

Risks of Atrazine Use to Federally Listed Endangered Barton Springs Salamanders (Eurycea sosorum)

August 22, 2006

Risks of Atrazine Use to Federally Listed Endangered Barton Springs Salamanders

(Eurycea sosorum)

Pesticide Effects Determination

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

August 22, 2006

Table of Contents

1.	Executive Summary	6
2.	Problem Formulation	8
2		
2		
2	1	
2		
	2.4.1 Environmental Fate and Transport Assessment	
	2.4.2 Mechanism of Action.	
	2.4.3 Use Characterization	
2		
2	1	
2		
2		
	2.8.1 Risk Hypotheses	
	2.8.2 Diagram.	
3.	Exposure Assessment	28
3	-	
3		
5	3.2.1 Background	
	3.2.2 Geology/Hydrogeology	
	3.2.3 Conceptual Model of Exposure	
	3.2.4 Existing Monitoring Data	
	3.2.5 Modeling Approach	
	3.2.5.1 Model Inputs	
	3.2.6 Individual Scenario Results	
	3.2.6.1 Residential	
	3.2.6.2 Turf	
	3.2.6.3 Fallow/Idle Land	
	3.2.6.4 Rights-of-Way	
	3.2.7 Characterization	
4.	Effects Assessment	64
4		
•	4.1.1 Toxicity to Freshwater Fish	
	4.1.1.1 Freshwater Fish: Acute Exposure (Mortality) Studies	
	4.1.1.2 Freshwater Fish: Chronic Exposure (Growth/Reproduction	
	4.1.1.3 Freshwater Fish: Sublethal Effects and Additional Open Li	iterature
	Information	
	4.1.2 Toxicity to Aquatic-phase Amphibians	
	4.1.2.1 Amphibians: Open Literature Data on Mortality	
	4.1.2.2 Amphibians: Open Literature Data on Sublethal Effects	
	4.1.3 Toxicity to Freshwater Invertebrates	71

Ջ.	Refe	rences	9
7.	Sum	mary of Direct and Indirect Effects to the Barton Springs Salamander	96
6	5.3	Assumptions Associated with the Acute LOCs	.95
,	6.2.5	J 1	
		laboratory tests	
	6.2.4		_
	6.2.3	j - C	94
	6.2.2	\mathcal{E} 1	
	6.2.1	\mathcal{L}	
	6.2		
	6.1.3		
	6.1.2		
	6.1.1	\mathcal{C}	
	6.1	Exposure Assessment Uncertainties	
6.	Unce	ertainties	
		·	
	3.4.3	(Freshwater Aquatic Plants)	9(
	5.2.3	` '	00
	5.2.2		
	5.2.1		
	5.2	Risk Description	
	3.	and/or Primary Productivity (Freshwater Aquatic Plants)	Q,
	5	(Freshwater Invertebrates	.ŏ₄
	5.	1.2.1 Evaluation of Potential Indirect Effects via Reduction in Food Items	Q'
	5.1.2		8
	5.1.1		
		Risk Estimation	
5.		Characterization	
_	Dial	Characterization	70
	4.4	Includin Dalavase Review	/ 8
		Endangered Species Levels of Concern	
		Use of Probit Slope Response Relationship to Provide Information on the	
		Community-Level Endpoints: Threshold Concentrations	
	4.1.5		
		1.4.1 Aquatic Plants: Laboratory Data	
	4.1.4	J 1	
		1.3.3 Freshwater Invertebrates: Open Literature Data	
		1.3.2 Freshwater Invertebrates: Chronic Exposure Studies	.72
	4.	1.3.1 Freshwater Invertebrates: Acute Exposure Studies	

List of Tables

Table 1.1. Effects Determination Summary for the Barton Springs Salamander	8
Table 2.1. Summary of Assessment Endpoints and Measures of Ecological Effect	. 25
Table 3.1. Information for the Barton Springs Salamander Endangered Species	
Assessment	. 29
Table 3.2. Summary of USGS Monitoring Data from the Four Springs Comprising	
Barton Springs	40
Table 3.3. Summary of PRZM/EZAMS Environmental Fate Data Used for Aquatic	2
Exposure Inputs for Atrazine Endangered Species Assessment for the	
Barton Springs Salamander	45
Table 3.4. Land Cover Adjustment Factors for the Action Area in the Barton Spring	gs
Segment of the Edwards Aquifer (BSSEA)	. 47
Table 3.5. Summary of PRZM Output EECs for all Modeled (Edge of Field	
Concentrations with Base Flow Incorporated).	
Table 3.6. Comparison of Maximum, Typical, and 90th Percentile Label Rates and	
Number of Applications	. 59
Table 3.7. Comparison of Residential and Rights-of-Way EECs Assuming Variable	е
Percentages of Overspray (0, 1, and 10%)	61
Table 3.8. Comparison of Residential EECs (granular) with 1% Over Spray and	
Variable Percentages of Impervious Surface (10 and 30%)	62
Table 3.9. Comparison of Residential EECs (granular) with 10% Overspray and	
Variable Percentages of Impervious Surface (10 and 30%)	. 62
Table 3.10. Comparison of Residential EECs (granular) Assuming Various	
Percentages of Treated ¹ / ₄ Acre Lot (10 and 50%)	
Table 4.1. Aquatic Toxicity Profile for Atrazine	
Table 4.2. 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 Spring	
Salamander via Direct Acute Effects on Dietary Items	
Table 5.3. Summary of RQs Used to Estimate Indirect Effects to the Barton Spring	
Salamander via Direct Chronic Effects on Dietary Items	
Table 5.4. Summary of RQs Used to Estimate Indirect Effects to the Barton Spring	
Salamander via Direct Effects on Aquatic Plants	
Table 5.5. Summary of RQs Used to Assess Potential Risk to Freshwater Invertebra	
Food Items of the Barton Springs Salamander	. 89
Table 5.6. Summary of Modeled Scenario Time-Weighted EECs with Threshold	
Concentrations for Potential Community-Level Effects	
Table 7.1. Effects Determination Summary for the Barton Springs Salamander	. 96

List of Figures

Figure 2.1 National Extent of Atrazine Use (lbs)	16
Figure 2.2. Barton Springs Complex	19
Figure 2.3. Barton Springs Salamander Action Area	23
Figure 2.4. Conceptual Model for Barton Springs Salamander	26
Figure 3.1. Barton Springs Segment of the Edwards Aquifer with HydroZones	31
Figure 3.2. Hydrogeologic Cross Section of the Barton Springs Segment of the Ed	lwards
Aquifer Showing Dominant Flow Pathways Within Each Hydrozone	33
Figure 3.3. Conceptual Model of Surface and Subsurface Flow Within the Barton	Springs
Segment of the Edwards Aquifer Relative to the Barton Springs Salam	ander34
Figure 3.4. Flow paths within Recharge Zone of the Barton Springs Segment of th	e
Edwards Aquifer	36
Figure 3.5. Location of Surface Water Sites within the Barton Springs Segment	41
Figure 3.6. Location of Groundwater Sites Within the Barton Springs Segment	42
Figure 3.7. Flow Hydrograph Data for Barton Springs	43
Figure 3.8. Conceptual Model of Paired Residential/Impervious Scenarios	50
Figure 3.9. Percentage of Impervious Surface Coverage in Vicinity of Barton Spri	ngs 52
Figure 3.10. Representative Time Series Output from Paired Residential/Impervio	us
PRZM Scenario for Granular Applications	53
Figure 3.11. Conceptual Model of Rights-of-Way Scenario	56
Figure 4.1. Summary of Reported Acute LC ₅₀ /EC ₅₀ Values in Freshwater Inverteb	rates
for Atrazine	72
Figure 4.2. Use of Threshold Concentrations in Endangered Species Assessment	77

Appendices

Appendix A	Ecological Effects Data
Appendix B	Supporting Information for the Aquatic Community-Level
	Threshold Concentrations
Appendix C	Supporting Information for the Scenario Development
Appendix D	Status and Life History of the Barton Springs Salamander
Appendix E	Stepwise Approach to Modeling for the Barton Springs
	Salamander Endangered Species Assessment (Using the
	Residential Scenario as an Example)
Appendix F	Incident Database Information
Appendix G	RQ Method and LOCs

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 the herbicide atrazine on the survival, growth, and reproduction of this Federally 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), the August 5, 2004 Joint Counterpart Endangered Species Act Section 7 Consultation Regulations specified in 50 CFR Part 402 (USFWS/NMFS, 2004a; FR 69 47732-47762), 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 action area for the Barton Springs salamander is defined by those areas within the hydro geologic framework of the Barton Springs segment of the Edwards Aquifer.

Environmental fate and transport models were used to estimate high-end exposure values expected to occur in the Barton Springs action area as a result of agricultural and non-agricultural atrazine use in accordance with label directions. Modeled concentrations provide "edge-of-field" estimates of exposure which are intended to represent atrazine concentrations transported with runoff water directly to Barton Springs via subsurface flow through the fractured karst limestone of the Edwards Aquifer. Estimated high-end exposure values were compared with available monitoring data, although the monitoring data are unlikely to capture the upper bounds of exposure due to sampling frequency. In general, the modeled peak exposure estimates are two to ten times higher than concentrations seen in the monitoring data, while the annual average concentrations based on modeling are consistent with those seen in monitoring. The highest overall modeled exposures were predicted to occur from residential uses of atrazine within the action area.

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 vertebrates, including fish, which are generally used as a surrogate for amphibians, as well as available aquatic-phase amphibian data from the open literature. Given that the salamander's prey items and habitat requirements are dependant on the availability of freshwater aquatic invertebrates and aquatic plants, respectively, toxicity information for these taxonomic groups is also discussed. In addition to the registrant-submitted and open literature toxicity information, indirect effects to Barton Springs salamanders, via impacts to aquatic plant community structure and function are also evaluated based on

time-weighted threshold concentrations that correspond to potential aquatic plant community-level effects.

Degradates of atrazine include hydroxyatrazine (HA), deethylatrazine (DEA), deisopropylatrazine (DIA), and diaminochloroatrazine (DACT). Comparison of available toxicity information for the degradates of atrazine indicates lesser aquatic toxicity than the parent for freshwater and estuarine/marine fish, invertebrates, and aquatic plants. Because the degradates are not of greater toxicological concern than atrazine, concentrations of the atrazine degradates are not assessed further, and the focus of this assessment is parent atrazine.

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 atrazine use within the action area has the potential to adversely affect the Barton Springs salamander via direct toxicity to the salamander 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 Spring 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 preliminary conclusion of "may effect". If a determination is made that use of atrazine within the action area "may affect" the Barton Spring salamander, additional information is considered to refine the potential for exposure and effects, and the best available data are 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.

The best available data suggest that atrazine will either have no effect or is not likely to adversely affect the Barton Springs salamander by direct toxic effects or by indirect effects resulting from effects to aquatic invertebrates and plants. A summary of the risk conclusions and effects determination for the Barton Springs salamander is presented in Table 1.1. Further information on the results of the effects determination is included as part of the Risk Description in Section 5.2.

Table 1.1. Effects Determination Summary for the Barton Springs Salamander

Assessment Endpoint	Effects Determination	Basis for Determination
Survival, growth, and reproduction of Barton Springs salamander individuals via direct effects	No effect	No acute and chronic LOCs are exceeded.
Indirect effects to Barton Springs salamander via reduction of prey (i.e., freshwater invertebrates)	May affect, but not likely to adversely affect	Acute LOCs are exceeded based on the most sensitive ecotoxicity value for the midge; however RQs for other dietary items (amphipods, leeches, snails) are less than LOCs. Based on the non-selective nature of feeding behavior in the Barton Springs salamander and low magnitude of anticipated individual effects to all evaluated prey species, atrazine is not likely to indirectly affect the Barton Springs salamander via a reduction in freshwater invertebrate food items. This finding is based on insignificance of effects (i.e., effects to freshwater invertebrates are not likely to result in "take" of a single Barton Springs salamander) and discountability (i.e., the effect to freshwater invertebrates is extremely unlikely to occur given the estimated individual event probability of 1 in 45.5 million).
Indirect effects to Barton Springs salamander via reduction of habitat and/or primary productivity (i.e., aquatic plants)	May affect, but not likely to adversely affect	Although atrazine use may directly affect individual vascular and non-vascular aquatic plants in Barton Springs, its use within the action area is not likely to adversely affect the Barton Springs salamander via indirect community-level effects to aquatic vegetation. Predicted 14-, 30-, 60-, and 90-day EECs for all modeled atrazine use scenarios within the action area are well below the threshold concentrations representing community-level effects. This finding is based on insignificance of effects (i.e., community-level effects to aquatic plants are not likely to result in "take" of a single Barton Springs salamander).

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. This assessment was completed in accordance with the August 5, 2004 Joint Counterpart Endangered Species Act (ESA) Section 7 Consultation Regulations specified in 50 CFR Part 402 (USFWS/NMFS, 2004a; FR 69 47732-47762). The structure of this risk assessment is based on guidance contained in EPA's *Guidance for Ecological Risk Assessment* (U.S. EPA, 1998), the Services' *Endangered Species Consultation Handbook* (USFWS/NMFS, 1998) and procedures outlined in the Overview Document (U.S. EPA, 2004).

2.1 Purpose

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)*. The purpose of this ecological risk assessment is to make an "effects determination," as directed in Section 7(a) (2) of the ESA, for the Barton Springs salamander (*Eurycea sosorum*) by evaluating the potential direct and indirect effects resulting from use of the herbicide atrazine (6-chloro-N-ethyl-N-isopropyl-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.

In this endangered species assessment, direct and indirect effects to the Barton Springs salamander are evaluated in accordance with the screening-level methodology described in the Agency's Overview Document (U.S. EPA, 2004). It should be noted, however, that the indirect effects analysis in this assessment utilizes more refined data than is generally available to the Agency. Specifically, a robust set of microcosm and mesocosm data and aquatic ecosystem models are available for atrazine that allowed EPA to refine the indirect effects associated with potential aquatic community-level effects (via aquatic plant community structural change and subsequent habitat modification) to the Barton Springs salamander. Use of such information is consistent with the guidance provided in the Overview Document, which specifies that "the assessment process may, on a case-by-case basis, incorporate additional methods, models, and lines of evidence that EPA finds technically appropriate for risk management objectives" (Section V, page 31 of U.S. EPA, 2004).

As part of the "effects determination", the Agency will reach one of the following three conclusions regarding the potential for atrazine to affect the Barton Springs salamander:

- "No effect";
- "May affect, but not likely to adversely affect"; or
- "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 atrazine'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 atrazine 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 and potential community-level effects to aquatic plants. 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

Atrazine is currently registered as a herbicide in the U.S. to control annual broadleaf and grass weeds in corn, sorghum, sugarcane, and other crops. In addition to food crops, atrazine is also used on a variety of non-food crops, forests, residential/industrial uses, golf course turf, recreational areas, and rights-of-way. It is one of the most widely used herbicides in North America (U.S. EPA, 2003a).

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

This ecological risk assessment is for currently registered uses of atrazine in 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 atrazine include hydroxyatrazine (HA), deethylatrazine (DEA), deisopropylatrazine (DIA), and diaminochloroatrazine (DACT). Comparison of available toxicity information for the degradates of atrazine indicates lesser aquatic toxicity than the parent for freshwater fish, invertebrates, and aquatic plants. Specifically, the available degradate toxicity data for HA indicates that it is not toxic to freshwater fish and invertebrates at the limit of its solubility in water. In addition, available aquatic plant degradate toxicity data for HA, DEA, DIA, and DACT report non-definitive EC₅₀ values (i.e., 50% effect was not observed at the highest test concentrations) at concentrations that are 700 to 10,000 times higher than the lowest reported aquatic plant EC₅₀ value for parent atrazine. Given the lesser toxicity of the degradates, as compared to the parent, the focus of this assessment is parent atrazine. A detailed summary of the available ecotoxicity information for all of the atrazine degradates is presented in Appendix A.

2.3 Previous Assessments

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

(i.e., lakes/reservoirs, streams, and estuarine areas) equaled or exceeded concentrations shown to cause adverse effects to aquatic communities and populations of aquatic organisms. The results of the refined aquatic ecological assessment indicated that exposure to atrazine is likely to result in adverse community-level and population-level effects to aquatic communities at concentrations greater than or equal to $10\text{-}20~\mu\text{g/L}$ on a recurrent basis or over a prolonged period of time.

During this time, the Agency extensively reviewed a probabilistic ecological risk assessment submitted by the registrant (Giddings et al., 2000). The Agency's review of Syngenta's probabilistic risk assessment is included in Appendix XVII of the 2003 atrazine IRED. EPA's refined risk assessment incorporates some of the data submitted by the registrant in its probabilistic risk assessment.

The results of the Agency's ecological assessments for atrazine are fully discussed in the January 31, 2003, Interim Reregistration Eligibility Decision (IRED)¹. Because the Agency had determined that atrazine shares a common mechanism of toxicity with the structurally-related chlorinated triazines simazine and propazine, a cumulative human health risk assessment for the triazines was necessary before the Agency could make a final determination of reregistration eligibility. However, the Agency issued the interim decision in order to identify risk reduction measures that were necessary to support the continued use of atrazine. The January 2003 IRED requires extensive drinking water monitoring in Community Water Systems (CWSs) where atrazine levels have exceeded or are predicted to have the potential to exceed drinking water levels of concern. In addition, the need for the following information related to potential ecological risks was established: 1) an ecological monitoring program of potentially vulnerable water bodies in corn, sorghum, and sugarcane use areas; and 2) further information on potential amphibian gonadal developmental responses to atrazine.

EPA issued an addendum on October 31, 2003 that updated the IRED issued on January 31, 2003 (U.S. EPA, 2003b). This addendum describes new scientific developments pertaining to ecological monitoring and mitigation of watersheds and potential effects of atrazine on endocrine-mediated pathways of amphibian gonadal development.

The January 2003 IRED required atrazine registrants to develop a watershed monitoring protocol. The resulting protocol identifies 40 indicator watersheds in corn and sorghum growing areas in which monitoring has been required for a two-year period within each watershed. The first 20 watersheds were monitored in 2004 and 2005. The second set of 20 watersheds was monitored in 2005, and the second year of sampling for these watersheds is currently in progress. The goal of the monitoring is to ascertain the extent to which any of the watersheds have streams with atrazine concentrations that could cause significant changes in aquatic plant community structure, the most sensitive endpoint in the aquatic ecosystem. Streams in watersheds exceeding the Agency's levels of concern will be subject to mitigation consistent with watershed management principles

¹ The 2003 Interim Reregistration Eligibility Decision for atrazine is available via the internet at http://www.epa.gov/oppsrrd1/REDs/0001.pdf

described by the Agency's Office of Water program requirements (http://www.epa.gov/owow/tmdl/). These monitoring sites are representative of 1,172 watersheds determined to be among the most vulnerable to atrazine surface water loading from use on corn and sorghum. Therefore, the results from the 40 watersheds will be used to determine if further monitoring or remedial efforts are needed in the larger population of watersheds. EPA has selected an atrazine level of concern (LOC) that is based on significant aquatic community effects consistent with those described in the Office of Pesticide Programs (OPP) 2003 ecological risk assessment (U.S. EPA, 2003a and b) and the Office of Water's (OW) draft atrazine aquatic life criteria (U.S. EPA, 2003c). Further discussion of the aquatic community-level LOC is provided in Section 4.2 and Appendix B of this assessment. Aqueous atrazine concentrations obtained from monitoring studies can be interpreted with the LOC to determine if a water body is likely to be significantly affected.

As discussed in the October 2003 IRED, the Agency also conducted an evaluation of the submitted studies regarding the potential effects of atrazine on amphibian gonadal development and presented its assessment in the form of a white paper for external peer review to a FIFRA Scientific Advisory Panel (SAP) in June 2003². In the white paper dated May 29, 2003, the Agency summarized seventeen studies consisting of both open literature and registrant-submitted laboratory and field studies involving both native and non-native species of frogs. The Agency concluded that none of the studies fully accounted for environmental and animal husbandry factors capable of influencing endpoints that the studies were attempting to measure. The Agency also concluded that the current lines-of-evidence did not show that atrazine produced consistent effects across a range of exposure concentrations and amphibian species tested.

Based on this assessment, the Agency concluded and the SAP concurred that there was sufficient evidence to formulate a hypothesis that atrazine exposure may impact gonadal development in amphibians, but there were insufficient data to confirm or refute the hypothesis (http://www.epa.gov/oscpmont/sap/2003/June/junemeetingreport.pdf). Because of the inconsistency and lack of reproducibility across studies and an absence of a dose-response relationship in the currently available data, the Agency determined that the data did not alter the conclusions reached in the January 2003 IRED regarding uncertainties related to atrazine's potential effects on amphibians. The SAP supported EPA in seeking additional data to reduce uncertainties regarding potential risk to amphibians. Subsequent data collection has followed the multi-tiered process outlined in the Agency's white paper to the SAP (U.S. EPA, 2003d). In addition to addressing uncertainty regarding the potential use of atrazine to cause these effects, these studies are expected to characterize the nature of any potential dose-response relationship. A data call-in for the first tier of amphibian studies was issued in 2005 and studies are on-going; however, as of this writing, results are not available.

_

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

2.4 Stressor Source and Distribution

2.4.1 Environmental Fate and Transport Assessment

The following fate and transport description for atrazine was summarized based on information contained in the 2003 IRED (U.S. EPA, 2003a). In general, atrazine is expected to be mobile and persistent in the environment. The main route of dissipation is microbial degradation under aerobic conditions. Because of its persistence and mobility, atrazine is expected to reach surface and ground water. This is confirmed by the widespread detections of atrazine in surface water and ground water. Atrazine is persistent in soil, with a half-life (time until 50% of the parent atrazine remains) exceeding 1 year under some conditions (Armstrong et al., 1967). Atrazine can contaminate nearby non-target plants, soil and surface water via spray drift during application. Atrazine is applied directly to target plants during foliar application, but preplant and pre-emergent applications are generally far more prevalent.

The resistance of atrazine to abiotic hydrolysis (stable at pH 5, 7, and 9) and to direct aqueous photolysis (stable under sunlight at pH 7), and its only moderate susceptibility to degradation in soil (aerobic laboratory half-lives of 3-4 months) indicates that atrazine is unlikely to undergo rapid degradation on foliage. Likewise, a relatively low Henry's Law constant (2.6 X 10^{-9} atm-m³/mol) indicates that atrazine will probably not undergo rapid volatilization from foliage. However, its relatively low octanol/water partition coefficient (Log $K_{ow} = 2$.7), and its relatively low soil/water partitioning (Freundlich K_{ads} values < 3 and often < 1) may somewhat offset the low Henry's Law constant value, thereby possibly resulting in some volatilization from foliage. In addition, its relatively low adsorption characteristics indicate that atrazine may undergo substantial washoff from foliage. It should also be noted that foliar dissipation rates for numerous pesticides have generally been somewhat greater than otherwise indicated by their physical chemical and other fate properties.

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

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

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

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

2.4.2 Mechanism of Action

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

2.4.3 Use Characterization

An analysis of available usage and land cover information, including extensive discussions with local experts in the fields of agriculture and soil science, was completed to determine which atrazine uses are likely to be present in the action area. This evaluation is intended to place priority on those atrazine use areas likely to be in closest proximity to the salamander's habitat. The analysis indicates that of all registered uses for atrazine, the non-agricultural uses are likely to result in the highest exposures to the salamander. This is due to the preponderance of potential residential and other non-agricultural use sites (i.e., recreational and rights-of-way) in the immediate vicinity of Barton Springs, and the fact that very little agricultural crops other than fallow uses for the Conservation Reserve Program (CRP) are actually grown in the action area. Further details on the analysis used to make this determination are discussed below and included in Appendix C.

Critical to the development of appropriate modeling scenarios and to the evaluation of the appropriate model inputs is an assessment of usage information. The Agency's Biological and Economic Analysis Division (BEAD) provided an analysis of both national and local use information for atrazine (Kaul et al., 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-NASS³ and EPA proprietary data⁴ 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 atrazine, including pounds of active ingredient applied, percent of crop treated, number of applications per acre, application rate per acre, and base acres 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 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. Estimates of the percent of each crop treated, the number of applications and the application rate in each county are assumed to be the same as the state level estimates. Apportioning the usage in this manner may underestimate or overestimate the actual usage in a particular county.

In this analysis, the Agency gathered information on the agricultural uses of atrazine in the three counties (Hays, Travis, and Blanco) located within or adjacent to the action area for atrazine in the context of the Barton Springs salamander. Information was available on crops for which atrazine is registered, amounts of atrazine used by county, application rates, methods of application, application timing, and intervals between applications. Usage information is critical in determining which uses should be modeled, while the application methods, intervals, and timing are critical model inputs for estimating atrazine exposure. While the modeling described below relies initially on maximum label application rates and numbers of applications, the information on typical ranges of application rates and number of applications is useful for characterization of the modeling results. In general, for agricultural uses, atrazine is used in limited amounts relative to national use patterns in Hays, Travis and Blanco counties.

Nationally atrazine has the second largest poundage of any herbicide in the U.S. and is widely used to control broadleaf and many other weeds, primarily in corn, sorghum and sugarcane. As a selective herbicide, atrazine is applied pre-emergence and post-emergence. Figure 2.1 presents the national distribution of atrazine use from data collected between 1998 and 2004 and used in the cumulative triazine assessment (U.S. EPA, 2006a; Kaul et al., 2005).

and state. See http://www.usda.gov/nass/pubs/estindx1.htm#agchem.

15

³ 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

⁴ US EPA proprietary usage databases provide estimates of pesticide usage for select agricultural use sties by chemical, crop and state.

National Distribution of Atrazine Use (lbs)

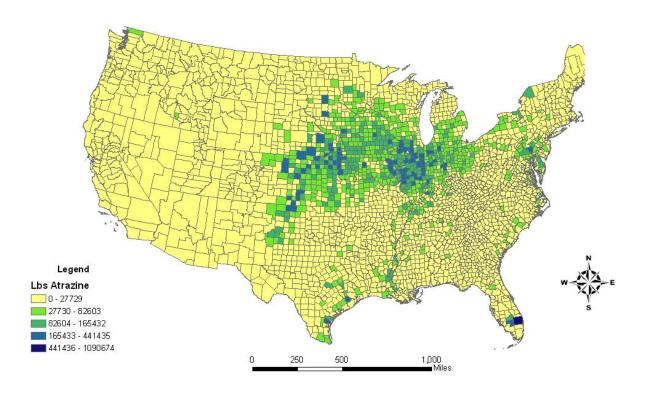


Figure 2.1 National Extent of Atrazine Use (lbs)

Locally, county level estimates of atrazine 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. In general, this information suggests that, in the three county area, approximately 20,000 lbs of atrazine were used on corn, sorghum, wheat, cotton, and pecans in descending order of total pounds applied.

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; see Appendix C for more detail) 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. The land cover analysis indicates that, of possible agricultural uses for atrazine, only the fallow/idle land use is likely to be present in the action area. Land cover data also suggest that many of the currently registered non-agricultural atrazine uses could not be excluded from the assessment (see Appendix C). However, the non-agricultural forestry use of atrazine 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). The usage analysis suggests that atrazine may be used on outdoor ornamental nurseries, although subsequent information (Shay, 2006, personal communication; DeLong-Amaya, 2006, personal communication; City of Austin, 2003a and b) suggests these uses are very limited in nature and are not assessed (see Appendix C for more detail). Based on this analysis, a suite of scenarios was developed, including a single agricultural scenario (fallow/idle land) and four non-agricultural scenarios (residential, impervious, rights-of-way, and turf) using local land cover, soils, and agronomic and climatic data specific to Travis and Hays counties in Texas.

Application rates, number of applications, and application intervals were also estimated (Zinn and Jones, 2006) for the three-county area. The minimum and maximum annual application rates for atrazine were obtained from EPA data sources. Application rates are provided at the state level for only crops grown in Blanco, Hays, and Travis counties on which atrazine is registered. The minimum application rate was reported as the minimum rate range. The 90th percentile application rate was reported as the highest application rate at which at least 90% of the averaged total area is treated. Therefore, at least 90% of the area is treated at this rate or less.

The only typical information available for a use site included in this assessment is for fallow land (Kaul et al., 2006). This was reported as meadow use; however for this analysis, it is used as a surrogate for atrazine use on fallow land under the CRP. Application rates are in units of pounds per acre. The minimum reported application rates for fallow ranged between 0.25 lbs/acre and 0.5 lbs per acre. The typical rate was reported as 0.8 lbs/acre for fallow, while the 90th percentile application rate for fallow was 2 lbs/acre. For fallow uses, the typical number of applications was 1. Information on typical intervals for fallow was not available. Overall, atrazine is applied as a preplant or pre-emergent herbicide to most sites in this part of Texas in late winter to mid spring. No information was available for other agricultural crops and no data were available for non-agricultural uses.

2.5 Assessed Species

A brief introduction to the Barton Springs salamander, including a summary of habitat, diet, and reproduction data relevant to this endangered species risk assessment is provided below. Further information on the status and life history of the Barton Springs salamander is provided in Appendix D.

The Barton Springs salamander, shown in Figure D.1 of Appendix D, 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 available information indicates that the Barton Springs salamander is restricted to the immediate vicinity of the four spring outlets that make up the Barton Springs complex (Figure 2.2), 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).

Based on survey results, Barton Springs salamanders appear to prefer areas near the spring outflows, with clean, loose substrate for cover, but may also be found in the aquatic plants, such as moss. In addition to providing cover, moss and other aquatic plants harbor a variety and abundance of the freshwater invertebrates that salamanders eat.

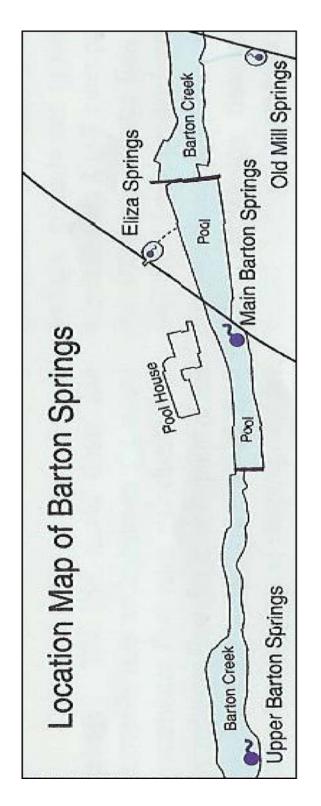


Diagram from Hauwert et al., Barton Springs Edwards Aquifer Conservation District Report

Figure 2.2. Barton Springs Complex (from Hauwert et al., 2005)

2.6 Action Area

It is recognized that the overall action area for the national registration of atrazine uses is likely to encompass considerable portions of the United States based on the large array of both agricultural and non-agricultural uses. However, the scope of this assessment limits consideration of the overall action area to those portions that may be applicable to the protection of the 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 atrazine may be expected to have on the environment, the exposure levels to atrazine that are associated with those effects, and the best available information concerning the use of atrazine 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 atrazine 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 2.3 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 Figure 2.2), 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 principally by the fracture nature of limestone within portion 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 via 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). A more detailed 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 atrazine 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) was analyzed and interviews with the local agricultural sector (Davis, 2006; Garcia, 2006; Perez, 2006; see Appendix C for more detail) were conducted to refine the characterization of potential atrazine 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 atrazine were unlikely, no areas could be excluded from the final action area based on usage and land cover data.

Finally, the environmental fate properties of atrazine 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 atrazine and the fact that atrazine 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 atrazine loading is considered significant are typically high use areas (Midwest corn belt), and the area surrounding Barton Springs is not a high use atrazine area, the expected loadings from atmospheric transport of atrazine are not expected to approach concentrations predicted by modeling (see Section 3.2.5).

Atrazine has been documented to be transported away from the site of application by both spray drift and volatilization. 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 significant route of exposure because the source area for atrazine reaching the springs is generally removed from the spring system where the salamander resides, and the atrazine 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 atrazine 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 atrazine, coupled with the available monitoring data, suggest that long range transport of volatilized atrazine 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 atrazine 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) 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 atrazine 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.

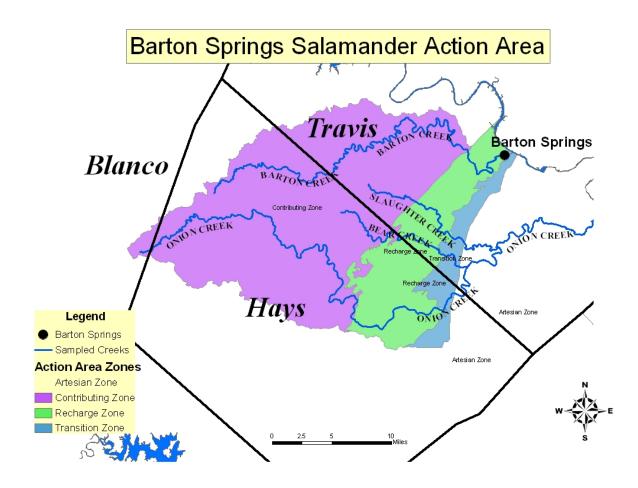


Figure 2.3. Barton Springs Salamander Action Area

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 atrazine (i.e., runoff), and the routes by which ecological receptors are exposed to atrazine-related contamination (i.e., direct contact).

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. 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. Specific measures of ecological effect are evaluated based on acute and chronic toxicity information from registrant-submitted

_

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

guideline tests that are performed on a limited number of organisms. Given that the results of the required registrant-submitted amphibian toxicity tests are not available for this assessment, it is assumed that fish and aquatic-phase amphibian toxicities are similar. Birds are generally considered as surrogates for terrestrial-phase amphibians; however, Barton Springs salamanders are neotenic (i.e., retain gills throughout their lives) and are aquatic-phase amphibians. Therefore, fish are used as a surrogate for amphibian/salamanders, in accordance with guidance specified in the Agency's Overview Document (U.S. EPA, 2004). Additional ecological effects data from the open literature, including effects data on salamanders and aquatic freshwater microcosm and mesocosm data were also considered.

Measures of effect from microcosm and mesocosm studies provide an expanded view of potential indirect effects of atrazine on aquatic organisms, their populations and communities in the laboratory, in simulated field situations, and in actual field situations. With respect to the microcosm and mesocosm data, threshold concentrations on aquatic community effects were determined from complex time variable atrazine exposure profiles (chemographs) within these experimental studies. Methods were developed to estimate ecological community responses for any possible atrazine chemograph based on the relationships in the micro- and mescocosm study results. This information was used to determine whether a certain exposure profile within a particular use site and/or action area may have exceeded a level of concern. Ecological modeling with the Comprehensive Aquatic Systems Model (CASM) (Bartell et al. 2000, Bartell et al. 1999, and DeAngelis et al., 1989) was used to calibrate the measured atrazine chemographs in order to estimate direct and indirect effects of atrazine and to project potential changes in aquatic community structure and function.

A complete discussion of all the toxicity data available for this risk assessment, including use of the CASM model and associated aquatic community-level threshold concentrations, and the resulting measures of ecological effect selected for each taxonomic group of concern are 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 atrazine is provided in Table 2.1.

Table 2.1. Summary of Assessment Endpoints and Measures of Ecological Effect

Assessment Endpoint	Measures of Ecological Effect		
1. Survival, growth, and reproduction of Barton	1a. Rainbow trout acute LC ₅₀		
Springs salamander individuals via direct effects	1b. Brook trout chronic NOAEC		
	1c. Open literature lab and field NOAEC data for		
	salamanders		
2. Survival, growth, and reproduction of Barton	2a. Midge acute EC ₅₀		
Springs salamander individuals via indirect effects	2b. Scud chronic NOAEC		
on prey (i.e., freshwater invertebrates)	2c. Acute EC/LC ₅₀ data for freshwater invertebrates		
	that are potential food items for the Barton Spring		
	salamander		
3. Survival, growth, and reproduction of Barton	3a. Vascular plant (duckweed) acute EC ₅₀		
Springs salamander individuals via indirect effects	3b. Non-vascular plant (freshwater algae) acute		
on habitat and/or primary productivity (i.e., aquatic	EC ₅₀		
plant community)	3c. Microcosm/mesocosm threshold concentrations		
	showing aquatic primary productivity community-		
	level effects		

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 atrazine to the environment. Based on the results of the 2003 atrazine IRED (U.S. EPA, 2003a), the following risk hypotheses are presumed for this endangered species assessment:

- Atrazine 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;
- Atrazine 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
- Atrazine 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 atrazine on the Barton Springs salamander is shown in Figure 2.4. 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.

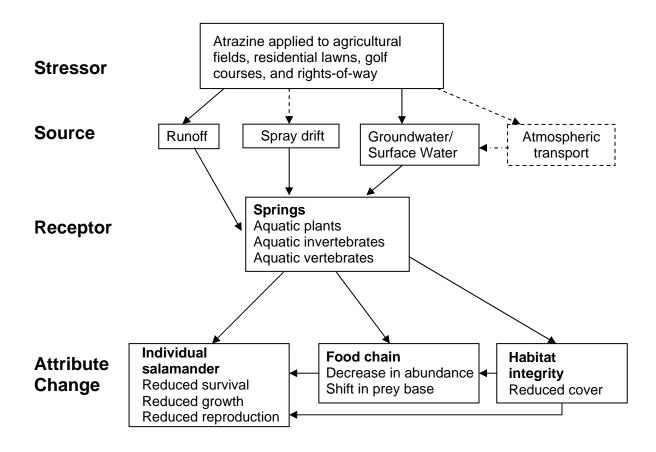


Figure 2.4. Conceptual Model for Barton Springs Salamander

The conceptual model provides an overview of the expected exposure routes for Barton Springs salamanders within the atrazine 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 atrazine 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 atrazine may indirectly affect the Barton Springs salamander via reduction in food and habitat availability. The available data indicate that atrazine is not likely to bioconcentrate in aquatic food items, with fish bioconcentration factors (BCFs) ranging from 2 to 8.5 (U.S. EPA, 2003c). Therefore, bioconcentration of atrazine in salamanders via the diet was not considered as a significant route of exposure.

Individual Barton Springs salamanders with the greatest potential to experience direct adverse effects from atrazine use are those that occur in surface water and/or groundwater with the highest concentrations of atrazine. 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, atrazine 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 atrazine use within Zilker Park, it is currently being revised to specifically restrict atrazine use within the Park in the future (personal communication with Elizabeth McVeety, pesticide applicator at Zilker Park, July 24, 2006). Given that atrazine is not used within the Barton Springs area, it is unlikely that atrazine 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 a significant route of exposure and are not further addressed in this risk assessment.

The source and mechanism of release of atrazine into surface and groundwater are ground and aerial application via foliar spray and coated fertilizer granules to agricultural (i.e., fallow/idle land) and non-agricultural sites (i.e., golf courses, residential lawns, rights-of-way, etc). Surface water runoff from the areas of atrazine 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 significant route of exposure because the source area for atrazine is generally removed from the spring system where the salamander resides, and the atrazine exposures that reach the springs do so via subsurface flow. Atmospheric transport is not considered as a significant 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 (Majewski et al., 2000; Majewski and Capel, 1995; Capel et al., 1994).

3. Exposure Assessment

3.1 Label Application Rates and Intervals

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

In the January and October 2003 IREDs, EPA stipulated numerous changes to the use of atrazine including label restrictions and other mitigation measures designed to reduce risk to human health and the environment (U.S. EPA, 2003a and b). Specifically pertinent to this assessment, the Agency entered into a Memorandum of Agreement (MOA) with the atrazine registrants. In the MOA, the Agency stipulated that certain label changes must be implemented on all manufacturing-use product labels for atrazine and on all end-use product labels for atrazine prior to the 2005 growing season including cancellation of certain uses, reduction in application rates, and requirements for harmonization across labels including setbacks from waterways. Specifically, the label changes stipulate no use of atrazine within 50 feet of sinkholes, within 66 feet of intermittent and perennial streams, and within 200 feet of lakes and reservoirs. The modeling discussed below predicts edge of field concentrations and thus spray drift is not quantitatively included in the predicted exposures. It is expected that a setback distance will result in a reduction in loading due to runoff across the setback zone; however, current models do not address this reduction quantitatively. Therefore, these restrictions are not quantitatively evaluated in this assessment. A qualitative discussion of the potential impact of these setbacks on estimated environmental concentrations of atrazine for the Barton Springs salamander is discussed further in Section 3.2.3. Table 3.1 provides a summary of label application rates for atrazine uses evaluated in this assessment.

Currently registered non-agricultural uses of atrazine within the Barton Springs action area include residential areas such as playgrounds and home lawns, turf (golf courses and recreational fields), and rights-of-way. Agricultural uses within the Barton Springs action area include fallow/idle land⁶ including Conservation Reserve Program (CRP) lands in Texas. According to use data gathered by EPA, there is no agricultural use of atrazine on corn or sorghum within the Barton Springs action area, although corn and sorghum represent the greatest use nationally.

Atrazine is formulated as liquid, wettable powder, dry flowable, and granular formulations. Application equipment for the agricultural uses includes ground

_

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

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

Table 3.1. Label Application Information for the Barton Springs Salamander Endangered Species Assessment1

Scenario	Maximum Application Rate (lbs/acre)	Maximum Number of Applications	Date of First Application	Formulation	Method of Application	Interval Between Applications
Residential	2.0	2	April 1	Granular	Ground	30 days
Residential	1.0	2	April 1	Liquid	Ground	30 days
Rights-of- Way	1.0	1	June 1	Liquid	Ground	NA
Fallow/ Idle land	2.25	1	November 1	Liquid	Ground and Aerial	NA
Turf	2.0	2	April 1	Granular	Ground	30 days
Turf	1.0	2	April 1	Liquid	Ground	30 days

 $¹⁻Based\ on\ 2003\ IRED$ and Label Change Summary Table memorandum dated June 12, 2006 (U.S. EPA, 2006b).

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 the springs. The Agency's PRZM model was used to provide edge of field estimates of exposure, which are assumed to be the concentrations of atrazine 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 atrazine 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 atrazine 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. Residential uses were modeled using pervious (1/4 acre lot) and impervious surface scenarios and weighting the output based on local data on the percentage of impervious surfaces in the action area region. Durations of exposure were used to match available ecotoxicity thresholds. The highest overall exposures were predicted to occur from the residential uses of atrazine that are in closest proximity to the spring system. In general, the exposure assessment yields modeled peak exposure estimates that are two to ten times 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 and D.4 of Appendix D. 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 aguifer of the Edwards Aguifer, which trends south-southwest 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 atrazine 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 atrazine exposure to the Barton Springs salamander. Figure 3.1 shows the extent of the BSSEA.

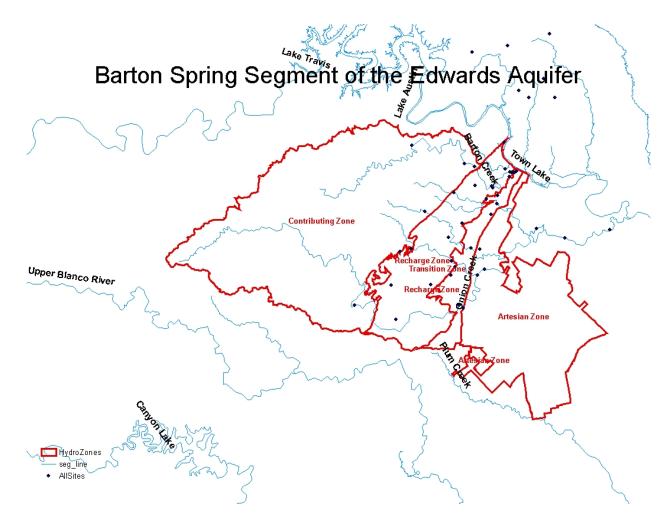


Figure 3.1. Barton Springs Segment of the Edwards Aquifer with HydroZones

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 atrazine reaching the Recharge Zone is likely to have the most immediate and significant impact on Barton Springs.

The Contributing Zone lies due west of the Recharge Zone. In this zone, runoff from sites treated with atrazine is transported via overland flow to surface water streams and ponds. Atrazine 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 atrazine with occasional shorter duration pulses of higher concentrations correlated with precipitation derived 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 Section D.4 of Appendix D.

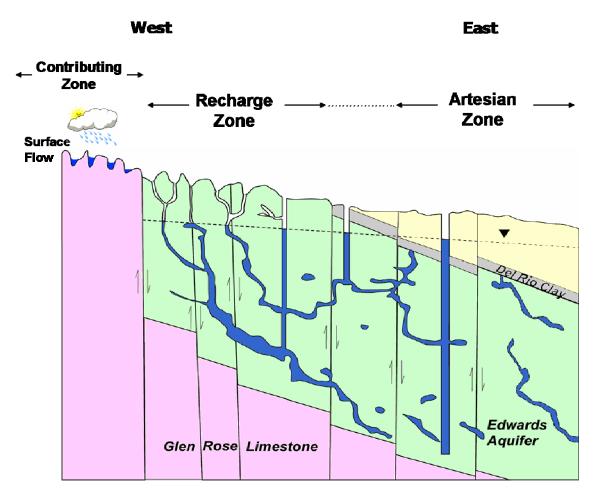


Figure 3.2. Hydrogeologic Cross Section of the Barton Springs Segment of the Edwards Aquifer Showing Dominant Flow Pathways Within Each Hydrozone (Taken from Mahler, 2005a)

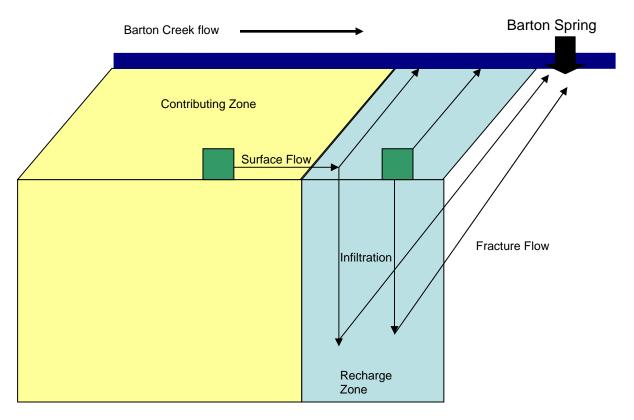


Figure 3.3. Conceptual Model of Surface and Subsurface Flow Within the Barton Springs Segment of the Edwards Aquifer Relative to the Barton Springs Salamander

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 occurr 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).

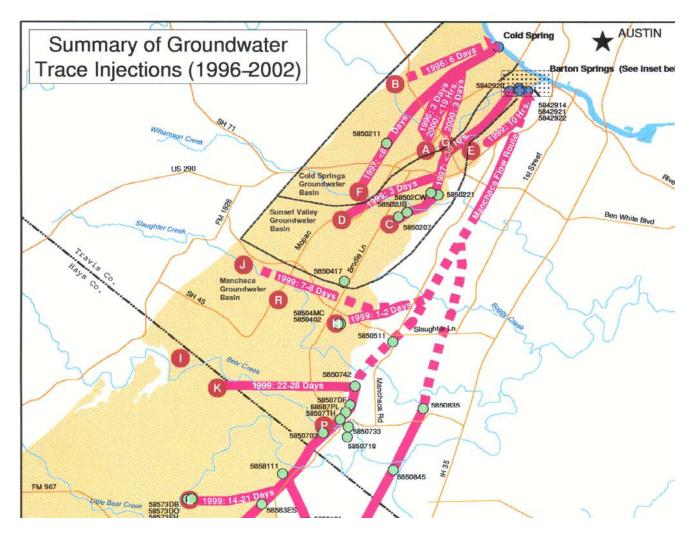


Figure 3.4. Flow paths within Recharge Zone of the Barton Springs Segment of the Edwards Aquifer (Taken from Mahler, 2005a; originally published in Hauwert et al., 2004)

3.2.3 Conceptual Model of Exposure

Given the understanding of the geology/hydrogeology described above, a combination of modeling and monitoring data is needed to assess the potential exposures from atrazine to the Barton Springs salamander. Routes of exposure are dependent on the location of registered use sites for atrazine within the action area (defined in Section 2.6 as the Contributing, Recharge, and Transition Zones), the location of those uses, and locations within the pool system (fractures versus pools) where the salamander resides. For instance, uses that are predominantly within the Recharge Zone of the BSSEA are likely to reach the springs via direct transport through the fractures within the karst zone. Atrazine originating from within the Recharge Zone is analogous to "edge of field" concentrations because atrazine applied within this area may be transported directly from the site of application to the subsurface fractures. Thus, atrazine may move directly from the edge of the treated field (or even from within it) into the fracture system. This route of exposure is expected to be the most conservative (e.g. represent the highest potential exposure).

The interconnected nature of the subsurface network can have a significant influence on mixing, dilution, storage and degradation of flow through karst (Field, 2004). The simplest, and for purposes of this assessment, most conservative assumption, is a straight conduit from the source to the springs (defined as a Type I karst network in Field, 2004). For the BSSEA, it is unclear how much, if any, interconnectedness exists in the Recharge Zone. Therefore, a conservative assumption is that the system is represented by a straight conduit network of fractures, and atrazine reaching the springs from source areas has limited potential for degradation and dilution. The conservativeness of this assumption is apparent when considering that source areas further removed from Barton Springs have the potential for some dilution and degradation both at the surface and subsurface.

Atrazine residues derived from application sites that are located predominantly within the Contributing Zone are expected to travel via overland flow to surface streams to the Recharge Zone where infiltration from "losing" streams is likely to occur. Thus, exposures from these sources are expected to be lower relative to the "edge of field" exposures that originate in the Recharge Zone. Therefore, the "edge of field" approach is likely to over-estimate exposure originating in these areas (i.e., the Contributing Zone).

Given 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 both agricultural and non-agricultural uses sites are present in the Recharge Zone, the Contributing Zone, or both.

Based on the hydrogeologic configuration, pesticide use is modeled in the Contributing, Recharge, and Transition Zones. PRZM is used to model edge-of-field runoff concentrations, assuming that the edge of field runoff concentration is transported directly to the Barton Springs system via flow through the fracture system. This exposure pathway represents a simplification of how the species may be exposed to pesticides used in the BSSEA; however, it provides an upper bound of potential exposures and is reasonable given that there are no available models for predicting pesticide fate and transport in karst systems. This approach is considered to be conservative because the conceptual model of transport does not include degradation or dilution for use sites in close proximity to the springs.

Analysis of land cover data and usage information suggests that limited agriculture is present in the Contributing and Recharge Zones of the BSSEA. In order to address the potential for atrazine exposure from use on these sites, a suite of PRZM modeling scenarios was developed for the specific agronomic, soil, and climatic data available for the BSSEA. As noted above, the action area for the development of the Barton Springs scenarios is comprised of three hydrologic zones of the BSSEA (in order of importance): 1) the Recharge Zone, which consists of a fractured karstic geology; 2) the Contributing Zone, where surface runoff may flow to the Recharge Zone; and 3) the Transition Zone, which has a remote potential to contribute to the Recharge Zone (http://www.edwardsaquifer.net/intro.html). Although the Transition Zone is considered in this assessment, primary emphasis is given to the Recharge Zone with secondary emphasis on the Contributing Zone. No scenarios were parameterized based solely on the Transition 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.

As previously discussed in Section 3.1, label changes, including the establishment of setback restrictions on application of atrazine around wells and sinkholes, perennial and intermittent streams, lakes, and reservoirs, were implemented as part of the IRED/MOA. Specifically, the label changes restrict atrazine use within 50 feet of sinkholes, 66 feet of intermittent and perennial streams, and 200 feet of lakes and reservoirs. These restrictions are not quantitatively evaluated in this assessment.

As stated previously, this assessment assumes that the estimated environmental concentration (EEC) is derived from an edge-of-field exposure; thus, spray drift is not a factor in the predicted assessment. However, the assessment also conservatively assumes that the edge of field exposure could occur where surface runoff enters the subsurface fractures via sinkholes and/or other conduits to the karst fractures adjacent to the site of application. The net effect of the 50 foot setback would reduce loadings to the subsurface where the setback zone consists of a healthy vegetated zone. Alternatively, where the setback zone consists of bare soil, or a poorly maintained vegetative zone, little reduction

in runoff would be likely to occur. Current models do not estimate the effect of setbacks on load reduction for runoff, although it is documented in the literature that well vegetated setbacks can result in a substantial reduction in pesticide load to surface water (USDA, NRCS, 2000). Specifically for atrazine, data reported in the USDA study indicate that well vegetated setbacks reduce atrazine loading to surface water by as little as 11% and as much as 100% of total runoff without a buffer. It is expected that the presence of a well-vegetated 50 foot setback between the site of application of atrazine and sinkholes could result in loading reductions to the subsurface system. Therefore, the aquatic EECs presented in this assessment are likely to over-estimate exposure in areas with well-vegetated setbacks. However, given the lack of quantifiable estimates of load reduction and available data on the effectiveness of vegetated zones surrounding sinkholes, ranges of potential exposures cannot be estimated. The label changes also specify setback distances of 66 and 200 feet for atrazine applications surrounding intermittent/perennial streams and lakes/reservoirs, respectively. Typically, the influence of setback distances on spray drift loading would be evaluated using AgDrift to estimate the impact of the setback on the fraction of drift reaching a surface water body (U.S. EPA, 2004). However, as previously discussed, spray drift was not considered as a significant route of exposure in this assessment.

Overall, it is expected that well vegetated and maintained setbacks will reduce overall loading of atrazine from runoff estimates presented in this assessment. However, these reductions cannot be quantified and are unlikely to be uniform across the action area.

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 atrazine was 3 μ g/L in the Upper Spring, however, most detections of atrazine were below 1 μ g/L. Given the nature of the flow regime within the springs, it is unlikely that these sampling events have captured the peak exposures.

Evaluation of long-term trends in the monitoring data suggests that atrazine concentrations have increased since the 1980s; however, given that recent detections could be related to improved sampling techniques and analytical methods with lower detection limits, these observations may reflect an increase in monitoring intensity rather than a trend in atrazine exposures. Long-term trends in the analysis of spring water data suggest that atrazine occurrence has been sporadic. Atrazine was not detected in spring

samples prior to 2000; however, this may be due to changes in detection limits and an increased attention in recent years on more frequent sampling and selected sampling tied to specific runoff events (as noted above). More recent sampling suggests that longer-term concentrations in the four springs tend to be less than 1 μ g/L, with most of the longer-term detections closer to 0.1 μ g/L. Consideration of only detections yields an overall average concentration of 0.45 μ g/L (excluding non-detections). Table 3.2 presents a summary of the site-specific monitoring data.

Table 3.2. Summary of USGS Monitoring Data from the Four Springs Comprising Barton Springs

Spring	Range of Sample Dates	# of Samples	# of Detects	Frequency of Detection	Maximum Concentration (μg/L)	Minimum Concentration (μg/L)	Average Concentration (µg/L)
Main	1978- 2005	78	60	77%	0.555	0.042	0.070
Upper	2001- 2005	44	44	100%	3.190	0.018	0.164
Old Mill	2001- 2005	12	12	100%	0.063	0.007	0.015
Eliza	2000- 2005	15	15	100%	0.112	0.007	0.036

Several degradates of atrazine were detected in the samples collected from the springs including deethyl-atrazine, 2-hydroxyatrazine, chlorodiamino-s-triazine, and deisopropylatrazine. All four degradates were detected in spring water at concentrations below those found for atrazine. In general, the concentrations of the degradates were 2 to 3 times less than atrazine during low flow (base flow) periods, while atrazine concentrations were 10 to 15 times higher than deethyl-atrazine during high flow (storm derived runoff events).

Analysis of the stream data suggests that trends similar to those seen in the spring data occurr in surface streams. Similar to the spring detections, most of the atrazine detections in surface streams have occurred since 2000. The highest detection of atrazine in the streams is 4.39 μ g/L in Slaughter Creek in 2005. Figure 3.5 shows the location of stream samples within the BSSEA.

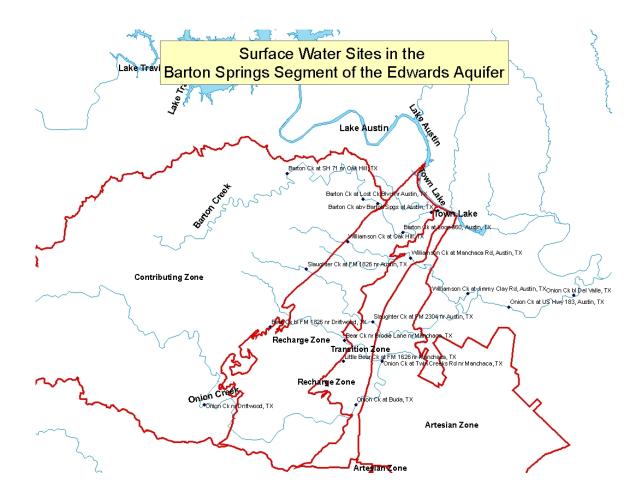


Figure 3.5. Location of Surface Water Sites within the Barton Springs Segment

Statewide surface water data for atrazine collected by the USGS NAWQA Program between 1993 and 2003 shows that atrazine was detected in 792 out of 866 samples (92% detection frequency). The highest detected concentration of atrazine in all of the NAWQA samples was 20 μ g/L in 1994 and 1995 from two sites in the Trinity River Study Unit in Ellis and Navarro counties (station ID numbers 321313096415201 and 321017096420099) that are outside of the action area for this assessment.

Analysis of the well data from the USGS collected between 2000 and 2005 suggests that atrazine was detected in groundwater from five locations within the BSSEA. In general, with the exception of a single sample analyzed from well # YD-58-34-617 in 2002 at 0.192 μ g/L, all atrazine detections were below 0.1 μ g/L. More commonly detected were the principal degradates of atrazine including deethyl-atrazine, 2-hydroxyatrazine, chlorodiamino-s-triazine, and deisopropylatrazine, none of which are included in this assessment (rationale for this exclusion is provided in the atrazine 2003 IRED and Section 2.2 of this assessment). Overall, the low frequency and magnitude of detection of atrazine suggests that its occurrence in baseflow may be a minimal source of exposure at Barton Springs. Figure 3.6 presents the location of the groundwater wells within the BSSEA.

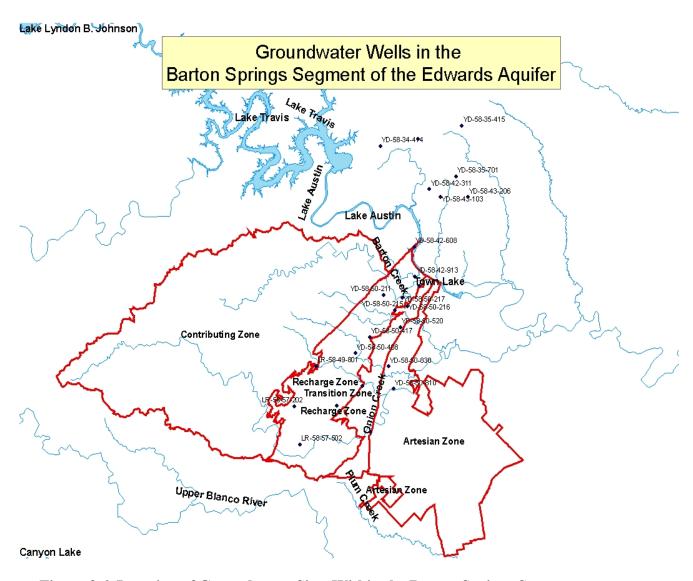


Figure 3.6. Location of Groundwater Sites Within the Barton Springs Segment

Overall, the monitoring data provided by the USGS indicate relatively consistent low-level concentrations of atrazine 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.

A key component of the total load reaching Barton Springs is represented by base flow. For this assessment, a reasonably conservative estimate of atrazine load arriving via base flow is represented by the average concentration in all springs of 0.45 $\mu g/L$. This estimation is conservative because it excludes all non-detections of atrazine and includes concentrations associated with storm events. The USGS has estimated base flow concentrations of atrazine to be less than 0.1 $\mu g/L$ (Mahler, 2005a). Information on the average flow rate through the spring system was evaluated. As noted above, Hauwert et al. (2004) estimated an average flow rate through the entire system of 53 cfs, while Mahler (2005a) reported an average of 56 cfs. Hydrograph data for Barton Springs from the USGS (Figure 3.7) yields an average flow of 62 cfs.

Mean Flow (cfs) for Barton Springs

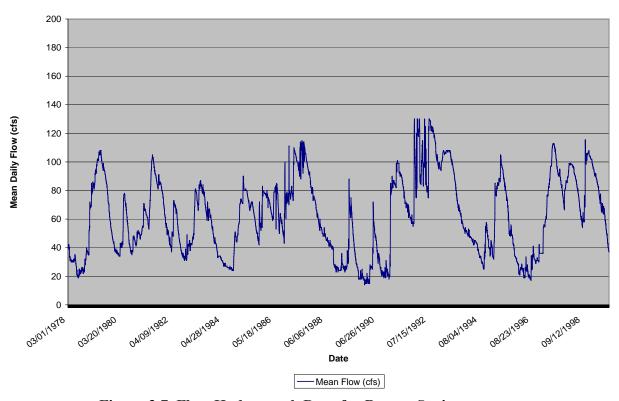


Figure 3.7. Flow Hydrograph Data for Barton Springs

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.

Loading of atrazine to the system via base flow is approximated using available monitoring data. The average concentration from the available USGS monitoring data from the four springs of $0.45~\mu g/L$ is added to the daily edge of field EEC. The volume associated with the base flow is accounted for by using the long-term average flow rate from the USGS hydrograph data for Barton Springs of 63 cfs. PRZM reports a daily flow at the edge of field in units of centimeters representing the depth of water leaving the 10 hectare field. An equivalent volume is calculated by multiplying the edge of field runoff depth by the total area of the 10 hectare field in centimeters. In order to use the average flow rate from the hydrograph data, the allocation of 63 cfs was back-calculated to the entire BSSEA drainage area of 900 square kilometers. This estimation yields an approximate additional flow through the Recharge Zone of 1,000,000,000 cm³/day (PRZM reports this as the depth of water running off the entire 10 hectare watershed, or 1 cm) which is added to the daily edge of field estimate.

Peak atrazine concentrations, as well as rolling time-weighted averages of 14 days, 21 days, 30 days, 60 days, and 90 days are calculated for comparison with various ecotoxicity endpoints (including aquatic community-level threshold concentrations) for atrazine.

3.2.5.1 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 2003 atrazine IRED (U.S. EPA, 2003a) and the cumulative triazine risk assessment (U.S. EPA, 2006a); 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/oppsrrd1/REDs/atrazine_ired.pdf

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.

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

Fate Property	Value	MRID (or source)	
Molecular Weight	215.7	MRID 41379803	
Henry's constant	2.58 x10 ⁻⁹	MRID 41379803	
Vapor Pressure	$3 \times 10^{-7} \text{ atm-m}^3/\text{mol}$	MRID 41379803	
Solubility in Water	33 mg/l	MRID 41379803	
Photolysis in Water	335 days	MRID 42089904	
Aerobic Soil Metabolism Half-lives	152 days	MRID 40431301 MRID 40629303 MRID 42089906	
Hydrolysis	Stable	MRID 40431319	
Aerobic Aquatic Metabolism (water column)	304 days	2x aerobic soil metabolism rate constant	

Fate Property	Value	MRID (or source)
Anaerobic Aquatic Metabolism (benthic)	608 days	MRID 40431323
		MRID 40431324
		MRID 41257901
Vac	00.701/~	MRID 41257902
Koc	88.78 ml/g	MRID 41257904
		MRID 41257905
		MRID 41257906
Application Efficiency	95 % for aerial 99 % for ground	default value ²
Spray Drift Fraction ¹	5 % for aerial 1 % for ground	default value ²

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

Unlike the Agency's standard ecological risk assessment methodology that relies on EECs derived using an EXAMS standard water body, edge of field concentrations were predicted for those use sites expected to reside within the action area. It is expected that infiltration directly into the fractured limestone represents the most direct route of exposure and is likely to yield the highest EECs. In this instance, PRZM alone was used to estimate the edge of field concentration. Unlike the typical approach used to estimate exposure using the EXAMS water body described above, the PRZM output from the *.zts file (daily time series data) associated with each scenario modeled was used. In this instance, the *.zts file provides daily estimates of atrazine exposures. The PE4v01.pl script was modified to provide time series output (TSER in PRZM) as opposed to the standard cumulative output (TCUM in PRZM) of runoff volume, mass transported with runoff, infiltration volume, mass transported with infiltration, eroded sediment, and mass of pesticide transported with sediment. The runoff volume and runoff mass were extracted and converted to a runoff concentration.

The standard approach for conducting ecological risk assessment assumes that 100% of the 10-hectare watershed is covered by the relevant use. This approach also assumes that the standard water body is adjacent to the edge of the field. In this assessment, the majority of the use sites, with the exception of residential uses, are either sporadically present within the action area or are predominant further south and west of the spring systems. Therefore, it is unlikely that edge of field concentrations for these use sites are equivalent to the standard assumption that the water body is receiving runoff from a small watershed that is adjacent to a field that is 100% cropped and treated. Although travel through the fractures is likely to be direct, there is also likely to be non-impacted fracture flow arriving simultaneously at the springs from the same runoff event. In order to account for this, an adjustment factor is applied to each of the use sites located outside the immediate area of the springs. These factors are based on an assessment of recent land cover data with specific cover types that correlate with the use sites. A summary of the land cover-based adjustment factors is presented in Table 3.4.

^{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

Table 3.4. Land Cover Adjustment Factors for the Action Area in the Barton Springs Segment of the Edwards Aquifer (BSSEA)

Scenario Modeled	Land Cover Adjustment Factor
Residential	70% of action area in vicinity of Barton Springs (% decreases with distance from spring system)
Impervious	30% of action area in vicinity of Barton Springs (% decreases with distance from spring system)
Turf	100% of action area in vicinity of Barton Springs and 28% of treatable golf course (based on limited occurrence of golf courses within action area)
Rights-of-Way	10% of action area in vicinity of Barton Springs (based on density near springs; % decreases with distance from spring system)
Fallow/Idle land	5% of action area in vicinity of Barton Springs (estimated from land cover of entire action area; majority of fallow/idle land is located at the southern and western edge of action area)

The edge of field concentrations are post-processed (see Appendix E for details) in order to provide durations of exposure. First, daily concentrations were calculated using the time series data described above for all 30 years of model output. Then, peak, 14-day, 21-day, 30-day, 60-day, and 90-day average concentrations were calculated across the entire 30 years of data. In order to match the standard PRZM/EXAMS output, the 30 years of daily and rolling averages were separated into individual years and the maximum value for each of the 30 years was calculated for both peak and rolling averages. Finally, the 30 years of maximum values were ranked and the 90th percentile from the rankings was selected as the final EEC for use in risk estimation.

The time series output generated by PRZM provides individual runoff events and does not capture the influence of base flow in the fracture zone. In order to account for the influence of base flow, the average of all monitoring data detections (excluded non-detections) was added to the time series output. For atrazine, the estimated base flow value of $0.45~\mu g/L$ is considered to be conservative because it includes recently analyzed storm runoff data and excludes all non-detections. In addition, the flow associated with the base flow exposures was estimated using USGS hydrograph data for Barton Springs and was also added into the exposure estimate. Addition of base flow exposure to the estimated exposure was intended to provide an upper bound on base flow and account for the limitations in the sample frequency that may not completely represent base flow.

The $0.45~\mu g/L$ estimate of base flow concentration is considered to be a conservative estimate for several reasons. First, the estimate is based on all detections from the site-specific USGS monitoring data and includes samples collected during high flow events. Second, the value excludes all non-detections. These first two points are important because the USGS estimates that the actual base flow concentration of atrazine from the non-runoff driven sampling is less than $0.1~\mu g/L$ (Mahler, 2005a).

Calculating an annual average concentration from the PRZM generated edge of field concentrations without the added 0.45 $\mu g/L$ value yields an annual average concentration of roughly 0.15 $\mu g/L$ that is consistent with the USGS estimate. Therefore, addition of the 0.45 $\mu g/L$ atrazine concentration is assumed to be conservative and protective. The analysis indicates that long term averaging of the PRZM edge of field output provides estimated exposures consistent (within a factor of two to three times) with the estimate of base flow predicted by the USGS from the site specific monitoring data. Therefore, the approach taken in this assessment is considered to be reasonable and protective. The approach is reasonable given that the monitoring data, though of high quality and targeted to Barton Springs, is not considered to be of sufficient robustness to provide an upper bound on both peak and longer-term exposures due to the limited sampling frequency in the study design. This approach is protective because use of the 0.45 $\mu g/L$ concentration as a base flow value is greater than any estimate of base flow available based on a limited monitoring data set.

The calculations described above were completed by a straight comparison of runoff mass divided by runoff volume to get a daily estimated EEC. However, a second step was included to evaluate the influence of infiltrating water on the overall exposure. This additional step was included to mimic the influence of infiltrating water into the fractured limestone of the Recharge Zone and to account for base flow concentrations associated with some amount of infiltrating water not accounted for in the runoff edge of field EEC. The additional volume of water predicted by PRZM to leach out of the bottom of the soil profile was added back into the edge of field concentration. This additional step accounts for recharge to the fractured limestone that is likely to occur from infiltration of runoff water through fractures/sinkholes and direct infiltration throughout the soil profile.

3.2.6 Individual Scenario Results

A total of four scenarios were developed for this assessment, including residential, turf, rights-of-way, and fallow/idle land. Of these, two scenarios are used in tandem with the impervious scenario (residential and rights-of-way). Discussions with local experts (Markwardt, 2006; Ward, 2006; Mason, 2006) suggest that atrazine is never used in the rights-of-way scenario within the action area. However, the rights-of-way scenario was modeled, given its potential as a use site in the action area. It should be noted, however, that the predicted EECs associated with rights-of-way are less relevant to actual exposures than the remaining scenarios and are presented for a qualitative comparison.

Model inputs were selected for atrazine using the most recent data available from the atrazine IRED (U.S. EPA, 2003a) and the triazine cumulative assessment (U.S. EPA,

2006a). A discussion of each assessed exposure scenario and a summary of the results for each is provided below. Copies of the model input files along with the stepwise approach for processing model output are provided in Appendix E.

3.2.6.1 Residential

The residential exposure scenario represents two scenarios modeled in tandem. The first scenario is intended to reflect runoff and erosion from a typical ¼ acre lot and reflects a typical urban/suburban use site with homeowner and professional applications. The residential lot scenario was developed using local soil information and a USDA runoff curve number developed specifically for ¼ acre lots (USDA, 1986). In order to justify the assumption of ¼ acre lot as a typical exposure scenario, publicly available data was reviewed from the United States Census (Census). Specifically, data from 2003 from the American Housing Survey (AHS) available at the following website was reviewed.

http://www.census.gov/hhes/www/housing/ahs

Initially, the data for all suburban homes available nationally was reviewed. It is assumed that most pesticide applications, particularly for herbicides, occur in suburban settings. In order to test the assumption of the ¼ acre lot as the best representation, AHS data for suburban homes that list total number of houses by lot size and by square footage of house (see Table 1C-3 at the AHS website above) was reviewed. With a total of 45,552,000 total units reported nationally for all suburban areas, 12,368,000 units (the largest class at 27%) were located on lots between 1/8 acre and ¼ acre, while 9,339,000 units (the second largest class at 21%) were located on lots between ¼ acre and ½ acre. Overall, the median lot size was 0.37 acre. This analysis suggests that the ¼ acre lot is a reasonable approximation of suburban pesticide use. The selection of the ¼ acre lot was an assumed to provide an estimate of potential exposure in an urban/suburban scenario where it is expected that most herbicide use will occur. It is believed that this representation provides a reasonable estimate of typical uses in an urban/suburban watershed particularly as it relates to the City of Austin and its rapidly developing outskirts.

The second scenario was developed to represent general impervious surfaces expected to be present in an urban/suburban watershed. Examples of representative impervious surfaces include roads, parking lots, and buildings. These surfaces are distinct from the impervious surfaces inherent in the ¼ acre lot (driveways, sidewalk, and house). The impervious surface uses a high-end curve number (98 out of a maximum of 100) to mimic the runoff expected from these surfaces. Using these in tandem allows for a weighting of the runoff potential of both surface types within a residential watershed that is different from the standard agricultural watershed that assumes uniform land cover. Figure 3.8 presents the conceptual model of the paired impervious/pervious scenario approach.

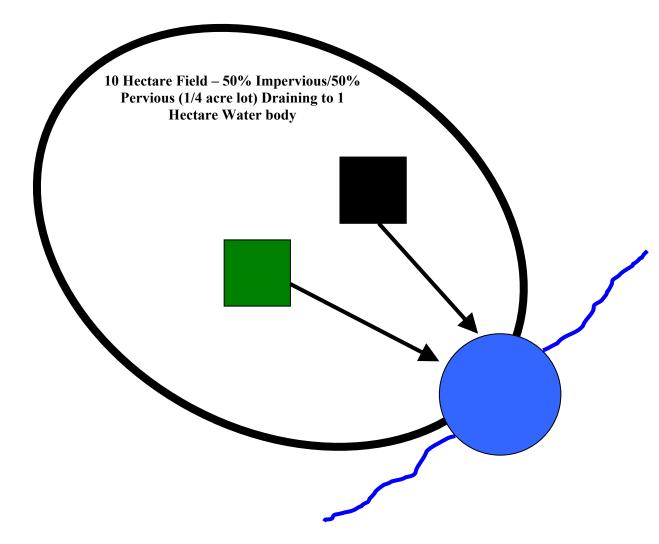


Figure 3.8. Conceptual Model of Paired Residential/Impervious Scenarios (green square represents ¼ acre lot while black square represents impervious surface scenario. Ratio of pervious to impervious surface based on best available land cover data)

For edge of field EECs, the output is weighted based on the percentage of impervious surface present in the action area and by the percentage of the ¼ acre lot treated. For this assessment, it is assumed that 30% of the action area in the vicinity of the spring system is impervious (see Figure 3.9). This assumption is reasonable given the density of residential development surrounding the springs. Outlying areas are likely to have lower percentages of impervious surfaces; however, residential areas in close proximity to the springs are likely to be most representative of the expected edge of field concentrations. Additional analysis of the impact of alternative assumptions for percent impervious

surface, overspray, and percentage of lot treated is included in Section 3.2.7. Because of the unique nature of the karst system, it is assumed that no direct spray drift will reach the spring system (personal communication with Elizabeth McVeety, pesticide applicator at Zilker Park, April 21, 2006). However, it is likely that some overspray may reach impervious surfaces in the residential setting. In order to account for overspray, the impervious surface was modeled using three separate assumptions. For the purposes of risk assessment, it is assumed that 1% of the application rate could reach the impervious surfaces surrounding each residential lot. This amount of overspray is not based on any empirical data (studies of this type were not identified); however, the assumption seems reasonable given the principal drift assumption for ground spray in ecological risk assessments is 1%. In order to test the assumption of 1% overspray and address the uncertainty associated with the lack of data for overspray, two alternate scenarios were modeled. The impervious surface was modeled with 0% overspray and 10% overspray to provide a lower bound (0% overspray) and an upper bound (10% overspray) on the 1% assumption. The results of these alternate modeling exercises are discussed more fully in Section 3.2.7 of this assessment.

In this exercise, it is also assumed that that 50% of the ¼ acre lot is treated with atrazine. This assumption was based on data from the AHS website and from professional judgment about typical features and the percentage of a typical lot those features might require. For example, the AHS survey data reports that of a total of 43,328,000 single detached homes in suburban areas, 10,124,000 (the largest group at 23%) were between 1,500 and 2,000 square feet, while 7,255,000 (the third largest group at 17%) were between 2,000 and 2,500 square feet, and 9,513,000 (the second largest group at 22%) were between 1,000 and 1,500 square feet. From this data, it is assumed that a typical home is 2,000 square feet with a 1,000 square foot footprint. Lower sized houses less than 1,500 square feet are more likely to represent single floor structures; thus, the 1,000 square foot estimate for a house footprint is considered to be reasonable.

In addition to the footprint of the typical house, it is also assumed that a typical house has a driveway of approximately 25 by 30 feet or 750 square feet and roughly 250 square feet of sidewalk. A typical suburban home was also assumed to have roughly 300 square feet of deck space and 900 square feet of garage. Finally, it was assumed that a substantial portion of the typical home would be landscaped with an estimate of 2,000 square feet. All of the previous estimates are based on professional judgment and are not derived from the AHS data. All of these areas are assumed to not be treated with a turf herbicide, resulting in a total area not treated with atrazine of 5,200 square feet. Taking a total ¼ acre lot size of 10,890 square feet and subtracting the untreated square footage yields a total remaining area of 5,690, or roughly 50% of the total lot that could be potentially treated.

The first assumption may result in an underestimation of exposure, given that more overspray of impervious surfaces is possible. The impact of this assumption is tested and characterized in Section 3.2.7. Note that this scenario represents general impervious surfaces within a watershed not part of the ¼ acre lot and includes roads, parking lots, and buildings among others where overspray from residential lots is expected to be

minimal. The ¼ acre lot, by comparison, was developed with a curve number reflective of the fact that the lot is covered with both pervious surfaces (grass and landscaped gardens) and impervious surfaces (driveways, sidewalks, and buildings). In this case, the assumption that 50% of the lot is treated likely overestimates the amount of landscaped area treated, but underestimates unintentional overspray of driveways and sidewalks. The impact of this assumption is also evaluated in Section 3.2.7. Overall, these are simplifying assumptions that are likely to provide a reasonable high-end estimate of exposure, given the limitations of the modeling approach.

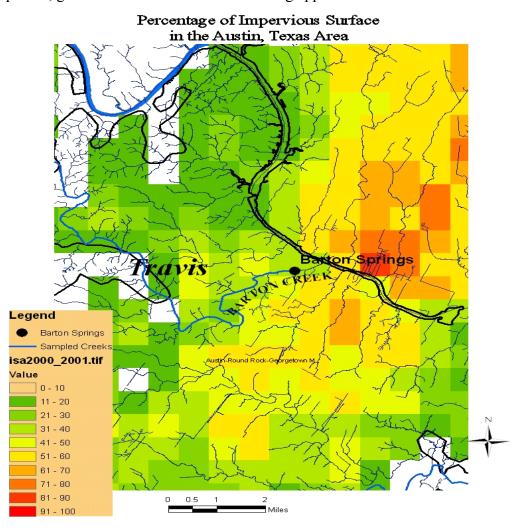


Figure 3.9. Percentage of Impervious Surface Coverage in Vicinity of Barton Springs

The combined edge of field concentrations are estimated using the *.zts output from PRZM as described above. In this paired scenario approach, the *.zts output from both the impervious and residential scenarios are weighted and added together to provide an

overall estimate of exposure. Non-detects in the weighted output were converted to $0.45 \,\mu g/L$ in order to capture the potential influence of base flow (described in more detail above).

Two categories of formulations are currently registered for atrazine use on residential sites, including granular and liquid formulations (wettable powder dry flowables). Both formulations were modeled separately because application rates are different (2 lbs/acre for granular and 1 lb/acre for liquid) and the standard assumption for modeling granular formulations is different from liquid formulations. Granular formulations are typically modeled as soil applied (CAM is set to 8 with a minimized incorporation depth of 1 cm) and 0% spray drift, as compared with a foliar application (CAM isset to 2 with a 4-cm depth of incorporation), which assumes the standard spray drift of 1% for ground applications. However, because spray drift is not assumed to contribute to the loadings in the spring and some overspray is expected to impervious surfaces, both residential scenarios (liquid and granular) were modeled assuming that 1% of the application rate is applied to the impervious surface.

Figure 3.10 graphically presents the runoff only time series output for the edge of field concentrations predicted for the granular application of atrazine to the paired residential/impervious scenario, assuming an overspray of 1% of the application rate to the impervious surface.

Residential (Granular) Edge of Field Concentration (ppb)

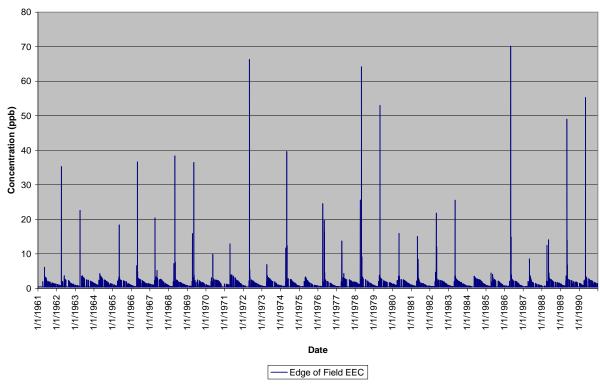


Figure 3.10. Representative Time Series Output from Paired Residential/Impervious PRZM Scenario for Granular Applications

3.2.6.2 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). For the Barton Springs assessment, the turf scenario is intended to represent golf course turf and recreational fields because residential uses are captured in the residential scenario (standard turf scenarios are typically used to represent both golf course, recreational, and residential uses). The standard approach for conducting ecological risk assessments assumes 100% of the 10-hectare watershed is covered by the relevant use. This approach also assumes that the receptor (EXAMS standard water body) is adjacent to the edge of the field. In this assessment, with the exception of the residential use sites described above, most of the other potential atrazine use sites are not adjacent to the receptor. Although travel through the fractures is likely to be direct, there is also likely to be nonimpacted fracture flow arriving simultaneously at the springs. In order to account for this, an adjustment factor was applied to each of the non-residential use sites. For turf, it was assumed that 100% of the watershed feeding the fractures is represented by golf course turf, but that the percentage of the golf course expected to be treated in this case is represented by the golf course adjustment factor of 28%. This seems reasonable given that the land cover analysis indicates that only a few golf courses are present within the action area. As with the residential scenario, a base flow concentration of 0.45 µg/L was added to the overall exposure.

Similar to the residential scenario, two categories of formulations are registered for atrazine use on turf sites. These are granular and liquid formulations (wettable powder dry flowables). Both formulations were modeled separately because application rates are different (2 lbs/acre for granular and 1 lbs/acre for liquid) and the standard assumption for modeling granular formulations is different from liquid formulations. Granular formulations are typically modeled as soil applied (CAM is set to 8 with a minimized incorporation depth of 1 cm) and 0% spray drift, as compared with a foliar application (CAM is set to 2 with a 4-cm depth of incorporation), which assumes the standard 1% spray drift for ground applications.

3.2.6.3 Fallow/Idle Land

The fallow/idle land scenario represents the only agricultural use present within in the action area. Generally, this scenario conceptually represents the potential application of atrazine to fallow lands under the Conservation Reserve Program (CRP) as most of the available information suggests that no agricultural use of atrazine occurs in the action area. It is also expected, given the available usage information, that fallow/idle land represents a minor use in the action area, although there are limitations associated with this analysis. Regardless, the scenario was included in the assessment to address all potential atrazine uses.

This scenario was developed similar to the standard PRZM scenarios. The bulk of this use pattern is located south and west of the receptor site; therefore, a land cover adjustment factor was applied to the modeled output. Although travel through the fractures is likely to be direct, non-impacted fracture flow is likely to arrive simultaneously at the springs, especially given that most of this use site is located within the Contributing Zone. To account for this, an adjustment factor was applied for each of the non-residential use sites. For the fallow/idle land scenario, this factor represents the percentage of the land cover within the action area that is in fallow/idle land. As shown in Table 3.4, the adjustment factor for fallow/idle land is 5%. As with the residential scenario, a base flow concentration of 0.45 μ g/L was added to the overall exposure.

3.2.6.4 Rights-of-Way

The rights-of-way scenario represents a vegetated buffer strip where atrazine could be applied adjacent to a water body. The vegetative strip as modeled is intended to represent treated buffers along roadways, railroad lines, and utility rights-of-way. Each of these sites is conceptualized as a naturally vegetated strip that runs linearly adjacent to a sensitive water body. Figure 3.11 presents the conceptual model of this scenario relative to a roadway, although a similar layout would be expected for rail and power rights-of-way. It is expected that the density of roads, railroads, and utility rights-of-way define the density of rights-of-way use within the watershed where pesticides may be applied. Land cover data was used to define this percentage. For this assessment, it is assumed that the maximum density of treated rights-of-way within the action area is 10%, which is expected to represent a slight over-estimation.

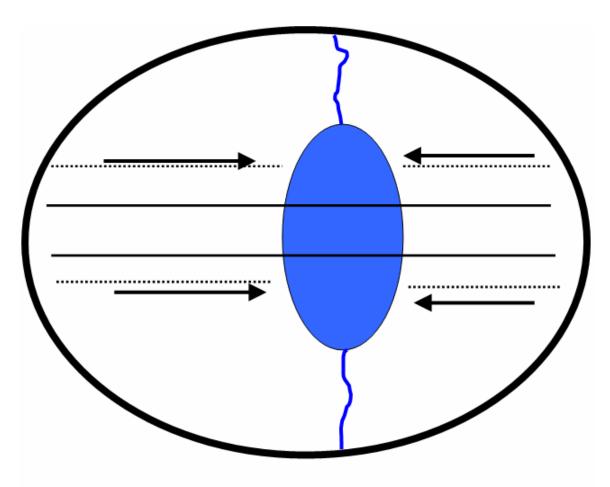


Figure 3.11. Conceptual Model of Rights-of-Way Scenario

In addition, the potential impact of impervious surfaces within the treated area was addressed to estimate its influence on overall runoff and exposure. For the roadway rights-of-way use, it is assumed that there is an equal amount of impervious and pervious surface within the treatment area, but that the atrazine treatment is likely to be restricted to the pervious portion of the rights-of-way. As with the residential scenario, this assumption accounts for a potential overspray to the impervious surface of 1%. Conversations with local experts (Markwardt, 2006; Ward, 2006; Mason, 2006) suggest that the rights-of-way scenario is likely to be conservative, given that atrazine is not typically used (glyphosate is reportedly the herbicide of choice in Hays and Travis counties) and treatment zones are typically one to four feet wide. As with the residential scenario, the impact of 0% and 10% over spray of the impervious surface on rights-of-way is characterized in Section 3.2.7 of this assessment.

As with the residential scenario, the edge of field EECs for both the runoff only and runoff plus infiltration scenarios were estimated. Similar to the residential scenario, a base flow concentration of $0.45 \,\mu g/L$ was added to the overall exposure.

Table 3.5 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 presented in Table 3.5 are used to derive risk quotients, which are presented as part of the Risk Characterization in Section 5

Table 3.5. Summary of PRZM Output EECs for all Modeled Scenarios (Edge of Field Concentrations with Base Flow Incorporated).

			90 th Perce	entile of 30 Ye	ars of Out	put				
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)
Residential Granular ¹	2	2	April 1	41.2	3.5	2.5	1.9	1.2	1.0	0.6
Residential Liquid ¹	1	2	April 1	26.6	2.5	1.8	1.5	1.0	0.8	0.5
Right-of- Way ¹	1	1	June 1	6.2	1.1	0.9	0.8	0.6	0.6	0.5
Fallow/Idle land	2.25	1	November 1	7.5	1.0	0.8	0.7	0.6	0.6	0.5
Turf – Granular	2	2	April 1	22.4	2.0	1.5	1.2	0.8	0.7	0.5
Turf - Liquid	1	2	April 1	16.2	1.7	1.3	1.0	0.7	0.6	0.5

In general, the exposure assessment yields modeled peak exposure estimates that are two to ten times higher than those seen in monitoring data, while the annual average concentrations are consistent with those seen in monitoring. The 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.

3.2.7 Characterization

Reported use information provides a sense of the actual use on sites similar to those assessed including fallow/idle land. In this instance, the data for fallow/idle land suggests that the 90th percentile application rate is similar to the maximum labeled use rate while the typical use application rate (equivalent to the average of all reported applications) is roughly half the labeled maximum rate used in this assessment. Table 3.6 summarizes the typical and 90th percentile rates and number of applications relative to those used in this assessment. If it were assumed that this pattern holds true for all uses (agricultural and non-agricultural), then modeling with the typical application rates would yield predicted exposures that are roughly half of those presented in Table 3.5.

Table 3.6. Comparison of Maximum, Typical, and 90th Percentile Label Rates and Number of Applications

Scenario	Maximum Application Rate (lbs/acre)	Maximum Number of Applications	90 th Percentile Application Rate (lbs/acre)	90 th Percentile Number of Applications	Typical Application Rate (lbs/acre)	Typical Number of Applications
Fallow/Idle land	2.25	1	2	1	0.9	1

In order to account for the variability in overspray, the residential scenario was modeled assuming two alternate scenarios of 0% and 10% overspray to impervious surfaces. The alternate assumptions are intended to provide a bound on the 1% assumption. Because both the residential and rights-of-way scenarios were modeled using the paired pervious/impervious approach, the alternate scenarios were modeled for both scenarios (residential was modeled for both granular and liquid formulations). The conservativeness of these assumptions is unknown, given a lack of data on this phenomenon. However, given that the impervious scenario is intended to represent nontarget surfaces such as roads, parking lots and buildings, it is seems reasonable to assume that 10% overspray is an over-estimation of what would likely occur to these off-site areas, while 0% may be an under-estimation.

In order to model the overspray, the binding coefficient was set to zero and the aerobic soil metabolism half-life was set to stable in lieu of actual data for the impervious scenario. It is assumed that non-binding would occur on these surfaces and that limited degradation would occur. The percentage overspray was then multiplied by the total application rate to yield an effective application rate for the overspray to impervious

surfaces. This analysis yielded an application rate on the impervious surface of 0.2 lbs/acre (0.23 kg/ha) for 10% overspray and 0.02 lbs/acre (0.023 kg/ha) for 1% overspray.

Comparison of the resulting EECs indicates that with 10% overspray, the overall increase in peak EECs is roughly 30%, while the longer-term EECs are increased by nearly 50%. For the 1% overspray assumption, there is very little increase in overall EECs for both peak and average EECs, as compared to the 0% overspray assumption. This is not unexpected, given the increased runoff, lack of binding, and lack of degradation being modeled. Without actual data on these processes, it is impossible to determine whether these exposures reflect reality, especially given that none of the monitoring data indicate concentrations approaching any of these EECs with or without overspray. The overspray comparison is presented in Table 3.7.

Other assumptions that can have a significant impact on the overall predicted EECs include the percentage of impervious surface and the percentage of ½ acre lot that is treated. In both instances, the relationship between the assumption and the predicted EEC is linear. The assumed action area impervious surface percentage of 30% in the vicinity of the Barton Springs decreases dramatically further south and west from the salamander's habitat. It is apparent from the available data that this value decreases to less than 10% the further south and west from the springs. The impact of this assumption was evaluated by readjusting the output to reflect the impact of a 10% impervious cover assumption on predicted exposures. In general, peak and longer-term average concentrations are generally doubled as the percentage of impervious decreases. The comparison of this analysis is presented in Table 3.8. This is likely due to the increase in treated area contributing more pesticide mass and an increase in the impervious surface, which yields greater amounts of non-contaminated runoff.

This impact of a decrease in impervious surface will hold only with the assumption of limited overspray. This assumption was explored by comparing the impact of the change in percentage of impervious surface on the 10% overspray scenario discussed above. In this case, peak EECs increase by roughly 50% while the averages are only slightly increased. The comparison of this analysis is presented in Table 3.9.

Finally, in this assessment it is assumed that 50% of the ¼ acre lot is treated. In order to test the significance of this assumption, the exposure scenario was re-run using a different assumption of 10% treatment of the ¼ acre lot. As expected, peak EECs are reduced by roughly a factor of five, while the longer term exposures are reduced by a factor of two to three times. The results of this comparison are presented in Table 3.10.

Table 3.7. Comparison of Residential and Rights-of-Way EECs Assuming Variable Percentages of Overspray (0, 1, and 10%)

			90 th Per	rcentile of 30	Years of Ou	ıtput				
Use Site	Application Rate (lbs/acre)	Number of Applications (interval)	First Application Date	Peak 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)
Residential - 1% Overspray ¹	2.0	2 (30 days)	April 1	41.2	3.5	2.5	1.9	1.2	1.0	0.6
Residential – No Overspray ¹	2.0	2 (30 days)	April 1	40.0	3.3	2.4	1.8	1.1	0.9	0.6
Residential - 10% Overspray ¹	2.0	2 (30 days)	April 1	51.7	5.6	4.1	3.4	2.3	1.7	0.9
Rights-of- Way - 1% Overspray	1.0	1	June 1	6.2	1.1	0.9	0.8	0.6	0.6	0.5
Rights-of- Way – No Overspray	1.0	1	June 1	5.9	1.1	0.9	0.8	0.6	0.6	0.5
Rights-of Way - 10% Overspray	1.0	1	June 1	9.5	1.9	1.7	1.5	1.1	0.9	0.6

^{1 –} Only the granular application was tested for characterization

Table 3.8. Comparison of Residential EECs (granular) with 1% Over Spray and Variable Percentages of Impervious Surface (10 and 30%)

	90 th Percentile of 30 Years of Output									
Use Site	Applicati on Rate (lbs/acre)	Number of Applications (interval)	First Application Date	Peak 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)
Residential - 30% Impervious	1.0	2 (30 days)	April 1	41.2	3.5	2.5	1.9	1.2	1.0	0.6
Residential – 10% Impervious	1.0	2 (30 days)	April 1	62.8	5.0	3.5	2.6	1.6	1.2	0.6

Table 3.9. Comparison of Residential EECs (granular) with 10% Overspray and Variable Percentages of Impervious Surface (10 and 30%)

			90 th Per	centile of 30	Years of Ou	itput				
Use Site	Applicati on Rate (lbs/acre)	Number of Applications (interval)	First Application Date	Peak 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)
Residential - 30% Impervious	1.0	2 (30 days)	April 1	51.7	5.6	4.1	3.4	2.3	1.7	0.9
Residential – 10% Impervious	1.0	2 (30 days)	April 1	67.1	6.0	4.2	3.2	2.0	1.5	0.7

Table 3.10. Comparison of Residential EECs (granular) Assuming Various Percentages of Treated ¼ Acre Lot (10 and 50%)

			90 th Per	centile of 30	Years of Ou	tput				
Use Site	Application Rate (lbs/acre)	Number of Applications (interval)	First Application Date	Peak 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)
Residential – 50% Treated	1.0	2 (30 days)	April 1	41.2	3.5	2.5	1.9	1.2	1.0	0.6
Residential – 10% Treated	1.0	2 (30 days)	April 1	9.5	1.3	1.0	0.9	0.7	0.6	0.5

4. Effects Assessment

This assessment evaluates the potential for atrazine to adversely affect the Barton Springs salamander. As previously discussed in Section 2.7, 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 vertebrates, including fish, which are generally used as a surrogate for amphibians, as well as available salamander toxicity data from the open literature. 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 atrazine. In addition to registrant-submitted and open literature toxicity information, indirect effects to Barton Springs salamanders, via impacts to aquatic plant community structure and function are also evaluated based on communitylevel threshold concentrations. Other sources of information, including use of the acute probit dose response relationship to establish the probability of an individual effect and reviews of the Ecological Incident Information System (EIIS), are conducted to further refine the characterization of potential ecological effects associated with exposure to atrazine. A summary of the available freshwater ecotoxicity information, the community-level endpoints, use of the probit dose response relationship, and the incident information for atrazine are provided in Sections 4.1 through 4.4, respectively.

With respect to atrazine degradates, including hydroxyatrazine (HA), deethylatrazine (DEA), deisopropylatrazine (DIA), and diaminochloroatrazine (DACT), it is assumed that they are of lesser toxicity as compared to the parent compound. Comparison of available toxicity information for the degradates of atrazine indicates lesser aquatic toxicity than the parent for freshwater fish, invertebrates, and aquatic plants. Specifically, the available degradate toxicity data for HA indicate that it is not toxic to freshwater fish and invertebrates at the limit of its solubility in water. In addition, available aquatic plant degradate toxicity data for HA, DEA, DIA, and DACT report nondefinitive EC₅₀ values (i.e., 50% effect was not observed at the highest test concentrations) at concentrations that are 700 to 10,000 times higher than the lowest reported aquatic plant EC₅₀ value for parent atrazine. Therefore, given the lesser toxicity of the degradates, as compared to the parent, concentrations of the atrazine degradates are not assessed, and the focus of this assessment is limited to parent atrazine. The available information also indicates that aquatic organisms are more sensitive to the technical grade (TGAI) than the formulated products of atrazine; therefore, the focus of this assessment is on the TGAI. A detailed summary of the available ecotoxicity information for all of the atrazine degradates and formulated products is presented in Appendix A.

4.1 Evaluation of Aquatic Ecotoxicity Studies

Toxicity endpoints are established based on data generated from guideline studies submitted by the registrant, and from open literature studies that meet the criteria for inclusion into the ECOTOX database maintained by EPA/Office of Research and Development (ORD) (U.S. EPA, 2004). Open literature data presented in this assessment were obtained from the 2003 atrazine IRED (U.S. EPA, 2003a) as well as information obtained on February 16, 2006. The February 2006 ECOTOX search included all open literature data for atrazine (i.e., pre- and post-IRED). In order to be included in the ECOTOX database, papers must meet the following minimum criteria:

- (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. Based on the results of the 2003 IRED for atrazine, potential adverse effects on sensitive aquatic plants and non-target aquatic organisms including their populations and communities, are likely to be greatest when atrazine concentrations in water equal or exceed approximately 10 to 20 µg/L on a recurrent basis or over a prolonged period of time (U.S. EPA, 2003a). Given the large amount of microcosm/mesocosm and field study data for atrazine, only effects data that are more conservative than the 10 µg/L aquatic-community effect level identified in the 2003 atrazine IRED were considered. In addition, data for taxa that are directly relevant to the Barton Springs salamander (i.e., aquatic-phase amphibians) were also 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 2.7. 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.

As described in Agency's Overview Document (U.S. EPA, 2004), the most sensitive endpoint for each taxa is evaluated. For this assessment, evaluated taxa include freshwater fish, freshwater aquatic invertebrates, and freshwater aquatic plants. Currently, no guideline tests exist for salamanders. Therefore, surrogate species were used as described in the Overview Document (U.S. EPA, 2004). In addition, aquatic-phase amphibian ecotoxicity data from the open literature are qualitatively discussed. Table 4.1 summarizes the most sensitive ecological 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. It should be noted that Appendix A also includes ecotoxicity data for taxonomic groups that are not relevant to this assessment (i.e., birds, estuarine/marine fish, invertebrates, and plants) because the Agency is completing endangered species assessments for other species concurrently with this assessment.

Table 4.1. Aquatic Toxicity Profile for Atrazine

Assessment Endpoint	Species	Toxicity Value Used in Risk Assessment	Citation MRID # (Author & Date)	Comment
Acute Direct Toxicity to Salamander	Rainbow trout ¹	96-hour LC ₅₀ = 5,300 μg/L Probit slope = 2.72	000247-16 (Beliles and Scott, 1965)	Acceptable
Chronic Direct Toxicity to Salamander	Brook trout ¹	NOAEC = $65 \mu g/L$ LOAEC = $120 \mu g/L$	000243-77 (Macek et al., 1976)	Acceptable: 7.2% reduction in length; 16% reduction in weight
Indirect Toxicity to Salamander via Acute Toxicity to Freshwater Invertebrates (i.e. prey items)	Midge	48-hour $LC_{50} = 720$ μ g/L Probit slope unavailable	000243-77 (Macek et al., 1976)	Supplemental: raw data unavailable
Indirect Toxicity to Salamander via Chronic Toxicity to Freshwater Invertebrates (i.e. prey items)	Scud	NOAEC = $60 \mu g/L$ LOAEC = $120 \mu g/L$	000243-77 (Macek et al., 1976)	Acceptable: 25 % reduction in development of F ₁ to seventh instar
Indirect Toxicity to Salamander via Acute Toxicity to Non-vascular aquatic plants	4 species of freshwater algae	1-week EC ₅₀ = 1 μ g/L	000235-44 (Torres & O'Flaherty, 1976)	Supplemental: 41 to 98% reduction in chlorophyll production; raw data unavailable
Indirect Toxicity to Salamander via Acute Toxicity to Vascular aquatic plants	Duckweed	14-day $EC_{50} = 37 \mu g/L$	430748-04 (Hoberg, 1993)	Supplemental: 50% reduction in biomass; NOAEC not determined

¹ Used as a surrogate for the Barton Springs salamander. Open literature data for the salamander are presented in Section 4.1.2.

Toxicity to aquatic fish and invertebrates is categorized using the system shown in Table 4.2 (U.S. EPA, 2004). Toxicity categories for aquatic plants have not been defined.

Table 4.2. Categories of Acute Toxicity for Aquatic Organisms

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

4.1.1 Toxicity to Freshwater Fish

As previously discussed, no guideline tests exist for salamanders; therefore, freshwater fish are used as surrogate species for amphibians including salamanders (U.S. EPA, 2004). The available open literature information on atrazine toxicity to aquatic-phase amphibians, which is provided in Section 4.1.2, shows that acute and chronic ecotoxicity endpoints for amphibians are generally less sensitive than fish. 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. A summary of acute and chronic freshwater fish data, including sublethal effects, is provided below.

4.1.1.1 Freshwater Fish: Acute Exposure (Mortality) Studies

Freshwater fish acute toxicity studies were used to assess potential direct effects to the Barton Springs salamander because direct acute toxicity guideline data on salamanders are unavailable. Atrazine toxicity has been evaluated in numerous freshwater fish species, including rainbow trout, brook trout, bluegill sunfish, fathead minnow, tilapia, zebrafish, goldfish, and carp, and the results of these studies demonstrate a wide range of sensitivity. The range of acute freshwater fish LC₅₀ values for atrazine spans one order of magnitude, from 5,300 to 60,000 μg/L; therefore, atrazine is categorized as moderately (>1,000 to 10,000 μg/L) to slightly (>10,000 to 100,000 μg/L) toxic to freshwater fish on an acute basis. The freshwater fish acute LC₅₀ value of 5,300 μg/L is based on a static 96-hour toxicity test using rainbow trout (*Oncorhynchus mykiss*) (MRID # 000247-16). No sublethal effects were reported as part of this study. A complete list of all the acute freshwater fish toxicity data for atrazine is provided in Table A-8 of Appendix A.

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

Similar to the acute data, chronic freshwater fish toxicity studies were used to assess potential direct effects to the Barton Springs salamander because direct chronic toxicity guideline data for salamanders do not exist. Freshwater fish full life-cycle studies for atrazine are available and summarized in Table A-12 of Appendix A. Following 44 weeks of exposure to atrazine in a flow-through system, statistically significant reductions in brook trout mean length (7.2%) and body weight (16%) were observed at a concentration of $120 \,\mu\text{g/L}$, as compared to the control (MRID # 000243-77). The

corresponding NOAEC for this study is 65 μ g/L. Although the acute toxicity data for atrazine show that rainbow trout are the most sensitive freshwater fish, available chronic rainbow trout toxicity data indicates that it is less sensitive to atrazine, on a chronic exposure basis, than the brook trout, with respective LOAEC and NOAEC values of 1,100 μ g/L and 410 μ g/L. Further information on chronic freshwater fish toxicity data for atrazine is provided in Section A.2.2 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 4.1.

Reported sublethal effects in rainbow trout show increased plasma vitellogenin levels in both female and male fish and decreased plasma testosterone levels in male fish at atrazine concentrations of approximately 50 μ g/L (Wieser and Gross, 2002 [MRID 456223-04]). Vitellogenin (Vtg) is an egg yolk precursor protein expressed normally in female fish and dormant in male fish. The presence of Vtg in male fish is used as a molecular marker of exposure to estrogenic chemicals. It should be noted, however, that there is a high degree of variability with the Vtg effects in these studies, which confounds the ability to resolve the effects of atrazine on plasma steroids and vitellogenesis.

In salmon, endocrine-mediated olfactory functions were affected at $0.5~\mu g/L$ atrazine (Moore and Lower, 2001). 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 atrazine at $0.5~\mu g/L$. Overall, the relationship between reduced olfactory response of males to the female 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 is not established. In addition, EPA did not use these data in development of the aquatic life water quality criteria for atrazine because the test material was not adequately described or translated. Furthermore, the study did not determine whether the decreased response of olfactory epithelium to specific chemical stimuli would likely impair similar responses in intact fish.

Although these studies raise concern about the effects of atrazine on endocrine-mediated functions in freshwater and anadromous fish, these effects are difficult to quantify because they are not clearly tied to the assessment endpoints for the Barton Springs salamander (i.e., survival, growth, and reproduction of individuals). In addition, differences in habitat and behavior of the tested fish species compared with the Barton Springs salamander suggest that the results are not readily extrapolated to salamanders. Furthermore, there is uncertainty associated with extrapolating effects observed in the laboratory to more variable exposures and conditions in the field. Therefore, potential sublethal effects on fish are evaluated qualitatively and not used as part of the quantitative risk characterization. Further detail on sublethal effects to fish is provided in Sections A.2.4a and A.2.4b of Appendix A.

4.1.2 Toxicity to Aquatic-phase Amphibians

Available toxicity information on potential atrazine-related mortality and sublethal effects to aquatic-phase amphibians (including salamanders) from the open literature is summarized below in Sections 4.1.2.1 and 4.1.2.2, respectively. Guideline ecotoxicity studies for amphibians are not available.

4.1.2.1 Amphibians: Open Literature Data on Mortality

Available acute data for amphibians, including the leopard frog (*Rana pipiens*), wood frog (*R. sylvaticas*), and American toad (*Bufo americanus*), indicate that they are relatively insensitive to atrazine with acute LC_{50} values > 20,000 µg/L (Allran and Karasov, 2001). Acute toxicity data are not available for the salamander.

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

4.1.2.2 Amphibians: Open Literature Data on Sublethal Effects

An evaluation and review of the results of submitted studies regarding potential atrazine effects data on amphibian gonadal development are presented in the Agency's White Paper (U.S. EPA, 2003d) and discussed in Section A.2.4c of Appendix A. As previously discussed in Section 2.3, the Agency has concluded that there is currently insufficient evidence to confirm or refute the hypothesis that atrazine exposure may impact gonadal development in amphibians. Therefore, the Agency has requested additional data from the registrant to reduce uncertainties regarding potential risk to amphibians. In addition to addressing uncertainty regarding the potential of atrazine to cause these effects, these studies are expected to characterize the nature of any potential dose-response

relationship. The initial results of these amphibian studies are expected to become available in late 2006 to early 2007; therefore, as of this writing, they are not available for inclusion in this endangered species risk assessment for the Barton Springs salamander.

Open literature data on sublethal effects of atrazine to aquatic-phase amphibians, including frogs and salamanders, are summarized below and discussed in greater detail in Section A.2.4d of Appendix A. The following information includes studies identified as part of the 2006 open literature search that were not reviewed as part of the Agency's 2003 White Paper (U.S. EPA, 2003d).

Frogs (Anurans)

The reviewed studies were classified as qualitative because they address issues of concern to the risk assessment, but are not appropriate for quantitative use due to uncertainties related to a lack of raw data and limitations in the study design. Further information on the study design and uncertainties associated with each of the reviewed studies are provided in Section A.2.4d and Table A-16 of Appendix A. In summary, the microcosm/mesocosm and chronic lab data for frogs indicate that sublethal effects to amphibians, such as reduced mass and length at metamorphosis may occur at atrazine exposure concentrations of approximately 200 μ g/L and higher under the conditions tested (Diana et al., 2000; Boone and James, 2003; and Gucciardo, 1999). Decreased frog weight (and length) at metamorphosis at \geq 200 μ g/L atrazine is hypothesized to result from atrazine's effect on algal populations, which are a primary source of food for developing anurans (Diana et al., 2000). Other factors, such as decreasing DO, pH, and macrophyte biomass following atrazine exposure may also contribute to observed sublethal effects.

In the lab, plasma testosterone was reduced in male frogs at atrazine concentrations of 259 μ g/L; however, an increase in aromatase activity (aromatase increases synthesis of 17 β -estradiol resulting in depletion of testosterone levels) was not observed (Hecker et al., 2005). Therefore, the mechanism associated with decreased testosterone levels in adult males is unclear.

The observed effect levels of $\sim 200 \,\mu\text{g/L}$ are greater than the aquatic community-level effects of 10-20 $\,\mu\text{g/L}$ documented in the January 2003 atrazine IRED (U.S. EPA, 2003a). In addition, uncertainties and associated limitations in the design of the reviewed studies are similar to the conclusions previously reported (U.S. EPA, 2003d).

Salamanders (Caudates)

The reviewed sublethal studies contain variable results with respect to atrazine exposures and sublethal effects to aquatic-phase salamanders. Two chronic studies on the streamside salamander (*A. barbouri*) and long-toed salamander (*A. macrodactylum*) show reduced mass and snout-vent length (SVL) at metamorphosis, in addition to accelerated metamorphosis, relative to controls, at atrazine concentrations ranging from 184 to 400 µg/L (Rohr et al., 2004; Forson and Storfer, 2006). The NOAEC values for these studies

range between 18.4 and 40 μ g/L. In another study, the time to metamorphosis was increased in small-mouthed salamanders (*A. texanum*) at the only concentration of atrazine tested (197 μ g/L); however, no effect on the time to metamorphosis was observed in spotted salamanders at the same concentration of atrazine (Boone and James, 2003).

The interaction of atrazine and one of the iridoviruses (the *Ambystoma tigrinum* virus [ATV]) was studied in long-toed salamanders by Forson and Storfer (2006). Larvae exposed to both atrazine and ATV had lower levels of mortality and ATV infectivity compared to larvae exposed to virus alone, suggesting that atrazine may compromise virus efficacy or improve salamander immune competency. Behavioral changes in locomotion (i.e., increased activity following tapping on tanks) were observed in streamside salamanders exposed to 400 μ g/L; however, this endpoint is not relevant to the assessment endpoints chosen for this risk assessment. It is unclear how increased larval salamander activity due to tank tapping in the lab would translate into reduced fitness in the wild.

All of the reviewed salamander studies from the open literature were classified as qualitative because they address issues of concern to the risk assessment, but are not appropriate for quantitative use due to uncertainties related to a lack of raw data and limitations in the study design. Further information on the study design and uncertainties associated with each of the reviewed studies is provided in Section A.2.4d and Table A-17 of Appendix A.

4.1.3 Toxicity to Freshwater Invertebrates

Freshwater aquatic invertebrate toxicity data were used to assess potential indirect effects of atrazine to the Barton Springs salamander. Direct effects to freshwater invertebrates resulting from exposure to atrazine may indirectly affect the Barton Springs salamander via reduction in available food. As discussed in Section D.5.1 of Appendix D, 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. Based on analysis of the stomach and fecal samples from a limited number of adult and juvenile Barton Springs salamanders, the most prevalent organisms found 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.

A summary of acute and chronic freshwater invertebrate data, including published data in the open literature since completion of the IRED (U.S. EPA, 2003a), is provided below in Sections 4.1.3.1 through 4.1.3.3.

4.1.3.1 Freshwater Invertebrates: Acute Exposure Studies

Atrazine is classified as highly toxic to slightly toxic to aquatic invertebrates. There is a wide range of EC_{50}/LC_{50} values for freshwater invertebrates with values ranging from

720 to $>33,000~\mu g/L$. The freshwater LC₅₀ value of 720 μ g/L is based on an acute 48-hour static toxicity test for the midge, *Chironomus tentans* (MRID # 000243-77). Further evaluation of the available acute toxicity data for the midge shows high variability with the LC₅₀ values, ranging from 720 to $>33,000~\mu g/L$. With the exception of the midge, reported acute toxicity values for the other five freshwater invertebrates (including the water flea, scud, stonefly, leech, and snail) are 3,500 μ g/L and higher. All of the available acute toxicity data for freshwater invertebrates are provided in Section A.2.5 and Table A-18 of Appendix A. The LC₅₀/EC₅₀ distribution for freshwater invertebrates is graphically represented in Figure 4.1. The columns represent the lowest reported value for each species, and the positive y error bar represents the maximum reported value. Values in parentheses represent the number of studies included in the analyses.

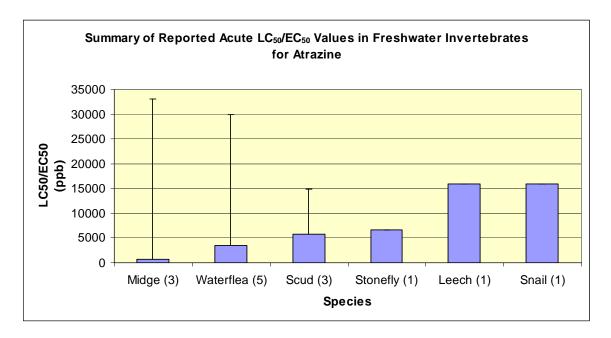


Figure 4.1. Summary of Reported Acute LC₅₀/EC₅₀ Values in Freshwater Invertebrates for Atrazine

4.1.3.2 Freshwater Invertebrates: Chronic Exposure Studies

The most sensitive chronic endpoint for freshwater invertebrates is based on a 30-day flow-through study on the scud (*Gammarus fasciatus*), which showed a 25% reduction in the development of F_1 to the seventh instar at atrazine concentrations of 140 μ g/L; the corresponding NOAEC is 60 μ g/L (MRID # 000243-77). Although the acute toxicity data for atrazine show that the midge (*Chironomus tentans*) is the most sensitive freshwater invertebrate, available chronic midge toxicity data indicate that it is less sensitive to atrazine, on a chronic exposure basis, than the scud, with respective LOAEC and NOAEC values of 230 μ g/L and 110 μ g/L. Additional information on the chronic toxicity of atrazine to freshwater invertebrates is provided in Section A.2.6 and Table A-20 of Appendix A.

4.1.3.3 Freshwater Invertebrates: Open Literature Data

One additional acute study for an underrepresented taxa of freshwater mussels was located in the open literature. The results of the study by Johnson et al. (1993) suggest that 48-hour exposures at atrazine concentrations up to 60 mg/L do not affect the survival of juvenile and mature freshwater mussels, *Anodonta imbecilis*; therefore, *A. imbecilis* is less acutely sensitive to atrazine than other freshwater invertebrates.

4.1.4 Toxicity to Aquatic Plants

Aquatic plant toxicity studies were used as one of the measures of effect to evaluate whether atrazine may affect primary production. 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 atrazine to affect primary productivity. Laboratory studies were used to determine whether atrazine may cause direct effects to aquatic plants. In addition, the threshold concentrations, described in Section 4.2, were used to further characterize potential community level effects to Barton Springs salamanders resulting from potential effects to aquatic plants. A summary of the laboratory data for aquatic plants is provided in Section 4.1.4.1. A description of the threshold concentrations used to evaluate community-level effects is included in Section 4.2.

4.1.4.1 Aquatic Plants: Laboratory Data

Numerous aquatic plant toxicity studies have been submitted to the Agency. A summary of the data for freshwater vascular and non-vascular plants is provided below. Section A.4.2 and Tables A-40 and A-41 of Appendix A include a more comprehensive description of these data.

The Tier II results for freshwater aquatic plants indicate that atrazine causes a 41 to 98% reduction in chlorophyll production of freshwater algae; the corresponding EC_{50} value for four different species of freshwater algae is 1 μ g/L, based on data from a 7-day acute study (MRID # 000235-44). Vascular plants are less sensitive to atrazine than freshwater non-vascular plants with an EC_{50} value of 37 μ g/L, based on reduction in duckweed growth (MRID # 430748-04).

Comparison of atrazine toxicity levels for three different algae endpoints suggests that the endpoints in decreasing order of sensitivity are cell count, growth rate and oxygen production (Stratton, 1984). Walsh (1983) exposed *Skeletonema costatum* to atrazine and concluded that atrazine is only slightly algicidal at relatively high concentrations (i.e., 500 & 1,000 µg/L). Caux et al. (1996) compared the cell count IC₅₀ and fluorescence LC₅₀ and concluded that atrazine is algicidal at concentrations affecting cell counts.

Abou-Waly et al. (1991) measured growth rates on days 3, 5, and 7 for two algal species. The pattern of atrazine effects on growth rates differs sharply between the two species. Atrazine had a strong early effect on *Anabaena flos-aquae* followed by rapid recovery in clean water (i.e., EC_{50} values for days 3, 5, and 7 are 58, 469, and 766 μ g/L, respectively). The EC_{50} values for *Selenastrum capricornutum* continued to decline from day 3 through 7 (i.e., 283, 218, and 214 μ g/L, respectively). Based on theses results, it appears that the timing of peak effects for atrazine may differ depending on the test species.

It should be noted that rrecovery from the effects of atrazine and the development of resistance to the effects of atrazine in some vascular and non-vascular aquatic plants has been reported and may add uncertainty to these findings. However, reports of recovery are often based on differing interpretations of recovery. Thus, before recovery can be considered as an uncertainty, an agreed upon interpretation is needed. The Agency believes that recovery has a simple and straight forward interpretation: a return to pre-exposure levels for the *affected population*, not for a replacement population of more tolerant species. Further research is needed to quantify the impact that recovery and resistance would have on aquatic plants.

4.1.5 Freshwater Field Studies

Microcosm and mesocosm studies with atrazine provide measurements of primary productivity that incorporate the aggregate responses of multiple species in aquatic plant communities. Because plant species vary widely in their sensitivity to atrazine, the overall response of the plant community may be different from the responses of the individual species measured in laboratory toxicity tests. Mesocosm and microcosm studies allow observation of population and community recovery from atrazine effects and of indirect effects on higher trophic levels. In addition, mesocosm and microcosm studies, especially those conducted in outdoor systems, incorporate partitioning, degradation, and dissipation, factors that are not usually accounted for in laboratory toxicity studies, but that may influence the magnitude of ecological effects.

Atrazine has been the subject of many mesocosm and microcosm studies in ponds, streams, lakes, and wetlands. The durations of these studies have ranged from a few weeks to several years at exposure concentrations ranging from $0.1~\mu g/L$ to $10,000~\mu g/L$. Most of the studies have focused on atrazine effects on phytoplankton, periphyton, and macrophytes; however, some have also included measurements on animals.

As described in the 2003 IRED for atrazine (U.S. EPA, 2003a), potential adverse effects on sensitive aquatic plants and non-target aquatic organisms including their populations and communities are likely to be greatest when atrazine concentrations in water equal or exceed approximately 10 to 20 μ g/L on a recurrent basis or over a prolonged period of time. A summary of all the freshwater aquatic microcosm, mesocosm, and field studies that were reviewed as part of the 2003 IRED is included in Section A.2.8a and Tables A-22 through A-24 of Appendix A. Given the large amount of microcosm and mesocosm and field study data for atrazine, only effects data less than or more conservative than the

 $10~\mu g/L$ aquatic community effect level identified in the 2003 IRED were summarized from the open literature search that was completed in February 2006. Field study data for aquatic-phase amphibians, including frogs and salamanders, are summarized in Section 4.1.2 and discussed in greater detail in Sections A.2.3 and A.2.4 of Appendix A. Based on the selection criteria for review of new open literature, all of the available studies show effects levels to freshwater fish, invertebrates, and aquatic plants at concentrations greater than $10~\mu g/L$.

Community-level effects to aquatic plants that are likely to result in indirect effects to the rest of the aquatic community, including the Barton Springs salamanders, are evaluated based on threshold concentrations. These screening threshold concentrations, which are discussed in greater detail in Section 4.2 and Appendix B, incorporate the available micro- and mesocosm data included in the 2003 IRED, as well as additional information gathered following completion of the 2003 atrazine IRED (U.S. EPA, 2003b and 2003e).

4.2 Community-Level Endpoints: Threshold Concentrations

In this endangered species assessment, direct and indirect effects to the Barton Springs salamander are evaluated in accordance with the screening-level methodology described in the Agency's Overview Document (U.S. EPA, 2004). If aquatic plant RQs exceed the Agency's non-listed species LOC (because the salamander does not have an obligate relationship with any one particular plant species, but rather relies on multiple plant species), based on available EC₅₀ data for vascular and non-vascular plants, risks to individual aquatic plants are assumed.

It should be noted, however, that the indirect effects analysis in this assessment is unique, in that the best available information for atrazine-related effects on aquatic communities is significantly more extensive than for other pesticides. Hence, atrazine effects determinations can utilize more refined data than is generally available to the Agency. Specifically, a robust set of microcosm and mesocosm data and aquatic ecosystem models are available for atrazine that allowed EPA to refine the indirect effects associated with potential aquatic community-level effects (via aquatic plant community structural change and subsequent habitat modification) to the Barton Springs salamander. Use of such information is consistent with the guidance provided in the Overview Document (U.S. EPA, 2004), which specifies that "the assessment process may, on a case-by-case basis, incorporate additional methods, models, and lines of evidence that EPA finds technically appropriate for risk management objectives" (Section V, page 31 of U.S. EPA, 2004). This information, which represents the best scientific data available, is described in further detail below and in Appendix B.

As previously mentioned in Section 2.3, the Agency has selected an atrazine level of concern (LOC) in the 2003 IRED (U.S. EPA, 2003a and b) that is consistent with the approach described in the Office of Water's (OW) draft atrazine aquatic life criteria (U.S. EPA, 2003c). Through these previous analyses (U.S. EPA, 2003a, b, and c), which reflect the current best available information, predicted or monitored aqueous atrazine concentrations can be interpreted to determine if a water body is likely to be significantly

affected via indirect effects to the aquatic community. Potential impacts of atrazine to plant community structure and function that are likely to result in indirect effects to the rest of the aquatic community, including the Barton Springs salamander, are evaluated as described below.

As described further in Appendix B, responses in microcosms and mesocosms exposed to atrazine were evaluated to differentiate no or slight, recoverable effects from significant, generally non-recoverable effects (U.S. EPA, 2003e). Because effects varied with exposure duration and magnitude, there was a need for methods to predict relative differences in effects for different types of exposures. The Comprehensive Aquatic Systems Model (CASM) (Bartell et al., 2000; Bartell et al., 1999; DeAngelis et al., 1989) was selected as an appropriate tool to predict these relative effects, and was configured to provide a simulation for the entire growing season of a 2nd and 3rd order Midwestern stream as a function of atrazine exposure. CASM simulations conducted for the concentration/duration exposure profiles of the micro- and mesocosm data showed that CASM seasonal output, represented as an aquatic plant community similarity index, correlated with the micro- and mesocosm effect scores, and that a 5% change in this index reasonably discriminated micro- and mesocosm responses with slight versus significant effects. The CASM-based index was assumed to be applicable to more diverse exposure conditions beyond those present in the micro- and mesocosm studies.

To avoid having to routinely run the CASM model, simulations were conducted for a variety of actual and synthetic atrazine chemographs to determine 14-, 30-, 60-, and 90-day average concentrations that discriminated among exposures that were unlikely to exceed the CASM-based index (i.e., 5% change in the index). It should be noted that the average 14-, 30-, 60-, and 90-day concentrations were originally intended to be used as screening values to trigger a CASM run (which is used as a tool to identify the 5% index change LOC), rather than actual thresholds to be used as an LOC (U.S. EPA, 2003e). The following threshold concentrations for atrazine were identified (U.S. EPA, 2003e):

- 14-day average = $38 \mu g/L$
- 30-day average = $27 \mu g/L$
- 60-day average = $18 \mu g/L$
- 90-day average = $12 \mu g/L$

Effects of atrazine on aquatic plant communities that have the potential to subsequently pose indirect effects to the Barton Springs salamander are best addressed using the robust set of micro- and mesocosm studies available for atrazine and the associated risk estimation techniques (U.S. EPA, 2003a, b, c, and e). The 14-, 30-, 60-, and 90-day threshold concentrations developed by EPA (U.S. EPA 2003e) are used to evaluate potential indirect effects to aquatic communities for the purposes of this endangered species assessment. Use of these threshold concentrations is considered appropriate because: (1) the CASM-based index meets the goals of the defined assessment endpoints for this assessment; (2) the threshold concentrations provide a reasonable surrogate for the CASM index; and (3) the additional conservatism built into the threshold concentration, relative to the CASM-based index, is appropriate for an endangered

species risk assessment (i.e., the threshold concentrations were set to be conservative, producing a low level (1%) of false negatives relative to false positives). Therefore, these threshold concentrations are used to identify potential indirect effects (via aquatic plant community structural change) to the Barton Springs salamander. If modeled atrazine EECs exceed the 14-, 30-, 60- and 90-day threshold concentrations following refinements of potential atrazine concentrations with available monitoring data, the CASM model could be employed to further characterize the potential for indirect effects. A step-wise data evaluation scheme incorporating the use of the screening threshold concentrations is provided in Figure 4.2. Further information on threshold concentrations is provided in Appendix B.

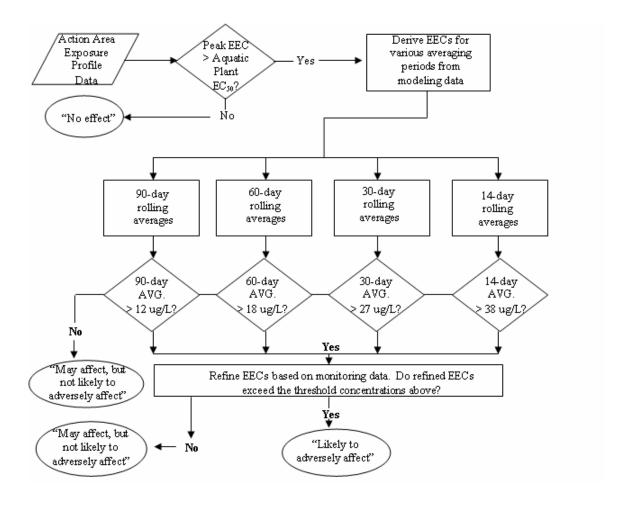


Figure 4.2. Use of Threshold Concentrations in Endangered Species Assessment

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

The Agency uses the probit dose response relationship as a tool for providing additional information on the potential for acute direct effects to individual listed species and aguatic 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 atrazine on par with the acute toxicity endpoint selected for RQ calculation. To accomplish this interpretation, the Agency uses the slope of the dose response relationship available from the toxicity study used to establish the acute toxicity measures of effect for each taxonomic group that is relevant to this assessment (i.e., freshwater fish used as a surrogate for aquatic-phase amphibians and freshwater invertebrates). The individual effects probability associated with the acute RQ is based on the mean estimate of the slope and an assumption of a probit dose response relationship. In addition to a single effects probability estimate based on the mean, upper and lower estimates of the effects probability are also provided to account for variance in the slope, if available. The upper and lower bounds of the effects probability are based on available information on the 95% confidence interval of the slope. A statement regarding the confidence in the estimated event probabilities is also included. Studies with good probit fit characteristics (i.e., statistically appropriate for the data set) are associated with a high degree of confidence. Conversely, a low degree of confidence is associated with data from studies that do not statistically support a probit dose response relationship. In addition, confidence in the data set may be reduced by high variance in the slope (i.e., large 95% confidence intervals), despite good probit fit characteristics.

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

4.4 Incident Database Review

A number of incidents have been reported in which atrazine has been associated with some type of environmental effect, with variable levels of certainty that atrazine caused the effects, ranging from unlikely to highly probable. As of the writing of the 2003 IRED (U.S. EPA, 2003a), 109 incidents were listed in the Ecological Incident Information System (EIIS) files under atrazine: 4 cases were listed as highly probable, 40 as probable, 50 as possible, 13 as unlikely, and 2 as unrelated. Atrazine alone is not very toxic to the birds, mammals, and aquatic animals cited in most of these incidents. In none of these cases has evidence been provided that firmly demonstrates that atrazine has produced the reported effects. Atrazine residues in fish tissue were measured in only one incident

reported as a fish kill (# I004021-004); however, many chemicals were identified and high profenofos levels were found. Therefore, the organophosphate was determined to be responsible for the large fish kill. In many cases, the inference of these reported incidents to atrazine is likely due to the widespread use of atrazine and the proximity of the atrazine application and timing to the occurrence of the incident.

Between October 26, 2000 and June 9, 2006, 8 incidents were listed in the EIIS involving the use of atrazine: 6 cases are listed as possible and 2 are listed as unlikely. The effects of these incidents ranged from major fish kills to minor burning of garden plants adjacent to a field treated with atrazine. Of these incidents, 5 were caused by drift, 1 by runoff and 2 because of misuse.

Of the 6 cases that were listed as "Possible," all were terrestrial and, therefore, not relevant to this assessment. In the two cases listed as "Unlikely," one resulted in the death of 50-60 bass, 2,000 crappie and 300-400 bluegills (IN: #I013987-001). Three chemicals, including terbufos, atrazine and acetochlor, were used in a product suspected to be present in the runoff. Tests were conducted in the two affected ponds and terbufos was the only chemical listed as being detected in both. It is not clear if atrazine and acetochlor were measured in the pond water analysis. However, it is likely that terbufos was responsible for the fish kill because it has a greater lethality to fish than atrazine and acetochlor.

One of the two reported "misuse" incidents caused substantial damage to aquatic animals (TN: # I016990-001) resulting in the death of 2,000 bluegill sunfish, 400 catfish, and a snake. This incident was credited to the dumping of 4 to 5 gallons of a product suspected of containing atrazine into a one half acre pond.

Based on the available incident information, supporting data is not available to clearly demonstrate that atrazine is the cause of the observed aquatic effects (i.e., death to fish). In addition, the best available toxicity information shows that atrazine is not directly toxic to freshwater fish (and aquatic-phase amphibians) at environmentally relevant concentrations (see Sections 4.1.1 and 4.1.2). Further information on the atrazine incidents reported in the 2003 IRED (U.S. EPA, 2003a) and a summary of uncertainties associated with all reported incidents are provided in Appendix F.

5. Risk Characterization

Risk characterization is the integration of the exposure and effects characterizations to determine the potential ecological risk from varying atrazine 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 effects determination (i.e., "no effect," "likely to adversely affect," or "may affect, but not likely to adversely affect") for the Barton Springs salamander.

5.1 Risk Estimation

Risk was estimated by calculating the ratio of the estimated environmental concentrations (EECs; see Table 3.5) and the appropriate toxicity endpoint (see Table 4.1). This ratio is the risk quotient (RQ), which is then compared to pre-established acute and chronic levels of concern (LOCs) for each category evaluated (Appendix G). Screening-level RQs are based on the most sensitive endpoints and modeled surface water concentrations from the following scenarios for atrazine:

- residential granular use @ 2 lb ai/A; 2 applications with 30 days between applications (assumes 1% over-application of atrazine granules to impervious surfaces)
- residential liquid use @ 1 lb ai/A; 2 applications with 30 days between applications (assumes 1% over-spray of atrazine to impervious surfaces)
- turf granular use @ 2 lb ai/A; 2 applications with 30 days between applications
- turf liquid use @ 1 lb ai/A; 2 applications with 30 days between applications
- rights-of-way liquid use @ 1 lb ai/A; 1 application (assumes 1% over-spray of atrazine to impervious surfaces)
- fallow/idle land use @ 2.25 lb ai/A; 1 application

In cases where the screening-level RQ exceeds one or more LOCs, additional factors, including Barton Springs salamander life history characteristics, refinement of the EECs using available monitoring data, and consideration of community-level threshold concentrations, are considered and used to characterize the potential for atrazine to affect the Barton Springs salamander. Risk estimations of direct and indirect effects of atrazine 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 atrazine degradates, including HA, DEA, DIA, and DACT, is assumed to be less than the parent compound; therefore, RQ values were not derived for the degradates.

5.1.1 Direct Effects

Direct effects associated with acute and chronic exposure to atrazine in Barton Springs are not expected to occur for the Barton Springs salamander. Risk quotients used to estimate direct effects to the Barton Springs salamander are provided in Table 5.1 below. Risk quotients were calculated only for the use that resulted in the highest EEC (granular residential use) because none of the acute or chronic LOCs were exceeded. These risk quotients are further characterized in Section 5.2.1.

Table 5.1. Summary of Direct Effect RQs for the Barton Springs Salamander

Effect to Barton Springs Salamander	Surrogate Species	Toxicity Value (μg/L)	EEC (μg/L)	RQ	Probability of Individual Effect	LOC Exceedance and Risk Interpretation
Acute Direct Toxicity	Rainbow trout	$LC_{50} = 5,300$	Peak: 41.2	0.008	1 in 1.7E+08 (1 in 1,870 to 1 in 5.82E+15) ^a	No ^b
Chronic Direct Toxicity	Brook trout	NOAEC = 65	60-day: 1.2	0.018	Not calculated for chronic endpoints	No ^b

^a Based on a probit slope of 2.72 with 95% confidence intervals of 1.56 and 3.89 (MRID# 000247-16).

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 salamander'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 atrazine 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

^b RQ < acute endangered species LOC of 0.05.

between the listed species and non-listed 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 concluded. If plant RQs are above non-listed 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 D, the Barton Springs salamander relies on multiple plant species, including aquatic moss, pondweed, arrowhead, water primrose, cabomba, and other aquatic plants for cover and as a source of habitat and food for the variety and abundance of aquatic invertebrates that salamanders eat.

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, respectively.

5.1.2.1 Evaluation of Potential Indirect Effects via Reduction in Food Items (Freshwater 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 and the distribution of the sensitivities of the prey organisms to atrazine. 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 Tables 5.2 and 5.3, respectively. Acute and chronic RQs are based on the most sensitive toxicity endpoint for the midge (EC $_{50}$ = 720 µg/L) and the scud (NOAEC = 60 µg/L), respectively.

Table 5.2. Summary of RQs Used to Estimate Indirect Effects to the Barton Springs Salamander via Direct Acute Effects on Dietary Items

Indirect Effect to Barton Springs Salamander	Surrogate Food Item / Toxicity Value (µg/L)	Use (appl. method; rate; # appl.; interval between appl.)	Peak EECs (μg/L)	RQ	Probability of Individual Effect ^a	LOC Exceedance and Risk Interpretation
Reduced Food Supply via Acute Direct	Midge EC50 = 720	Residential (granular; 2 lb ai/A; 2 appl.; 30 d	41.2	0.057	1 in 4.55E+07	Yes ^b
Toxicity to		interval)				
Invertebrates		Residential (ground liquid; 1 lb ai/A; 2 appl.; 30 d interval)	26.6	0.037	1 in 6.72E+09	No ^c
			22.4	0.031	1 in 6.29E+10	No ^c
		Turf (ground liquid; 1 lb ai/A; 2 appl.; 30 d interval)	16.2	0.023	1 in 3.53E+12	No ^c
		Fallow/Idle land (aerial liquid; 2.25 lb ai/A; 1 appl.)	7.5	0.010	1 in 1.46E+18	No ^c
3.01		Rights-of-Way (liquid; 1 lb ai/A; 1 appl.)	6.2	0.009	1 in 8.97E+18	No ^c

^a Slope information on the toxicity study that was used to derive the RQ for freshwater invertebrates is not available. Therefore, the probability of an individual effect was calculated using a probit slope of 4.4, which is the only technical grade atrazine value, reported in the available freshwater invertebrates studies that may serve as food items for the salamander; 95% confidence intervals could not be calculated based on the available data (Table A-18; Taylor et al., 1991; MRID# 452029-17).

For freshwater invertebrates, acute RQs exceed the acute risk to the listed species LOC of 0.05 for the residential granular use (2 lb ai/A) only. Acute RQs for the other modeled atrazine uses are less than the listed species LOC. Because the listed species LOC is exceeded for the residential granular use, atrazine use related to residential granular applications has the potential to indirectly affect the Barton Springs salamander via reduction in the availability of sensitive aquatic invertebrate food items. However, this analysis was based on the most sensitive aquatic invertebrate endpoint of freshwater species tested in laboratory studies and did not consider the range of aquatic invertebrate species sensitivity to atrazine or the specific dietary requirements of the Barton Springs

^b RQ > acute listed species LOC of 0.05. Further evaluation of the range of freshwater invertebrate species sensitivity to atrazine and dietary requirements of the Barton Springs salamander is completed in Section 5.2.2.

^c RQ < acute listed species LOC of 0.05.

salamander. Therefore, additional characterization of the potential for atrazine to affect freshwater invertebrate food items of the Barton Springs salamander is presented as part of the Risk Description in Section 5.2.2.

Table 5.3. Summary of RQ and LOC Used to Estimate Indirect Effects to the Barton Springs Salamander via Direct Chronic Effects on Dietary Items

Indirect Effect to Barton Springs Salamander	Surrogate Food Item / Toxicity Value (µg/L)	Use (appl. method; rate; # appl.; interval between appl.)	21-day EECs (μg/L)	RQ	LOC Exceedance and Risk Interpretation
Reduced Food Supply via Chronic Direct Toxicity to Invertebrates	Scud NOAEC = 60	Residential (granular; 2 lb ai/A; 2 appl.; 30 d interval)	2.5	0.04	No ^a

^a RQ < chronic risk LOC of 1.0.

As shown in Table 5.3, the chronic LOC is not exceeded for freshwater invertebrates, based on the use that results in the highest EECs (granular residential use). Therefore, indirect effects to the Barton Springs salamander based on direct chronic effects to dietary items are not expected to occur.

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

Potential indirect effects from effects on habitat and/or primary productivity were assessed using RQs from freshwater aquatic vascular and non-vascular plant data as a screen. If aquatic plant RQs exceed the Agency's non-listed species LOC (because the salamander relies on multiple plant species), potential community level effects are evaluated using the threshold concentrations, as described in Section 4.2. Risk quotients used to estimate potential indirect effects to the Barton Springs salamander from effects on aquatic plants primary productivity are summarized in Table 5.4.

Table 5.4. Summary of RQs Used to Estimate Indirect Effects to the Barton Springs Salamander via Direct Effects on Aquatic Plants

Indirect Effect to Barton Springs Salamander	Use (appl. method; rate; # appl.; interval between appl.)	Peak EECs (μg/L)	Non-vascular plant RQ $(EC_{50} = 1 \mu g/L^a)$	Vascular plant RQ (EC50 = 37 μg/L ^b)	LOC Exceedance and Risk Interpretation
Reduced Habitat and/or Primary	Residential (granular; 2 lb ai/A; 2 appl.; 30 d interval)	41.2	41.2	1.11	Yes ^c
Productivity via Direct Toxicity to Aquatic	Residential (ground liquid; 1 lb ai/A; 2 appl.; 30 d interval)	26.6	26.6	0.72	Yes ^d
Plants	Turf (granular; 2 lb ai/A; 2 appl.; 30 d interval)	22.4	22.4	0.61	Yes ^d
	Turf (ground liquid; 1 lb ai/A; 2 appl.; 30 d interval)	16.2	16.2	0.44	Yes ^d
	Fallow/Idle land (aerial liquid; 2.25 lb ai/A; 1 appl.)	7.5	7.5	0.20	Yes ^d
	Rights-of-Way (liquid; 1 lb ai/A; 1 appl.)	6.2	6.2	0.17	Yes ^d

^a Based on 1-week EC₅₀ value of 1 μ g/L for four species of freshwater algae (MRID # 000235-44).

Based on the results shown in Table 5.4, LOCs (RQ \geq 1.0) for direct effects to aquatic non-vascular plants are exceeded for all modeled atrazine use scenarios; LOCs for direct effects to aquatic vascular plants are exceeded only for the residential granular use scenario. Therefore, atrazine has the potential to indirectly affect the Barton Springs salamander via direct effects on both non-vascular and vascular aquatic plants for the residential granular scenario, and via direct effects on non-vascular aquatic plants for all modeled use scenarios. However, this screening-level analysis was based on the most sensitive EC50 value from all of the available freshwater aquatic plant toxicity information. No known obligate relationship exists between the Barton Springs salamander and any single freshwater plant species; therefore, listed species RQs using the NOAEC/EC05 values for aquatic plants were not derived. Further analysis of the time-weighted EECs relative to their respective threshold concentrations is necessary to determine whether effects to individual plant species would likely result in community-level effects. This analysis is presented as part of the Risk Description in Section 5.2.3.

^b Based on 14-day EC₅₀ value of 37 μg/L for duckweed (MRID # 430748-08).

^c RQ > non-listed species LOC of 1.0 for both non-vascular and vascular plants. Direct effects to non-vascular and vascular aquatic plants are possible. Further evaluation of the EECs relative to the threshold concentrations (for community-level effects) is necessary.

^d RQ > non-listed aquatic plant species LOC of 1.0 for non-vascular plants; RQ < non-listed plant species LOC of 1.0 for vascular plant. Direct effects to non-vascular aquatic plants are possible. Further evaluation of the EECs relative to the threshold concentrations (for community-level effects) is necessary.

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, a "no effect" determination is made, based on atrazine'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.

Following a "may affect" determination, additional information is considered to refine the potential for exposure at the predicted levels based on the life history characteristics (i.e., habitat range, feeding preferences, etc) of the Barton Spring salamander and potential community-level effects to aquatic plants. Based on the best available information, the Agency uses the refined evaluation to distinguish those actions that "may affect, but are not likely to adversely affect" from those actions that are "likely to adversely affect" the Barton Springs salamander.

The criteria used to make determinations that the effects of an action are "not likely to adversely affect" the Barton Springs salamander include the following:

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

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

5.2.1 Direct Effects to the Barton Springs Salamander

Respective acute and chronic RQs of 0.008 and 0.018 (based on the modeled EECs from the residential granular scenario assuming 1% overspray and 30% impervious surfaces) are well below the Agency's acute and chronic risk LOCs for all modeled uses of atrazine within the action area. Using an upper bound assumption of residential granular use EECs, based on 10% overspray and 10% impervious surfaces (peak EEC = 67.1 μ g/L and 60-day EEC = 2.0 μ g/L; see Table 3.9), also results in respective acute and chronic RQs of 0.013 and 0.03 that are less than the Agency's LOCs. 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.

The probability of an individual event to the Barton Springs salamander was calculated for the acute RQ of 0.008, based on the dose response curve slope from the acute toxicity study for the rainbow trout of 2.72 (MRID # 000247-16). The corresponding estimated chance of an individual acute mortality to the Barton Springs salamander at an RQ level of 0.008 (based on the acute toxic endpoint for surrogate freshwater fish) is 1 in 170 million. It is recognized that extrapolation of very low probability events is associated with considerable uncertainty in the resulting estimates. In order to explore the possible bounds to such estimates, the upper and lower default values for the rainbow trout dose response curve slope estimate (95% C.I.: 1.56 to 3.89) were used to calculate upper and lower estimates of the effects probability associated with the acute RQ. The respective lower and upper effects probability estimates are 1 in 1,870 (0.05%) and 1 in 5.82E+15 (~1.7E-14%). Given the low probability of an individual mortality occurrence and acute and chronic RQs that are well below LOCs, atrazine is not likely to cause direct adverse effects to the Barton Springs salamander.

Further lines-of-evidence that atrazine is unlikely to cause direct adverse effects to the Barton Springs salamander are provided by the information in the open literature. As previously discussed, the Agency has concluded that there is currently insufficient evidence to confirm or refute the hypothesis that atrazine exposure may impact gonadal development in amphibians (U.S. EPA, 2003d). Further examination of the available open literature data for aquatic-phase amphibians (discussed in Section 4.1.2) shows that exposure to atrazine does not cause direct acute and/or chronic mortality at environmentally relevant concentrations similar to the upper bounds of the modeled EECs. Reported sublethal effects to aquatic-phase amphibians show reduced weight and length at metamorphosis for frogs and salamanders at atrazine exposure concentrations of approximately 200 and accelerated metamorphosis in salamanders at concentrations of approximately 184 μ g/L; however, no effects to growth or time to metamorphosis have been reported at concentrations of \leq 68 μ g/L, similar to the upper bound of modeled EECs for atrazine uses within the action area. Therefore, direct effects to the survival, growth, and reproduction of Barton Springs salamanders are unlikely to occur.

As discussed in Section 4.1.1.3, several open literature studies raise concern about sublethal effects of atrazine on endocrine-mediated functions in freshwater fish, which are used as a surrogate for aquatic-phase amphibians. However, the significance of these effects is difficult to quantify because they are not quantitatively linked to changes in survival, growth, and reproduction of individuals (i.e., the assessment endpoints for the Barton Springs salamander). In addition, differences in habitat and behavior of the tested species compared with the Barton Springs salamander suggest that the results may not be relevant to this assessment. Furthermore, there is uncertainty associated with extrapolating effects observed in the laboratory to more variable exposures and conditions in the field. Further details on potential atrazine-related sublethal effects to fish are provided in Appendix A.

A review of the available aquatic incidents shows that only two incidents involving fish kills have been reported from 2000 through 2006. One of the two incidents was reported as "unlikely" (#I0139876-001) and the other was reported as a "misuse" (#I013550-003). Based on all reported aquatic incidents for atrazine, none were reported in Texas and none of the incidents reported effects to aquatic-phase amphibians. Further information on all of the reported aquatic incidents for atrazine is provided in Section 4.4 and Appendix F. Uncertainties related to the use of incident information from the Ecological Incident Information System (EIIS) are also discussed in Appendix F.

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)

The results of the screening-level risk assessment for the Barton Springs salamander suggest the potential for direct acute adverse effects to freshwater invertebrates, based on the residential granular use of atrazine at 2 lb ai/A (assuming 1% overspray and 30% impervious surfaces). The acute RQ of 0.057 exceeds the listed species of 0.05; therefore, atrazine use related to residential granular applications has the potential to indirectly affect the Barton Springs salamander via reduction in the availability of sensitive aquatic invertebrate food items.

However, this analysis was based on the lowest LC₅₀ value of 720 μ g/L for the midge (*Chironomus* spp.). Consideration of all acute toxicity data for the midge shows a wide range of sensitivity within and between species of the same genus (2 orders of magnitude) with values ranging from 720 to >33,000 μ g/L. Although the midge is a component of the Barton Spring salamander's diet, this species reportedly consumes a wide range of freshwater invertebrates that also include ostracods, copepods, snails, amphipods, mayfly larvae, leeches, and adult riffle beetles. Available acute toxicity values for other freshwater invertebrates that are included in the Barton Spring salamander's diet (i.e., amphipods, leeches, and snails) are 5,700 μ g/L and higher.

The potential for atrazine to elicit indirect effects to Barton Springs salamanders via effects on food items is dependent on several factors including: (1) the potential magnitude of effect on freshwater invertebrate individuals and populations; and (2) the number of prey species potentially affected relative to the expected number of species needed to maintain the dietary needs of the Barton Springs salamander. Together, these data provide a basis to evaluate whether the number of individuals within a prey species is likely to be reduced such that it may indirectly affect the Barton Springs salamander. Table 5.5 presents acute RQs and the probability of individual effects for dietary items of the Barton Springs salamander including midges, amphipods, leeches, and snails. The species sensitivity distribution of all acute toxicity data for freshwater aquatic invertebrates tested is represented in Figure 4.1. This analysis considers only acute risk to aquatic invertebrate food items as chronic risk quotients for invertebrates were less than the Agency's LOC. Even at the upper bound of EECs (21-day EEC of 4.2 μg/L from Table 3.9) based on assumptions of 10% overspray and 10% impervious surfaces for the residential granular use scenario), the chronic RQ of 0.07 is well below the LOC.

Table 5.5. Summary of RQs Used to Assess Potential Risk to Freshwater Invertebrate Food Items of the Barton Springs Salamander

Barton Springs Salamander Food Item Species	Acute Toxicity Value Range (µg/L) (No. of Studies)	RQ Range (based on an EEC of 40 μg/L)	Probability of Individual Effect*	Risk Interpretation	
Midge	720 - >33,000 (5)	<0.01 - 0.057	Up to 1 in 4.55E+07	Atrazine may affect sensitive food items, such as the midge; however the low probability of an individual effect to the midge is not likely to indirectly affect the Barton Springs salamander via reduction in midge prey items.	
Amphipod	5,700 – 14,900			Based on low probability of individual effects and RQs that are	
Leech	>16,000 (1) <0.01	< 0.01	<1 in	well below acute LOCs, atrazine is not likely to indirectly affect the	
Snail	>16,000 (1)		1.46E+18	Barton Springs salamander via reduction in amphipod, leech, or snail prey items.	

^{*}The probability of an individual effect was calculated using a probit slope of 4.4, which is the only technical grade atrazine value, reported in the available freshwater invertebrates studies that may serve as food items for the salamander; 95% confidence intervals could not be calculated based on the available data (Table A-18).

As shown in Table 5.5, the listed species LOC is exceeded for the midge (RQ = 0.057), based on the LC₅₀ value of 720 μ g/L. However, acute RQs based on the other acute toxicity data for the midge are \leq 0.04, less than the acute risk to endangered species LOC. Sufficient dose-response information was not available to allow for an estimation of the probability of an individual effect on the midge. Therefore, the probability of an individual effect was calculated using a probit dose response curve slope of 4.4; this is

the only slope for technical grade atrazine reported in available ecotoxicity data for freshwater invertebrates that are a component of the Barton Springs salamander's diet (amphipod; MRID # 452029-17). Based on a probit slope of 4.4, the probability of an individual mortality to the midge at an RQ of 0.057 is approximately 1 in 45.5 million (2.2E-08%).

Acute LOCs are not exceeded for the other dietary items of the Barton Springs salamander including the amphipod, leech or snail, based on the residential granular use EEC (assuming 1% overspray and 30% impervious surfaces). In addition, acute RQs based on the upper bound residential granular peak EEC of 67.1 µg/L (assuming 10% overspray and 10% impervious surfaces) are also less than acute LOCs for these food items in the salamander's diet.

Based on the non-selective nature of feeding behavior in the Barton Springs salamander and low magnitude of anticipated individual effects to all evaluated prey species, atrazine is not likely to indirectly affect the Barton Springs salamander via a reduction in freshwater invertebrate food items. This finding is based on insignificance of effects (i.e., effects to freshwater invertebrates are not likely to result in "take" of a single Barton Springs salamander) and discountability (i.e., the effect to freshwater invertebrates is extremely unlikely to occur given the estimated individual event probability of 1 in 45.5 million). Therefore, the effects determination for the assessment endpoint of indirect effects on the Barton Springs salamander via direct effects on prey (i.e., freshwater invertebrates) is "may affect, but not likely to adversely affect."

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

Direct adverse effects to non-vascular aquatic plants are possible, based on all modeled atrazine uses within the action area. In addition, direct effects to vascular plants are possible, based on the residential granular use of atrazine. Based on these direct effects, atrazine may indirectly affect the Barton Springs salamander via direct effects on aquatic plants. Therefore, the time-weighted EECs (for 14-day, 30-day, 60-day, and 90-day averages) were compared to their respective time-weighted threshold concentrations to determine whether potential effects to individual plant species would likely result in community level effects. As discussed in Section 4.2, concentrations of atrazine from the exposure profile at a particular use site and/or action area that exceed any of the following time-weighted threshold concentrations indicate that changes in the aquatic plant community structure could be affected:

- 14-day average = $38 \mu g/L$
- 30-day average = $27 \mu g/L$
- 60-day average = $18 \mu g/L$
- 90-day average = $12 \mu g/L$

A comparison of the 14-, 30-, 60-, and 90-day EECs for the Barton Springs salamander with the atrazine threshold concentrations representing potential aquatic community-level effects is provided in Table 5.6.

Table 5.6. Summary of Modeled Scenario Time-Weighted EECs with Threshold Concentrations for Potential Community-Level Effects

W 6	14-day		30-day		60-day		90-day	
Use Scenario	EEC (μg/L)	Threshold Conc. (µg/L)	EEC (μg/L)	Threshold Conc. (µg/L)	EEC (μg/L)	Threshold Conc. (µg/L)	EEC (μg/L)	Threshold Conc. (μg/L)
Res. (granular) (1% OS; 30% IS) / (10% OS; 10% IS)	3.5 / 6.0		1.9 / 3.2		1.2 / 2.0		1.0 / 1.5	
Res. (liquid)	2.5		1.5		1.0		0.8	
Turf (granular)	2.0	38	1.2	27	0.8	18	0.7	12
Turf (liquid)	1.7		1.0		0.7		0.6	
Rights-of- way	1.1		0.8		0.6		0.6	
Fallow/ Idle land	1.0		0.7		0.6		0.6	

OS = overspray

IS = impervious surfaces

Based on the results of this comparison, predicted 14-, 30-, 60-, and 90-day EECs for all modeled atrazine use scenarios (including the upper bound residential granular use EECs assuming 10% overspray and 10% impervious surfaces) are well below the threshold concentrations representing community-level effects. Although atrazine use may directly affect individual aquatic plants in Barton Springs, its use within the action area is not likely to adversely affect the Barton Springs salamander via indirect community-level effects to aquatic vegetation. This finding is based on insignificance of effects (i.e., community-level effects to aquatic plants are not likely to result in "take" of a single Barton Springs salamander) Therefore, the effects determination for the assessment endpoint of indirect effects on the Barton Springs salamander via direct effects on habitat and/or primary productivity of aquatic plants is "may affect, but not likely to adversely affect."

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 an order of magnitude above the highest detection found in any of the four springs. In general, the monitoring data should be considered a lower bound on exposure, while modeling represents an upper bound. Factors influencing the over-estimation of exposure include the assumption of no degradation, dilution, or mixing in the subsurface transport from edge of field to springs. The modeling exercise conservatively assumes that the spring and atrazine application site are adjacent. In reality, there are likely to be processes at work which cannot be accounted for in the modeling that will reduce the predicted exposures. In addition, the impact of setbacks on runoff estimates has not been quantified, although these buffers, especially those that are well-vegetated, are likely to result in significant reduction in runoff loading of atrazine.

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 Impact of Vegetative Setbacks on Runoff

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

6.1.3 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.

Uncertainty 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 atrazine is applied, the percent of a watershed that is cropped, and the percent of crops in that watershed that are actually treated with atrazine 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 atrazine, 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 are not available for salamanders; therefore, freshwater fish are used as surrogate species for aquatic-phase amphibians including salamanders. The available open literature information on atrazine 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. 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 Acute freshwater invertebrate toxicity data for the midge

The initial acute risk estimate for freshwater invertebrates was based on the lowest toxicity value from *Chironomus* studies, which showed a wide range of sensitivity within and between species of the same genus (2 orders of magnitude). Therefore, acute RQs based on the most sensitive toxicity endpoint for freshwater invertebrates may represent an overestimation of potential direct risks to freshwater invertebrates and indirect effects to the Barton Springs salamander via a reduction in available food.

6.2.4 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 atrazine. Additional environmental stressors may decrease the Barton Spring salamander's sensitivity to the herbicide, although there is the possibility of additive/synergistic reactions. Timing, peak concentration, and duration of exposure are critical in terms of evaluating effects, and these factors 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.

6.2.5 Use of threshold concentrations for community-level endpoints

For the purposes of this endangered species assessment, threshold concentrations are used to predict potential indirect effects (via aquatic plant community structural change) to the Barton Springs salamander. The conceptual aquatic ecosystem model used to develop the threshold concentrations is intended to simulate the ecological production dynamics in a 2nd or 3rd order Midwestern stream; however, the model has been correlated to microand mesocosm studies, which were derived from a wide range of experimental studies (i.e., jar studies to large enclosures in lentic and lotic systems), that represent the best available information for atrazine-related community-level endpoints.

The threshold concentrations are predictive of potential atrazine-related community-level effects in aquatic ecosystems, such as Barton Springs, where the species composition may differ from those included in the micro- and mesocosm studies. Although it is not possible to determine how well the responses observed in the micro- and mesocosm studies reflect the Barton Springs aquatic community, estimated high-end atrazine exposure concentrations in the action area (from modeled EECs) are predicted to be between 10 to 30 times lower than the community-level threshold concentrations, depending on the modeled atrazine use and averaging period. Given that threshold concentrations were derived based on the best available information from available community-level data for atrazine, these values are intended to be protective of the aquatic community, including the Barton Springs salamander. Additional uncertainties associated with use of the screening thresholds to estimate community-level effects are discussed in Section B.8 of Appendix B.

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 the midge (one of the dietary food items of the Barton Springs salamander). Acute ecotoxicity data from the midge was used to derive RQs for freshwater invertebrates. Based on a lack of dose-response information for the midge, the probability of an individual effect was calculated using the only probit dose response curve slope value reported in available freshwater invertebrate ecotoxicity data for technical grade atrazine. Therefore, a probit slope value of 4.4 for the amphipod, which is also a component of the Barton Springs salamander's diet, was used to estimate the probability of an individual effect on the freshwater invertebrates. It is unclear whether the probability of an individual effect for freshwater invertebrates other than amphipods would be higher or lower, given a lack of dose-response information for other freshwater invertebrate species. However, the assumed probit dose response slope for freshwater

invertebrates of 4.4 would have to decrease to approximately 1 to 2 to cause an effect probability ranging between 1 in 10 and 1 in 100, respectively, for freshwater 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 atrazine 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.

Table 7.1. Effects Determination Summary for the Barton Springs Salamander

Assessment Endpoint	Effects determination	Basis for Determination
Survival, growth, and reproduction of Barton Springs salamander individuals via direct effects	No effect	No acute and chronic LOCs are exceeded.
Indirect effects to Barton Springs salamander via reduction of prey (i.e., freshwater invertebrates)	May affect, but not likely to adversely affect	Acute LOCs are exceeded based on the most sensitive ecotoxicity value for the midge; however RQs for other dietary items (amphipods, leeches, snails) are less than LOCs. Based on the non-selective nature of feeding behavior in the Barton Springs salamander and low magnitude of anticipated individual effects to all evaluated prey species, atrazine is not likely to indirectly affect the Barton Springs salamander via a reduction in freshwater invertebrate food items. This finding is based on insignificance of effects (i.e., effects to freshwater invertebrates are not likely to result in "take" of a single Barton Springs salamander) and discountability (i.e., the effect to freshwater invertebrates is extremely unlikely to occur given the estimated individual event probability of 1 in 45.5 million).
Indirect effects to Barton Springs salamander via reduction of habitat and/or primary productivity (i.e., aquatic plants)	May affect, but not likely to adversely affect	Although atrazine use may directly affect individual vascular and non-vascular aquatic plants in Barton Springs, its use within the action area is not likely to adversely affect the Barton Springs salamander via indirect community-level effects to aquatic vegetation. Predicted 14-, 30-, 60-, and 90-day EECs for all modeled atrazine use scenarios within the action area are well below the threshold concentrations representing community-level effects. This finding is based on insignificance of effects (i.e., community-level effects to aquatic plants are not likely to result in "take" of a single Barton Springs salamander).

8. References

- Abou-Waly, Hoda, M. M. Abou-Setta, H. N. Nigg and L. L. Mallory. 1991.

 Growthresponse of freshwater algae, *Anabaena flos-aquae* and *Selenastrum capricornutum* to Atrazine and hexazinone herbicides. Bull. Environ. Contam. Toxicol. 46:223-229
- Allran, J. W. and Karasov, W. H. 2001. Effects of Atrazine on Embryos, Larvae, and Adults of Anuran Amphibians. *Environ.Toxicol.Chem.* 20: 769-775. EcoReference No.: 59251.
- Armstrong, D. E., C. Chester and R. F. Harris. 1967. Atrazine hydrolysis in soil. Soil Sci. Soc. Amer. Proc. 31:61-66.
- Bartell, S.M., G. Lefebvre, G. Aminski, M. Carreau, and K.R. Campbell. 1999. An ecosystem model for assessing ecological risks in Quebec rivers, lakes, and reservoirs. Ecol. Model. 124:43-67.
- Bartell, S.M., K.R. Campbell, C.M. Lovelock, S.K. Nair, and J.L. Shaw. 2000. Characterizing aquatic ecological risk from pesticides using a diquat dibromide case study III. Ecological Process Models. Environ. Toxicol. Chem. 19(5):1441-1453.
- Beliles, R. P. and W. J. Scott, Jr. 1965. Atrazine safety evaluation on fish and wildlife Bobwhite quail, mallard ducks, rainbow trout, sunfish, goldfish): Atrazine: Acute toxicity in rainbow trout. Prepared by Woodard Res. Corp.; submitted by Ciba-Geigy Corp., Greensboro, NC. (MRID No. 000247-16).
- Boone, M. D. and James, S. M. 2003. Interactions of an Insecticide, Herbicide, and Natural Stressors in Amphibian Community Mesocosms. *Ecol.Appl.* 13: 829-841. EcoReference No.: 81455.
- Brune, G. 1981. Springs of Texas, Volume I. Branch-Smith, Inc., Fort Worth, Texas.
- Capel, P. D., M. Lin and P. J. Wotzka. 1994. Pesticides in rains in Minnesota 1991-1993: An interim report. Minnesota Dept. Agr., Agronomy Serv. Div., p. 26. (Additional pages on 1994 data).
- Caux, Pierre-Yves, L. Menard, and R.A. Kent. 1996. Comparative study of the effects of MCPA, butylate, atrazine, and cyanazine on *Selenastrum apricornutum*. Environ. Poll. 92(2):219-225.
- Chippindale, P.T. 1993. Evolution, phylogeny, biogeography, and taxonomy of Central Texas spring and cave salamanders, *Eurycea* and *Typhlomolge* (Plethodontidae: Hemidactyliini). Dissertation, University of Texas at Austin, Austin, Texas.

- City of Austin (COA). 2003a. Unpublished Land Use Data. http://www.ci.austin.tx.us/landuse/. Accessed 15 February 2005.
- City of Austin. 2003b. Land Use Data (ftp://coageoid01.ci.austin.tx.us/GIS-Data/Regional/coa_gis.html).
- City of Austin Parks and Recreation Department (PARD) Integrated Pest Management Plan (IPM). 2005. April 27, 2005.
- Davis, B. 2006. County Extension Agent, Texas Cooperative Extension (Hays County) Personal Communication.
- DeAngelis, D.L., S.M. Bartell, and A.L. Brenkert. 1989. Effects of nutrient recycling and food-chain length on resilience. Amer. Nat. 134(5):778-805.
- DeLong-Amaya, A. 2006. Ladybird Johnson Wildflower Center, Personal Communication.
- Diana, S. G., Resetarits, W. J. Jr., Schaeffer, D. J., Beckmen, K. B., and Beasley, V. R. 2000. Effects of Atrazine on Amphibian Growth and Survival in Artificial Aquatic Communities. *Environ.Toxicol.Chem.* 19: 2961-2967. EcoReference No.: 59818.
- Field, M. 2004. Forecasting Versus Predicting Solute Transport in Solution Conduits for Estimating Drinking-Water Risks. Acta Carsologica, XXXIII/II, pp 115-150.
- Forson, D. and Storfer, A. 2006. Effects of Atrazine and Iridovirus Infection on Survival and Life-History Traits of the Long-Toed Salamander (Ambystoma macrodactylum). *Environ.Toxicol.Chem.* 25: 168-173. EcoReference No.: 82033.
- Garcia, E. 2006. Natural Resources Conservation Service District Conservationist (Travis County), Personal Communication.
- Giddings, J.M., T.A. Anderson, L.W. Hall, Jr., R.J. Kendall, R.P. Richards, K.R. Solomon, and W.M. Williams. 2000. Aquatic Ecological Risk Assessment of Atrazine A Tiered Probabilistic Approach, A Report of an Expert Panel. Novartis Crop Protection, Inc., Greensboro, NC, Section 5.3.1.5, page 147, June 23, 2000.
- Gucciardo, L. S. 1999. The Use of Anuran Larvae to Determine Chronic and Acute Toxicological Effects from Exposure to Atrazine and Metolachlor. *Ph.D.Thesis, Iowa State Univ., Ames, IA* 164 p. EcoReference No.: 78286.
- Hauwert, N., Johns, D. Hunt, B., Beery, J., Smith, B., and Sharp, J.M., Jr. 2004. Flow Systems of the Edwards Aquifer Barton Springs segment Interpreted from tracing and associated field studies.: So. Texas Geol. Soc. /Austin Geol. Soc. Symposium on the Edwards Aquifer, San Antonio, TX, in press.

- Hecker, M., Park, J. W., Murphy, M. B., Jones, P. D., Solomon, K. R., Van der Kraak, G., Carr, J. A., Smith, E. E., Du Preez, L., Kendall, R. J., and Giesy, J. P. 2005. Effects of Atrazine on CYP19 Gene Expression and Aromatase Activity in Testes and on Plasma Sex Steroid Concentrations of Male African Clawed Frogs (Xenopus laevis). *Toxicol.Sci.* 86: 273-280. EcoReference No.: 79287.
- Hoberg, J. R. 1993. Atrazine technical: Toxicity to duckweed, (*Lemna gibba*). SLI Rep. No. 93-4-4755. Prepared by Springborn Laboratories, Inc., Wareham, MA.; submitted by Ciba-Geigy Corporation, Greensboro, NC. (MRID No. 430748-04).
- Johnson, I. C., Keller, A. E., and Zam, S. G. 1993. A Method for Conducting Acute Toxicity Tests with the Early Life Stages of Freshwater Mussels. *In: W.G.Landis, J.S.Hughes, and M.A.Lewis (Eds.), Environmental Toxicology and Risk Assessment, ASTM STP 1179, Philadelphia, PA* 381-396.
- Kaul, M., T. Kiely, and A.Grube. 2005. Triazine pesticides usage data and maps for cumulative risk assessment, D317992. Unpublished EPA report.
- Kaul, M. and J. Carter. 2005. Use Sites for Blanco, Hays, and Travis Counties in Texas for Atrazine, Simazine, Carbaryl, Metolachlor, Diazinon, and Prometon (D322226).
- Kaul, M., Kiely, T., Jones, A., and Widawsky, D. 2006. Atrazine, Simazine, Carbaryl, Metolachlor, Diazinon, and Prometon Usage for Blanco, Hays, and Travis Counties in Texas to Support the Barton Springs Endangered Species Assessment (D322226, D322266 & D322267).
- Macek, K. J., K. S. Buxton, S. Sauter, S. Gnilka and J. W. Dean. 1976. Chronic toxicity of atrazine to selected aquatic invertebrates and fishes. US. EPA, Off. Res. Dev., Environ. Res. Lab. Duluth, MN. EPA-600/3-76-047. 49 p. (MRID # 000243-77).
- Maher, B. 2005a. The Case of the Anomalous Springs. Using all the clues to solve a karst mystery. USGS presentation to the ESI France-UT Karst Workshop; February 8, 2005.
- Maher, B. 2005b. Research Hydrologist, U.S. Geological Survey, Texas District. Personal Communication.
- Majewski, M.S., and P.D. Capel. 1995. Pesticides in the Atmosphere Distribution, Trends, and Governing Factors: Ann Arbor Press, Chelsea, Michigan, 228 p.
- Majewski, M.S., W.T. Foreman, and D.A. Goolsby. 2000. Pesticides in the atmospheres of the Mississippi River Valley, Part I Rain. Sci. Tot. Environ. 248:201-212.

- Markwardt, D. 2006. Vegetation Management Specialist. Texas DOT, Maintenance Division, Personal Communication.
- Mason, J. 2006. Vegetation Management Specialist. Texas DOT, Maintenance Division, Personal Communication.
- McVeety, E. 2006. Pesticide Applicator at Zilker Park, City of Austin Parks and Recreation Department, Personal Communication.
- Moore, A. and Lower, N. 2001. The Impact of Two Pesticides on Olfactory-Mediated Endocrine Function in Mature Male Atlantic Salmon (Salmo salar L.) Parr. *Comp.Biochem.Physiol.B* 129: 269-276. EcoReference No.: 67727.
- Perez, C., 2006, District Conservationist (Hays County), Personal Communication.
- Pipkin, T. and M. Frech, editors. 1993. Barton Springs eternal. Softshoe Publishing, Austin, Texas.
- Rohr, J. R., Elskus, A. A., Shepherd, B. S., Crowley, P. H., McCarthy, T. M., Niedzwiecki, J. H., Sager, T., Sih, A., and Palmer, B. D. 2003. Lethal and Sublethal Effects of Atrazine, Carbaryl, Endosulfan, and Octylphenol on the Streamside Salamander (Ambystoma barbouri). *Environ.Toxicol.Chem.* 22: 2385-2392. EcoReference No.: 71723.
- Rohr, J. R., Elskus, A. A., Shepherd, B. S., Crowley, P. H., McCarthy, T. M., Niedzwiecki, J. H., Sager, T., Sih, A., and Palmer, B. D. 2004. Multiple Stressors and Salamanders: Effects of an Herbicide, Food Limitation, and Hydroperiod. *Ecol.Appl.* 14: 1028-1040. EcoReference No.: 81748
- Schulz, A., F. Wengenmayer and H. M. Goodman. 1990. Genetic engineering of herbicide resistance in higher plants. Plant Sci. 9:1-15.
- Shay, K. 2006. Water Quality Education Manager, City of Austin Watershed Protection and Development Review Board, Personal Communication.
- Slade, R. J., Jr., Dorsey, M. E., Stewart, S. L. 1986. Hydrology and Water Quality of the Edwards Aquifer Associated with Barton Springs in the Austin Area, Texas, U.S. Geological Survey Water Resources Investigations Report 86-4036.
- Stratton, G. W. 1984. Effects of the herbicide atrazine and its degradation products, alone and in combination, on phototrophic microorganisms. Bull. Environ. Contam. Toxicol. 29:35-42. (MRID # 45087401).
- Taylor, E.J., S.J. Maund, and D. Pascoe. 1991. Toxicity of four common pollutants to the freshwater macroinvertebrates *Chironomus riparius* Meigen (Insecta:

- Diptera) and *Gammarus pulex* (L.) (Crustacea: Amphipoda). Arch. Environ. Contam. Toxicol. 21:371-376. (MRID 452029-17).
- Torres, A. M. R. and L. M. O'Flaherty. 1976. Influence of pesticides on *Chlorella*, *Chlorococcum*, *Stigeoclonium* (Chlorophyceae), *Tribonema*, *Vaucheria* (Xanthophyceae) and *Oscillatoria* (Cyanophyceae). Phycologia 15(1):25-36. (MRID # 000235-44).
- U. S. Department of Agriculture (USDA), Natural Resources Conservations Service (NRCS). 2000. Conservation Buffers to Reduce Pesticide Losses. Natural Resources Conservation Service. Fort Worth, Texas. 21pp.
- USDA Soil Conservation Service. 1986. Urban Hydrology for Small Watersheds. Technical Release 55, 2nd edition, June 1986.
- U.S. Environmental Protection Agency (EPA). 1998. Guidance for Ecological Risk Assessment. Risk Assessment Forum. EPA/630/R-95/002F, April 1998.
- U.S. EPA. 2003a. Interim Reregistration Eligibility Decision (IRED) for Atrazine.

 Office of Pesticide Programs. Environmental Fate and Effects Division. January 31, 2003. http://www.epa.gov/oppsrrd1/REDs/0001.pdf
- U.S. EPA. 2003b. Revised Atrazine Interim Reregistration (IRED). Office of Pesticide Programs. Environmental Fate and Effects Division. October 31, 2003. http://www.epa.gov/oppsrrd1/REDs/0001.pdf
- U.S. EPA. 2003c. Ambient Aquatic Life Water Quality Criteria for Atrazine Revised Draft. Office of Water, Office of Science and Technology, Health and Ecological Criteria Division, Washington, D.C. EPA-822-R-03-023. October 2003.
- U.S. EPA. 2003d. White paper on potential developmental effects of atrazine on amphibians. May 29, 2003. Office of Pesticide Programs, Washington D.C. Available at http://www.epa.gov/scipoly/sap.
- U.S. EPA. 2003e. Atrazine MOA Ecological Subgroup: Recommendations for aquatic community Level of Concern (LOC) and method to apply LOC(s) to monitoring data. Subgroup members: Juan Gonzalez-Valero (Syngenta), Douglas Urban (OPP/EPA), Russell Erickson (ORD/EPA), Alan Hosmer (Syngenta). Final Report Issued on October 22, 2003.
- U.S. EPA. 2004. Overview of the Ecological Risk Assessment Process in the Office of Pesticide Programs. Office of Prevention, Pesticides, and Toxic Substances. Office of Pesticide Programs. Washington, D.C. January 23, 2004.
- U.S. EPA. 2005. Pesticide Root Zone Model (PRZM) Field and Orchard Crop Scenarios: Guidance for Selecting Field Crop and Orchard Scenario Input

- Parameters, Version II. Water Quality Tech Team, Environmental Fate and Effects Division, Office of Pesticide Programs, U. S. Environmental Protection Agency. Washington, D.C. 20460. April, 2005.
- U.S. EPA. 2006a. Cumulative Risk Assessment for the Chlorinated Triazines. Office of Pesticides Programs. EPA-HQ-OPP-2005-0481. Washington, D.C. March 28, 2006.
- U.S. EPA. 2006b. Memorandum from Special Review and Reregistration Division to Environmental Fate and Effects Division: Errata Sheet for Label Changes Summary Table in the January 2003 Atrazine IRED. June 12, 2006.
- U.S. Fish and Wildlife Service (USFWS). 2005. Barton Springs Salamander (*Eurycea sosorum*) Recovery Plan. Southwest Region, USFWS, Albuquerque, New Mexico.
- U.S. Fish and Wildlife Service (USFWS) and National Marine Fisheries Service (NMFS). 1998. Endangered Species Consultation Handbook: Procedures for Conducting Consultation and Conference Activities Under Section 7 of the Endangered Species Act. Final Draft. March 1998.
- USFWS/NMFS. 2004a. 50 CFR Part 402. Joint Counterpart Endangered Species Act Section 7 Consultation Regulations; Final Rule. FR 47732-47762.
- USFWS/NMFS. 2004b. Letter from USFWS/NMFS to U.S. EPA Office of Prevention, Pesticides, and Toxic Substances. January 26, 2004. (http://www.fws.gov/endangered/consultations/pesticides/evaluation.pdf).
- U.S. Geological Survey (USGS), National Mapping Division, Rocky Mountain Mapping Center. 2003. Edwards Aquifer Land Use / Land Cover. Denver, Colorado. Accessed online at http://www.tceq.state.tx.us/gis/lulc.html.
- Walsh, G. E. 1983. Cell death and inhibition of population growth of marine unicellular algae by pesticides. Aquatic Toxicol. 3:209-214. (MRID # 45227731).
- Ward, D., 2006, Travis County Highway Department, Personal Communication.
- Wieser, C. M. and T. Gross. 2002. Determination of potential effects of 20 day exposure of atrazine on endocrine function in adult largemouth bass (*Micropterus salmoides*). Prepared by University of Florida, Wildlife Reproductive Toxicology Laboratory, Gainesville, FL, Wildlife No. NOVA98.02e; submitted by Syngenta Crop Protection, Inc., Greensboro, NC. (MRID No. 456223-04).
- Zinn, N. and A. Jones. 2006. Application Timing and Refined Usage Data for Atrazine, Simazine, Metolachlor, and Prometon in Three Texas Counties (Blanco, Hays, and Travis), D322539.