted by simazine concentrations of 1,000 µg/L, although one study reported a reduction in zooplankton biomass in the post-treatment period. In most of the studies, the fish are older life stages such as fingerlings and/or adults, which are not normally as sensitive to pesticides as larval and fry stages. In addition to indirect effects associated with low DO, the results of one field study suggest a possible direct effect of simazine on the feeding response of channel catfish, following direct application of 1,300 µg/L to earthen channel catfish ponds infested with stonewort. The reviewed field studies are qualitatively evaluated in this risk assessment because observed adverse effects associated with simazine exposure are likely the result of a complex interaction of several parameters rather than simazine concentration alone. Further discussion of the open literature field

studies for freshwater fish and invertebrates is provided in Section A.4.8 and summarized in Table A.18 of Appendix A.

The open literature contains a large amount of information on the toxicity of simazine to aquatic plants; however, the majority of data report toxicity values that are higher (i.e., not as sensitive) than the endpoints reported in the submitted studies. A number of open literature papers, which characterize unique endpoints to aquatic plants, present data with endpoint values that are more sensitive than the submitted endpoints, or discuss aquatic plant succession and recovery following simazine application are discussed below. Tables A-23 and A-18 of Appendix A provide a summary of the open literature laboratory and *in situ* studies, respectively, on the effects of simazine to aquatic plants. Based on the results of the *in situ* and laboratory studies, it appears that simazine results in a reduction of chlorophyll a in periphyton and phytoplankton at simazine levels between 500 and 1,000 µg/L. Other studies show increased chlorophyll a production at simazine concentrations of <0.05 ug/L. In addition, despite the apparent sensitivity of the blue-green algae Anabaena flos-aquae to simazine, the results of one open literature study suggest possible resistance and shifts in the aquatic periphytic plant community to blue-green alga at the higher simazine treatment levels of 5,000 µg/L. Simazine resistance has also been reported in seeds and tubers of *Potamogeton foliosus*. There is evidence to suggest that recovery occurs in algae upon removal of simazine from the site of action, with the recovery inversely proportional to the prior exposure level. In one study, recovery of macrophytes was noted within two to three months following application of simazine granules at 25 lb doses (% ai was not reported). Further detail on the open literature data for aquatic plants is discussed in Section A.5.3.

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

The Agency uses the probit dose response relationship as a tool for providing additional information on the potential for acute direct effects to individual listed species and aquatic animals that may indirectly affect the listed species of concern (U.S. EPA, 2004). As part of the risk characterization, an interpretation of acute RQ for listed species is discussed. This interpretation is presented in terms of the chance of an individual event (i.e., mortality or immobilization) should exposure at the EEC actually occur for a species with sensitivity to simazine 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. However, based on a review of the acute toxicity for simazine, no dose response information is available to estimate a slope for this analysis; therefore, a default slope assumption of 4.5 (with lower and upper bounds of 2 to 9) (Urban and Cook, 1986) is used.

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 lower and upper confidence bounds of that estimate) as the slope parameter for the spreadsheet. In addition, the acute RQ is entered as the desired threshold.

4.3 Incident Database Review

A review of the EIIS database for ecological aquatic incidents involving simazine was completed on May 22, 2006. Nine freshwater aquatic incidents involving fish kills were reported for simazine between the years of 1976 and 1995. Six incidents have a certainty index of "highly probable" or "probable," and the other three have certainty indices of "possible" and "unlikely." Six incidents resulted from treatment of a lake, pond, or lagoon; two incidents were associated with simazine use on corn and from simazine use along railroad tracks; and the treatment site for the other incident was not reported. In a number of the incidents involving direct application of simazine to lakes, ponds, and lagoons, the legality of use was listed as "misuse" or "undetermined." For those incidents where the legality of use is reported as "registered use," the volume of the water bodies is not provided; therefore, it is unclear whether simazine was applied in accordance with its intended use. The six incidents involving direct application of simazine to water are summarized in Appendix D. All occurred prior to 1996, when label language was clarified to restrict direct applications to ornamental ponds and aquaria greater than 1,000gallons. It is important to note that in a number of the incidents involving direct application of simazine to water, low DO, caused by decaying aquatic vegetation, is attributed as an indirect effect related to the fish kills. The certainty index associated with the remaining three incidents (those resulting from use on corn, railroad tracks, and an unspecified treatment site) was reported as "unlikely."

Of the nine reported incidents, three were reported in California, two were reported in Nebraska, two were reported in South Carolina, and one was reported in Michigan and in Tennessee. Fish species listed in these kills include smelt, bullheads, stickleback, striped bass, bluegills, channel catfish, croaker, menhaden, mullet, northern pike, pinfish, yellow perch, sea trout, black bullhead, and fathead minnows.

5. Risk Characterization

Risk characterization is the integration of the exposure and effects characterizations to determine the potential ecological risk from varying simazine use scenarios within the action area and likelihood of direct and indirect effects on the Barton Springs salamander. The risk characterization provides an estimation and a description 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 Barton Springs salamander and/or its habitat (i.e., "no effect," "likely to adversely affect," or "may affect, but not likely to adversely affect").

5.1 Risk Estimation

Risk was estimated by calculating the ratio of the estimated environmental concentration (Table 3.7) and the appropriate toxicity endpoint (Table 4.2). This ratio is the risk quotient (RQ), which is then compared to established acute and chronic levels of concern (LOCs) for each category evaluated (Appendix E). Screening-level RQs are based on the most sensitive endpoints and the following surface water concentration scenarios for simazine:

- orchard use @ 4 lbs ai/A; 1application
- turf granular use @ 2 lbs ai/A; 2 x 1 lb applications with 30 days between applications
- non-cropland aerial use @ 5 lbs ai/A; 1 application
- residential turf use @ 2 lbs ai/A; 2 x 1 lb applications with 30 days between applications
- aggregate EEC based on combined orchard, turf, non-cropland, and residential simazine uses within the action area

In cases where the screening-level RQ exceeds one or more LOCs (i.e., "may affect"), additional factors, including Barton Springs salamander life history characteristics and refinement of the screening-level EECs using available monitoring data, are considered and used to characterize the potential for simazine to adversely affect the Barton Springs salamander. Risk estimations of direct and indirect effects of simazine to the Barton Springs salamander are provided in Sections 5.1.1 and 5.1.2, respectively.

As previously discussed in the effects assessment, the toxicity of the simazine degradates has been shown to be less than the parent compound based on the available toxicity data for freshwater fish, invertebrates, and aquatic plants; therefore, the focus of the risk characterization is parent simazine (i.e., RQ values were not derived for the degradates).

5.1.1 Direct Effects

Direct effects to the Barton Springs salamander associated with acute and chronic exposure to simazine are based on the most sensitive toxicity data available for freshwater fish. Acute and chronic RQs used to estimate potential direct effects to the Barton Springs salamander are provided in Table 5.1.

Direct effects associated with acute and chronic exposure to simazine are not expected to occur for the Barton Springs salamander because RQs are well below LOCs based on all combined uses of simazine within the action area. These RQs are further characterized in Section 5.2.1.

Table 5.1 Summary of Direct Effect RQs for the Barton Springs Salamander						
Direct Acute	Fathead	$LC_{50} = 6,400$	Peak = 7.6	< 0.01	1 in 1.28E+41 (1	No ^c
Toxicity	minnow				in 1.01E+09 to 1	
					in 1.35E+160)	
Direct Chronic		NOAEC =	60-day = 0.4	< 0.01	Not calculated for	No ^c
Toxicity		960			chronic endpoints	

^a Screening-level aggregate EEC for all combined uses of simazine within the action area (from Table 3.7).

5.1.2 Indirect Effects

Pesticides have the potential to exert indirect effects upon listed species by inducing changes in structural or functional characteristics of affected communities. Perturbation of forage or prey availability and alteration of the extent and nature of habitat are examples of indirect effects.

In conducting a screen for indirect effects, direct effects LOCs for each taxonomic group (i.e., freshwater fish, invertebrates, and aquatic plants) are employed to make inferences concerning the potential for indirect effects upon listed species that rely upon non-listed organisms in these taxonomic groups as resources critical to their life cycle (U.S. EPA, 2004). This approach used to evaluate indirect effects to listed species is endorsed by the Services (USFWS/NMFS, 2004b). If no direct effect listed species LOCs are exceeded for non-endangered organisms that are critical to the Barton Springs salmander's life cycle, the concern for indirect effects to the Barton Springs salamander is expected to be minimal.

If LOCs are exceeded for freshwater invertebrates that are prey items of the Barton Springs salamander, there is a potential for simazine to indirectly affect the salamander by reducing available food supply. In such cases, the dose response relationship from the toxicity study used for calculating the RQ of the surrogate prey item is analyzed to estimate the probability of acute effects associated with an exposure equivalent to the EEC. The greater the probability that exposures will produce effects on a taxa, the greater the concern for potential indirect effects for listed species dependant upon that taxa (U.S. EPA, 2004).

As an herbicide, indirect effects to the Barton Springs salamander from potential effects on primary productivity of aquatic plants are a principle concern. If plant RQs fall between the endangered species and non-endangered species LOCs, a no effect determination for listed species that rely on multiple plant species to successfully complete their life cycle (termed plant dependent species) is determined. If plant RQs are

^b A probit slope value for the acute fathead minnow toxicity test is not available; therefore, the effect probability was calculated based on a default slope assumption of 4.5 with upper and lower confidence intervals of 2 and 9 (Urban and Cook, 1986).

^c RQ < acute endangered species LOC of 0.05.

above non-endangered species LOCs, this could be indicative of a potential for adverse effects to those listed species that rely either on a specific plant species (plant species obligate) or multiple plant species (plant-dependent) for some important aspect of their life cycle (U.S. EPA, 2004). Based on the information provided in Appendix C, the Barton Springs salamander does not rely on a specific plant species (i.e., the salamander does not have an obligate relationship with a specific species of aquatic plant).

In summary, the potential for indirect effects to the Barton Springs salamander was evaluated using methods outlined in U.S. EPA (2004) and described below in Sections 5.1.2.1 and 5.1.2.2.

5.1.2.1 Evaluation of Potential Indirect Effects via Reduction in Food Items (Freshwater Fish and Invertebrates)

Potential indirect effects from direct effects on animal food items (i.e., freshwater invertebrates) were evaluated by considering the diet of the Barton Springs salamander. Barton Springs salamanders feed on a wide range of freshwater aquatic invertebrates including ostracods, copepods, chironomids, snails, amphipods, mayfly larvae, leeches, and adult riffle beetles. The most prevalent invertebrates found in stomach and fecal samples from a limited number of adult and juvenile Barton Springs salamanders were ostracods, amphipods, and chironomids (USFWS, 2005). However, data on the relative percentage of each type of aquatic invertebrate in the salamander's diet are not available. The RQs used to characterize potential indirect effects to the Barton Springs salamander from direct acute and chronic effects on freshwater invertebrate food sources are provided in Table 5.2. Acute and chronic RQs are based on the most sensitive toxicity endpoint for *Daphnia* ($TL_{50} = 1,000 \mu g/L$ and $NOAEC = 2,000 \mu g/L$), respectively. Uncertainties associated with use of a NOAEC value that is two times higher than the acute endpoint for the same genus of freshwater invertebrate (*Daphnia*) are addressed in Section 5.2.2.

Indirect effects to the Barton Springs salamander via reduction in the food supply of freshwater invertebrates are unlikely to occur because acute and chronic RQs (based on combined uses of simazine within the action area) for freshwater invertebrates are well below their respective LOCs. These RQs are further characterized in Section 5.2.2.

Reduced Food	Daphnia	$TL_{50} = 1,000$	Peak =	< 0.01	1 in 6.45E+21 (1	No ^c
Supply via Acute			7.6		in 1.22E+05 to 1	
Direct Toxicity to					in 2.31E+83)	
Invertebrates						
Reduced Food	Daphnia	NOAEC =	21- day =	< 0.01	Not calculated for	No ^c
Supply via Chronic	·	2,000	0.8		chronic endpoints	

Direct Toxicity to			
Invertebrates			

^a Screening-level aggregate EEC for all combined uses of simazine within the action area (from Table 3.7).

5.1.2.2 Evaluation of Potential Indirect Effects via Reduction in Habitat and/or Primary Productivity (Freshwater Aquatic Plants)

Potential indirect effects to the Barton Springs salamander based on impacts to habitat and/or primary production were assessed using RQs from freshwater aquatic vascular and non-vascular plant data as a screen. RQs used to estimate potential indirect effects to the Barton Springs salamander from effects on aquatic plant primary productivity are summarized in Table 5.3.

Based on the results shown in Table 5.3, LOCs for direct effects to aquatic non-vascular and vascular plants are not exceeded for all combined uses of simazine within the action area. Therefore, simazine is not likely to indirectly affect the Barton Springs salamander via direct effects on aquatic plants. No known obligate relationship exists between the Barton Springs salamander and any single freshwater non-vascular and/or vascular plant species; therefore, endangered species RQs using the NOAEC/EC₀₅ values for aquatic plants were not derived. These RQs are further characterized in Section 5.2.3.

Reduced Habitat and/or	7.6	0. 21	0.05	No ^d
Primary Productivity				
via Direct Toxicity to				
Aquatic Plants				

^a Screening-level aggregate EEC for all combined uses of simazine within the action area (from Table 3.7).

5.2 Risk Description

The risk description synthesizes an overall conclusion regarding the likelihood of adverse impacts leading to an effects determination (i.e., "no effect," "may affect, but not likely to adversely affect," or "likely to adversely affect") for the Barton Springs salamander.

If the RQs presented in the Risk Estimation (Section 5.1) show no indirect effects and LOCs for the Barton Springs salamander are not exceeded for direct effects (RQs <

^b A probit slope value for the acute fathead minnow toxicity test is not available; therefore, the effect probability was calculated based on a default slope assumption of 4.5 with upper and lower confidence intervals of 2 and 9 (Urban and Cook, 1986).

^c RQ < acute endangered species LOC of 0.05.

 $^{^{}b}$ Based on a 5-day EC50 value of 36 μ g/L for blue green algae (MRID 426624-01).

^c Based on a 14-day EC50 value of 140 µg/L for duckweed (MRID 425037-04).

^dRQ < non-listed plant species LOC of 1.0.

LOC), a "no effect" determination is made, based on simazine's use within the action area. If, however, indirect effects are anticipated and/or exposure exceeds the LOCs for direct effects (RQs > LOC), the Agency concludes a preliminary "may affect" determination for the FIFRA regulatory action regarding simazine. A summary of the results of the risk estimation (i.e., "no effect" vs. "may affect" finding) presented in Sections 5.1.1 and 5.1.2 is provided in Table 5.4 for direct and indirect effects to the Barton Springs salamander.

A description of the risk and effects determination for each of the established assessment endpoints for the Barton Springs salamander is provided in Sections 5.2.1 through 5.2.3.

5.2.1 Direct Effects to the Barton Springs Salamander

Acute and chronic RQs based on aggregate combined uses of simazine in the action area are well below the Agency's acute and chronic risk LOCs. In addition, consideration of the simazine monitoring data from Barton Springs confirms that simazine concentrations (< 1 μ g/L) are well below exposures associated with adverse effects to freshwater fish. As previously discussed, direct effects to the Barton Springs salamander were based on freshwater fish data, which are used as a surrogate for aquatic-phase amphibians.

Because raw data was not provided as part of the acute toxicity study for the fathead minnow, information is unavailable to estimate a slope for the dose response curve. Therefore, the probability of an individual effect to the Barton Springs salamander was calculated based on a default assumption of 4.5 (with lower and upper bounds of 2 and 9) (Urban and Cook, 1986). The corresponding estimated chance of an individual acute mortality to the Barton Springs salamander at an RQ level of 0.001 is 1 in 1.28E+41

(with respective upper and lower bounds of 1 in 1.01E+09 to 1 in 1.35E+160). Given the low probability of an individual mortality occurrence and acute and chronic RQs that are well below LOCs, simazine is not likely to cause direct adverse effects to the Barton Springs salamander.

No toxicity information on lethal and/or sublethal effects of simazine to aquatic-phase amphibians is available, based on a comprehensive search of the open literature. As discussed in Section 4.1.1.3, one open literature study raises questions about sublethal effects of simazine on endocrine-mediated olfactory functions in anadromous fish. Consideration of the sublethal data indicates that effects associated with endocrinemediated olfactory functions may occur in anadromous fish including salmon at simazine concentrations as low as 0.1 µg/L (Moore and Lower, 2001). However, there are a number of limitations in the design of this study, which are addressed in detail in Section A.4.3 of Appendix A, that preclude quantitative use of the data in this risk assessment. For example, Moore and Lower (2001) exposed epithelial tissue (after removal of skin and cartilage) and not intact fish to simazine, and potential solvent effects could not be reconciled (i.e., no negative control was tested). Furthermore, no quantitative relationship is established between reduced olfactory response (measured as electrophysiological response) of epithelial tissue to the priming hormone in the laboratory and reduction in salmon reproduction (i.e., the ability of male salmon to detect, respond to, and mate with ovulating females) in the wild. In summary, it is not possible to quantitatively link the sublethal effects to the selected endpoints for the Barton Springs salamander (i.e., survival, growth, and reproduction of individuals). Also, effects to reproduction, growth, and survival were not observed in the full life cycle studies at levels that produced the reported sublethal effects (Appendix A).

A number of freshwater microcosm, mesocosm, and field studies are available for simazine, although the lowest concentration of simazine tested in these studies was 1,000 μg/L, well above environmentally relevant concentrations. In many of the studies (summarized in Section A.4.8 of Appendix A), adverse effects to freshwater fish in field studies following simazine application are attributed to indirect effects including a combination of low DO and reduced food resources, rather than direct toxicity of simazine. Therefore, the available field study data are inadequate to determine whether simazine applications to aquatic habitats at levels of approximately 1,000 µg/L result in adverse effects to freshwater fish either by direct toxicity or indirect effects such as low DO, lost food/habitat resources, and/or decreased ecosystem productivity in the absence of macrophytes. In addition to indirect effects associated with low DO, the results of a field study by Tucker and Boyd (1978) suggest a possible direct effect of simazine on the feeding response of channel catfish, following direct application of 1,300 µg/L to earthen channel catfish ponds infested with stonewort. However, the application rate of simazine used in this study is approximately three times higher than current labels allow and direct applications to water are restricted to ornamental ponds and aquariums of 1,000 gallons or less.

Nine freshwater aquatic incidents involving fish kills were reported for simazine between the years of 1980 and 1995. As described in Section 4.3, the majority of these incidents

were the result of direct application of simazine to water bodies not in accordance with the current label restrictions for direct applications to ornamental ponds and aquaria less than 1,000 gallons. The remaining three incidents were associated with simazine use on corn (not a use pattern associated with the action area for the salamander) and from simazine use along railroad tracks; however, the certainty index associated with these incidents was reported as "unlikely". A complete list of all the aquatic incidents involving simazine is included in Appendix D.

In summary, the Agency concludes a "no effect" determination for direct effects to the Barton Springs salamander, via mortality, growth, or fecundity, based on all available lines of evidence.

5.2.2 Indirect Effects via Reduction in Food Items (Freshwater Invertebrates)

Potential direct acute and chronic adverse effects to freshwater invertebrates, and resulting indirect effects to the Barton Springs salamander via reduction in food items, are unlikely, based on aggregate combined uses of simazine in the action area. Acute and chronic RQs for aquatic invertebrates are well below the Agency's LOCs with respective values of 0.007 and 0.0004. In addition, available simazine monitoring data from Barton Springs show simazine concentrations of less than 1 µg/L, less than exposures associated with adverse effects to freshwater invertebrates.

Acute RQ values were derived based on toxicity data on technical grade simazine for *Daphnia magna*, which appears to be the most sensitive of all the freshwater invertebrates studied. Information is unavailable to estimate a slope for the doseresponse curve because the raw data was not provided as part of the acute toxicity study for *D. magna*; therefore, the probability of an individual effect to a freshwater invertebrate was calculated based on a default assumption of 4.5 (with lower and upper bounds of 2 and 9). The corresponding estimated chance of an individual acute mortality and/or immobilization to a freshwater invertebrate at an RQ level of 0.007 is 1 in 6.45E+21 (with respective upper and lower bounds of 1 in 1.22E+05 to 1 in 2.31E+83).

Chronic RQs for invertebrates were less than the Agency's LOC, based on the highest 21-day modeled EECs for all simazine uses (0.8 μ g/L) and a *Daphnia* NOAEC value of 2,000 μ g/L for the 80% formulated product of simazine. As previously discussed in Section 4.1.2.2, chronic toxicity data for freshwater invertebrates using the TGAI are not available, although acute data for freshwater fish show that the formulated products of simazine are less toxic than the TGAI. Therefore, use of the formulated product chronic toxicity for freshwater invertebrates may underestimate potential effects, given the available data for freshwater fish. In addition, there is uncertainty associated with the NOAEC value of 2,000 μ g/L because no adverse effects to parental mortality or production of offspring were observed at the highest test concentration, despite an acute TL₅₀ value (1,000 μ g/L) for the same genus of freshwater invertebrate (*Daphnia*) that is two times lower than the chronic NOAEC. In order to characterize this uncertainty, the highest 21-day modeled EEC for simazine is compared to the lower TL₅₀ value; based on this comparison, the 21-day modeled EEC (0.8 μ g/L) is well below the TL₅₀ value of

1,000 µg/L. Chronic effects for freshwater invertebrates would have to be more than 3 orders of magnitude lower than the acute freshwater invertebrate endpoint to result in a level of effect that exceeds the LOC for freshwater invertebrates at the predicted levels of simazine exposure. One chronic open literature study on *Daphnia pulex* confirms that no adverse effects on growth or reproduction occur at simazine exposure concentrations $\leq 2,000 \,\mu\text{g/L}$ (Carter, 1981), well above environmentally relevant concentrations. In addition, the available field data indicate that benthic macroinvertebrates are generally not adversely impacted by simazine concentrations of 1,000 $\mu\text{g/L}$. Therefore, chronic risks to freshwater invertebrates and potential indirect effects to Barton Spring salamanders that consume them as prey are not expected.

The potential for simazine to elicit indirect effects to Barton Springs salamanders via effects of food items is dependent on several factors including: (1) the potential magnitude of effect on freshwater 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 Barton Springs salamander. Based on the low probability of an individual mortality/immobilization occurrence (1.5E-21%) to freshwater invertebrates and resulting RQs (based on the most sensitive freshwater invertebrate food item) that are well below LOCs, simazine is not likely to cause indirect adverse effects to the Barton Springs salamander via a reduction in food items. Therefore, the Agency concludes a "no effect" determination for indirect effects to the Barton Springs salamander via reduction in freshwater invertebrate food items, based on all available lines of evidence.

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

Direct adverse effects to non-vascular and vascular aquatic plants are unlikely because RQs based on aggregate combined uses of simazine in the action area and the most sensitive aquatic plant data are well below the Agency's LOCs. In addition, available simazine monitoring data from Barton Springs show simazine concentrations of less than 1 μ g/L, well below concentrations shown to cause adverse effects in aquatic plants. Therefore, simazine use in the action area is not expected to adversely affect the Barton Springs salamander via reduction in habitat and/or primary productivity.

Based on the results of the *in situ* and laboratory studies from the open literature (summarized in Tables A-23 and A-18 of Appendix A), it appears that simazine results in a reduction of chlorophyll a in periphyton and phytoplankton at simazine levels between 500 and 1,000 µg/L. Other studies show increased chlorophyll a production at low concentrations of simazine (\leq 0.05 µg/L). In addition, despite the apparent sensitivity of the blue-green algae *Anabaena flos-aquae* to simazine, the results of one open literature study suggest possible resistance and shifts in the aquatic periphytic plant community to blue-green alga at the higher simazine treatment levels of 5,000 µg/L. Simazine resistance has also been reported in seeds and tubers of *Potamogeton foliosus*. There is also evidence to suggest that recovery occurs in algae upon removal of simazine from the site of action, with the recovery inversely proportional to the prior exposure level. The

results of the available open literature confirm that simazine-related effects to aquatic plants within Barton Springs are unlikely to occur.

Based on the weight-of-evidence, a "no effect" determination is concluded for indirect effects to the Barton Springs salamander via reduction in freshwater aquatic plants that provide habitat and/or primary productivity.

6. Uncertainties

6.1 Exposure Assessment Uncertainties

Overall, the uncertainties inherent in the exposure assessment tend to result in overestimation of exposures. This is apparent when comparing modeling results with monitoring data. In particular, peak exposures are generally one to two orders of magnitude above the highest detection found in any of the four springs. In general, the monitoring data should be considered as a lower bound on exposure, while modeling represents an upper bound. Factors influencing the over-estimation of exposure include the assumption of no degradation or mixing in the subsurface transport from edge of field to springs. The modeling exercise conservatively assumes that simazine application sites are adjacent to the springs. In addition, the exposures in this assessment are being driven by a single use (residential) and conservatively assume that 50% of the potential use site is treated simultaneously. In reality, there are likely to be processes at work which cannot be accounted for in the modeling that will reduce the predicted exposures, and it is unlikely that residential use of simazine would be applied as a broadcast application across the entire action area.

6.1.1 Modeling Assumptions

Overall, the uncertainties addressed in this assessment cannot be quantitatively characterized. However, given the available data and the tendency to rely on conservative modeling assumptions, it is expected that the modeling results in an overprediction in exposure. In general, the simplifying assumptions used in this assessment appear from the characterization in Section 3.2.7 to be reasonable especially in light of the analysis completed and the available monitoring data. There are also a number of assumptions that tend to result in exposure over-estimation that cannot be quantified, but can be qualitatively described. For instance, modeling for each use site assumes (with the exception of the rights-of-way scenario) that the entire 10-hectare watershed is taken up by the respective use pattern. The assessment assumes that all applications have occurred concurrently on the same day at the exact same application rate. This is unlikely to occur in reality, but is a reasonable conservative assumption in lieu of actual data.

6.1.2 PRZM Modeling Inputs and Predicted Aquatic Concentrations

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 (PRZM) is a process or "simulation" model that calculates what happens to a

pesticide in a farmer's field on a day-to-day basis. It considers factors such as rainfall and plant transpiration of water, as well as how and when the pesticide is applied. It has two major components: hydrology and chemical transport. Water movement is simulated by the use of generalized soil parameters, including field capacity, wilting point, and saturation water content. The chemical transport component simulates 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 adds 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 open environment 90 percent of the time. Mobility input values are chosen to be representative of conditions in the open 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.

Additionally, the rate at which simazine is applied, the percent of a watershed that is cropped, and the percent of crops in that watershed that are actually treated with simazine may be lower than the Agency's default assumptions including use of the maximum allowable application rate, treatment of the entire crop, and the estimated area within a watershed planted with agricultural crops. The geometry of a watershed and limited meteorological data sets also add to the uncertainty of estimated aquatic concentrations.

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

6.2.2 Use of surrogate species effects data

Guideline toxicity tests and open literature data on simazine are not available for salamanders or any other aquatic-phase amphibian; therefore, freshwater fish are used as surrogate species for aquatic-phase amphibians including salamanders. Although no data is available for simazine, the available open literature information on atrazine (a closely related triazine herbicide) toxicity to aquatic-phase amphibians shows that acute and chronic ecotoxicity endpoints for aquatic-phase amphibians are generally about 3 to 4 times less sensitive than freshwater fish. Given that atrazine and simazine share a similar mode of action, it is assumed that same relationship in toxicity between freshwater fish and aquatic-phase amphibians would apply to simazine. Therefore, endpoints based on freshwater fish ecotoxicity data are assumed to be protective of potential direct effects to aquatic-phase salamanders including the Barton Springs salamander, and extrapolation of the risk conclusions from the most sensitive tested species to the Barton Springs salamander is likely to overestimate the potential risks to those species. Efforts are made to select the organisms most likely to be affected by the type of compound and usage pattern; however, there is an inherent uncertainty in extrapolating across phyla. In addition, the Agency's LOCs are intentionally set very low, and conservative estimates are made in the screening level risk assessment to account for these uncertainties.

6.2.3 Extrapolation of long-term environmental effects from short-term laboratory tests

The influence of length of exposure and concurrent environmental stressors to the Barton Springs salamander (i.e., urban expansion, habitat modification, decreased quantity and quality of water in Barton Springs, predators, etc.) will likely affect the species response to simazine. It is possible that additional environmental stressors may increase the Barton Spring salamander's sensitivity to the herbicide. Timing, peak concentration, and duration of exposure are critical in terms of evaluating effects, and these factors will vary both temporally and spatially within the action area. Overall, the effect of this variability may result in either an overestimation or underestimation of risk. However, as previously discussed, the Agency's LOCs are intentionally set very low, and conservative estimates are made in the screening level risk assessment to account for these uncertainties.

6.2.4 Exposure to Pesticide Mixtures

In accordance with the Overview Document and the Services Evaluation Memorandum (U.S. EPA, 2004; USFWS/NMFS, 2004), this assessment considers the single active ingredient of simazine, as well as available information on registered products containing multiple active ingredients in addition to simazine. However, the assessed species and its environments may be exposed to multiple pesticides simultaneously. Interactions of other toxic agents with simazine could result in additive effects, synergistic effects, or antagonistic effects. The available data suggest that pesticide mixtures involving simazine may produce additive effects with other triazine herbicides. Mixtures that have been studied include simazine with other herbicides including atrazine. A number of study authors claim additive effects in aquatic non-vascular plants.

As previously discussed, evaluation of pesticide mixtures is beyond the scope of this assessment because of the myriad of factors that cannot be quantified based on the available data. Those factors include identification of other possible co-contaminants and their concentrations, differences in the pattern and duration of exposure among contaminants, and the differential effects of other physical/chemical characteristics of the receiving waters (e.g. organic matter present in sediment and suspended water). Evaluation of factors that could influence additivity is beyond the scope of this assessment and is beyond the capabilities of the available data to allow for an evaluation. However, it is acknowledged that not considering mixtures could over- or under-estimate risks depending on the type of interaction and factors discussed above.

6.2.5 Sublethal Effects

For an acute risk assessment, the screening risk assessment relies on the acute mortality endpoint as well as a suite of sublethal responses to the pesticide, as determined by the testing of species response to chronic exposure conditions and subsequent chronic risk assessment. Consideration of additional sublethal data in the assessment is exercised on a case-by-case basis and only after careful consideration of the nature of the sublethal effect measured and the extent and quality of available data to support establishing a plausible relationship between the measure of effect (sublethal endpoint) and the assessment endpoints.

Open literature is useful in identifying sublethal effects associated with exposure to simazine. These effects in freshwater fish include, but are not limited to, decreased response from olfactory epithelium and effects on endocrine-mediated processes. However, no data are available to link the sublethal measurement endpoints to direct mortality or diminished reproduction, growth and survival that are used by OPP as assessment endpoints. While the study by Moore and Lower (2001) attempted to relate the results of olfactory perfusion assays to decreased predator avoidance and homing response in salmon, there a number of uncertainties associated with the study that limit its utility. OPP acknowledges that sublethal effects have been associated with simazine exposure in aquatic systems; however, there are insufficient data to definitively link the measurement endpoints to assessment endpoints. To the extent to which sublethal effects are not considered in this assessment, the potential direct and indirect effects of simazine on the Barton Springs salamander may be underestimated.

6.3 Assumptions Associated with the Acute LOCs

The risk characterization section of this endangered species assessment includes an evaluation of the potential for individual effects. 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 for the effects study corresponding to the taxonomic group for which the LOCs are exceeded.

Sufficient dose-response information was not available to estimate the probability of an individual effect on freshwater fish (used as a surrogate for the Barton Springs

salamander) and freshwater invertebrates (dietary food items of the Barton Springs salamander). Based on a lack of dose-response information for the midge, the probability of an individual effect was calculated using the default probit dose response curve slope of 4.5 (with lower and upper confidence intervals of 2 and 9) (Urban and Cook, 1986). It is unclear whether the probability of an individual effect for the salamander (based on toxicity data from freshwater fish) and invertebrates would be higher or lower, given a lack of simazine dose-response information for other freshwater species. However, the assumed probit dose response slope for freshwater fish and invertebrates of 4.5 would have to decrease to approximately 0.4 to 1.1 to cause an effect probability ranging between 1 in 10 and 1 in 100, respectively, for freshwater fish and invertebrates.

7. Summary of Direct and Indirect Effects to the Barton Springs Salamander

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 simazine to the Barton Springs salamander. A summary of the risk conclusions and effects determination for the Barton Springs salamander, given the uncertainties discussed in Section 6, is presented in Table 7.1.

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