Office of Prevention, Pesticides,
and Toxic Substances

Risks of Diazinon Use to the Federally Listed Endangered Barton Springs Salamander *(Eurycea sosorum)*

Pesticide Effects Determination

Environmental Fate and Effects Division
Office of Pesticide Programs
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1. Executive Summary

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

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

Based on use estimates provided from the Biological and Economic Assessment Division and from discussions with U. S. Department of Agriculture extension agents in the Austin, TX, area, diazinon is not used to any great extent in the vicinity of Barton Springs. However, current uses of diazinon are not prohibited in the Austin area.

Environmental fate and transport models were used to estimate high-end exposure values that could occur at the edge of use sites and in water in the Barton Springs action area as a result of potential agricultural and ornamental diazinon use in accordance with label directions. Modeled concentrations in the Barton Springs provide estimates of exposure that are intended to represent possible diazinon concentrations originating from all potential use sites. Transport of water containing diazinon could occur in surface water in the contributing zone and in the recharge zone predominantly from subsurface flow through the fractured karst limestone of the Edwards Aquifer. Estimated 1-in-10-year peak exposure values for the Barton Springs were aggregated from all potential use sites and used in risk estimation. Estimated peak exposure values were consistent with maximum concentrations reported in monitoring data taken in the springs. However, monitoring conducted in Barton Springs subsequent to the cancellation of all residential uses and the phase-out of many agricultural uses indicate that diazinon is below the level of detection even following high rain run-off events.

The highest potential exposure was predicted to occur from use of diazinon on ornamentals due to the unlimited number of applications allowed on the labels up to a practical limit of 26 applications (EPA Reg. No. 2935-408, 4581-392, 5905-248, 19713-91, 19713-492, 66222-9, 66222-10, 66222-103, 11556-123, 39039-3, 39039-6, 61483-78, 61483-80, and 61483-92). However, reduction of the number of applications to ornamentals allowed on the labels to only one would not reduce acute risk estimates for listed invertebrates to below the level of concern.

The assessment endpoints for the Barton Springs salamander include direct toxic effects on the survival, reproduction, and growth of the salamander itself, as well as indirect effects, such as reduction of the prey base and/or modification of its habitat. Direct effects to the Barton Springs salamander are based on toxicity information for freshwater fish, which are generally used as a surrogate for 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.

Degradates of diazinon include diazoxon and oxypyrimidine. Comparison of available toxicity information for oxypyrimidine indicates lesser aquatic toxicity than the parent for freshwater and estuarine/marine fish, invertebrates, and aquatic plants. However, diazoxon is more toxic than the parent compound. Because oxypyrimidine is not of greater toxicological concern than diazinon, concentrations of this degradate are not assessed further. Submitted environmental fate studies for diazinon do not identify diazoxon, as it does not form >10% of residues. Since diazoxon is relatively short-lived in the environment and its concentrations relative to the parent are expected to be low, this assessment focuses on parent diazinon alone. The assessment is considered protective though since the surrogate species (rrainbow trout) used to assess the direct acute toxicity of diazinon to the Barton Springs salamander is orders of magnitude more sensitive than similar data for aquatic-phase amphibians.

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) for Federally-listed endangered species to identify if diazinon use within the action area has any direct or indirect effect on the Barton Springs salamander. Based on estimated environmental concentrations for the currently registered uses of diazinon, RQ values are below the Agency's LOC for direct acute effects on the Barton Springs salamander; this represents a "no effect" determination. There is a potential to directly affect the Barton Springs salamander on a chronic exposure basis and through indirect effects to its invertebrate forage base. However, exposure data combined with likelihood of individual effect estimates indicate that both direct chronic effects on the salamander and potential indirect effects on the salamander's forage base are not likely to adversely affect (NLAA) the Barton Springs salamander. A summary of the risk conclusions and effects determination for the Barton Springs salamander is presented in **Table 1**. Based on these results, an informal consultation with the U. S. Fish and Wildlife Service under Section 7 of the Endangered Species Act should be initiated to seek concurrence with the NLAA determinations.

Table 1. Diazinon Effects Determination Summary for the Barton Springs Salamander.

Assessment Endpoint	Effects Determination	Basis for Determination
Acute mortality	No effect	Acute LOC is not exceeded based on the most sensitive surrogate freshwater vertebrate data.
Chronic survival, growth, and reproduction effects on Barton Springs salamander individuals via direct effects	May affect but not likely to adversely affect	Although there is uncertainty regarding the potential for chronic effects on growth since available chronic toxicity data fail to establish a definitive chronic NOEC, estimated environmental concentrations and monitoring data are sufficiently low to render the likelihood of chronic effects low and as such is considered discountable.
Indirect effects to Barton Springs salamander via reduction of prey (*i.e.*, freshwater invertebrates)	May affect but not likely to adversely affect	Acute risk to endangered species LOCs are exceeded based on the most sensitive aquatic invertebrates evaluated; however, the likelihood of individual effects is low and as such is considered discountable.
Indirect effects to Barton Springs salamander via reduction of habitat and/or primary productivity (*i.e.*, aquatic plants)	No effect	Diazinon use does not directly affect individual non-vascular aquatic plants in Barton Springs. Estimated peak EECs for all modeled diazinon use scenarios within the action area are well below the threshold concentration for aquatic, non-vascular plants. Although there are no toxicity data for aquatic vascular plants, the data for nonvascular aquatic plants and vascular terrestrial plants and the lack of any reported field incidents involving plants indicate that plants are less sensitive to diazinon than animals.

2. Problem Formulation

Problem formulation provides a strategic framework for the risk assessment. By identifying the important components of the problem, it focuses the assessment on the most relevant life history stages, habitat components, chemical properties, exposure routes, and endpoints. The structure of this risk assessment is based on guidance contained in 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 conducted consistent with settlement of the court case "*Center for Biological Diversity and Save Our Springs Alliance v. Leavitt, No. 1:04CV00126-CKK"* filed January 26, 2004. The purpose of this ecological risk assessment is to make an "effects determination," under Section 7(a) (2) of the Endangered Species Act, for the Barton Springs salamander *(Eurycea sosorum),* by evaluating the potential direct and indirect effects resulting from use of the insecticide diazinon (O,O-diethyl-O-2-isopropyl-4-methyl-6-pyrimidinyl-phosphorothioate) on the survival, growth, and/or reproduction of this federally listed 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).

As part of the "effects determination", the Agency will reach one of the following three conclusions regarding the potential for diazinon 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 LOCs for the Barton Springs salamander are not exceeded for direct effects, a "no effect" determination is made, based on diazinon's use within the action area. If, however, indirect effects are anticipated and/or estimated 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 diazinon 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 organisms. 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

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 pesiticide may be used. Product labels (also known as end-use labels) describe the formulation type, acceptable methods of application, approved used sites, and any restrictions on how applications may be conducted. This assessment involves an evaluation of risks to the salamander from potential uses of diazinon, in accordance with the approved product labels. The use of diazinon is termed "the action."

Diazinon was one of the most widely used insecticides in the U. S. for residential as well as agricultural pest control. However, a December 2000 agreement with the technical registrants terminated all indoor residential uses and phased out and cancelled all outdoor residential uses of diazinon by December 31, 2004. Additionally, all registrations for granular products, except use on lettuce in California and Arizona and two current Section 24c registrations for control of cranberry girdler in the Pacific Northwest were cancelled by 2005. Some mitigation measures were identified in the 2002 IRED but not implemented until January 2007, including deletion of aerial applications for all uses except on lettuce, cancellation of all seed treatment uses, and cancellation of foliar applications to all vegetable crops except honeydew melons in California to control leafhoppers. For most uses, only one application per growing season is allowed. Crops with dormant-season and in-season uses, *e.g.* stone fruits, are limited to a single application per season, for a total of two applications per year. Section 3 registrations on succulent beans, succulent peas, peppers, potatoes, and squash were cancelled by August 2004; watercress was phased out by 2006.

Oxypyrimidine is the primary degradate of diazinon and is seen in both the laboratory studies and field studies. Diazoxon, an intermediate degradate which degrades further to oxypyrimidine, was detected at low levels in field dissipation studies, but was not reported to be a major degradate in laboratory studies. In monitoring studies in California, diazoxon has been detected in air and precipitation samples. Comparison of available toxicity information for the degradates of diazinon indicates that oxypyrimidine is practically nontoxic to aquatic (fish and invertebrates) and terrestrial animals (birds) on an acute exposure basis and it is practically nontoxic to terrestrial animals (birds) on a subacute dietary exposure basis. Diazoxon, a relatively short-lived degradate, has similar toxicity to that of the parent and is very highly toxic to birds on an acute oral exposure basis and is highly toxic to birds on a subacute dietary exposure basis; diazaoxon is highly toxic to aquatic-phase amphibians. A detailed summary of the available ecotoxicity information for the diazinon degradates is presented in **Appendix A**.

2.3 Previous Assessments

2.3.1 Diazinon

The Agency completed a screening-level ecological risk assessment for diazinon use in February 2000 (U.S. EPA 2002). This assessment was based on laboratory ecotoxicological data submitted by the registrant in support of reregistration and from data 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 diazinon. The results of the Agency's ecological assessments for diazinon are fully discussed in the July 31, 2006, Interim Reregistration Eligibility Decision (IRED) (U.S. EPA 2006).

Because the Agency had determined that diazinon shares a common mechanism of toxicity with the structurally-related organophosphate insecticides, it is included in a preliminary cumulative human health risk assessment for the organophosphate pesticides which was developed in 2000.

2.3.2. Barton Springs Salamander

The Agency has also completed (U. S. EPA 2006) an ecological risk assessment evaluating the potential effects of the herbicide atrazine on the Barton Springs salamander. The atrazine assessment was another component of the settlement of the court case "*Center for Biological Diversity and Save Our Springs Alliance v. Leavitt, No. 1:04CV00126-CKK"*. Conclusions regarding atrazine use in its action area were that it would have no direct effect on the Barton Springs salamander's growth, reproduction or survival; furthermore, atrazine was not likely to indirectly affect the salamander through adverse effects on the salamander's prey or through adverse effects on aquatic plants.

2.4 Stressor Source and Distribution

2.4.1 Environmental Fate and Transport Assessment

The following fate and transport description for diazinon is consistent with the information contained in the initial 2002 IRED (U.S. EPA, 2002). Diazinon is mobile and moderately persistent in the environment. As shown in **Table 2** it degrades by microbial metabolism as well as the abiotic processes of hydrolysis and photolysis. Aerobic soil metabolism half-lives were 37 and 38 days in two laboratory studies. No acceptable anaerobic microbial metabolism data were submitted. Hydrolysis half-lives were 12, 138 and 77 days at pH's 5, 7 and 9 respectively. Photolysis occurred with half-lives of 17 to 37 hours on soil and 37 days in aqueous solution. The dominant degradation process is expected to depend on environmental conditions.

Diazinon is relatively mobile in soil, as Freundlich partition coefficients estimated from batch equilibrium studies ranged from 3.7 (1/n=0.60) to 23.4 (1/n=0.93) in sandy and loamy soils and were 114 (1/n=0.70) in an unclassified soil rich in organic carbon. However, Freundlich exponents were often less than 0.9. Diazinon binding in soil is correlated with organic carbon content, with a K_{OC} range of 439 to 854 L/kg_{oc}. Italian researchers reported that in 25 soils tested, R_f values indicate that diazinon was slightly mobile in 80% of soils tested and immobile in 20%. In saturated columns, diazinon was shown to leach in light textured soils with low organic matter (Arienzo *et al.*, 1994). In column leaching studies submitted to the Agency, diazinon residues which had been aged 30 days were shown to be mobile in columns of Lowell sand, Hanford sandy loam, Huntington loam and Armor silty clay soils.

Diazinon does volatilize, as indicated by its vapor pressure (1.40×10^{-4} torr at 20°C) and by detections in air, rain, and fog, as reported by USGS and other researchers and summarized by EPA in the IRED.

Field dissipation studies reported half-lives ranging from 5 to 20 days, which is consistent with the laboratory data. Studies were done with three different formulations (granular, wettable powder and emulsifiable concentrate) and there were no apparent differences in field dissipation among the three formulation types.

Table 2. General chemical properties and environmental fate parameters of stabilized diazinon.[1]

Chemical/Fate Parameter	Value	Source
Molecular mass	304.3	Product chemistry
Vapor pressure (20°C)	1.40×10^{-4} torr	U.S. EPA, 1988
Henry's Law Constant	1.40×10^{-6} atm m^3/mol	U.S. EPA, 1988
Water solubility (20°C)	40 mg/L	U.S. EPA, 1988
Octanol-to-water partition coefficient (K_{OW})	2.5×10^4	U.S. EPA, 1988
Freundlich soil-to-water partition coefficients (K_f) for adsorption (soil type)	5.6 (1/n = 0.63) (sand) 113.5 (1/n = 0.70) (unclassified) 11.7 (1/n = 0.77) (loam) 3.7 (1/n = 0.60) (sand) 4.5 (1/n = 0.55) (loamy sand) 23.4 (1/n = 0.93) (sandy clay loam)	MRID 00118032
Organic carbon normalized partition coefficients (K_{OC})[2]	439, 485, 560, 638, 720, 854 L/kg_{OC}	MRID 00118032
Hydrolysis half-lives (23-25°C)	12 d (pH 5) 138 d (pH 7) 77 d (pH 9)	MRID 40931101
Aqueous photolysis half-life	37 days	MRID 40863401
Soil photolysis half-life	17.3 hrs 37.4 hrs	MRID 00153229 MRID 00153230
Aerobic soil metabolism half-lives	37.4 days 38.0 days	MRID 40028701 MRID 44746001
Fish bioconcentration	542x (edible) 583x (viscera) 542x (whole fish)	MRID 40660808

[1] Some chemical properties of the stabilized technical diazinon used in product formulations differ from those of unstable technical diazinon.
[2] K_{OC} values were calculated based on K_f values for adsorption (*e.g.*, $K_{OC} = K_f$ (adsorption) ÷ % organic carbon).

The environmental fate characteristics of diazinon are consistent with those of compounds expected to occur in water resources. There is a considerable amount of evidence showing that diazinon occurs in both ground and surface water as a result of nonagricultural and agricultural uses, especially as a result of the residential uses which are no longer permitted.

Diazinon bioconcentrated to roughly 500x in bluegill tissue. Depuration was rapid with 96% removal after 7 days.

Oxypyrimidine (2-isopropy-6-methyl-4-pyrimidinol) is the primary degradate of diazinon and is seen in both the laboratory studies and field studies. While quantitative kinetic estimates of oxypyrimidine are not available, it appears to be more persistent than diazinon. In a soil column leaching study, oxypyrimidine was the most mobile residue and occurred as 39% to 53% of the applied in the leachate.

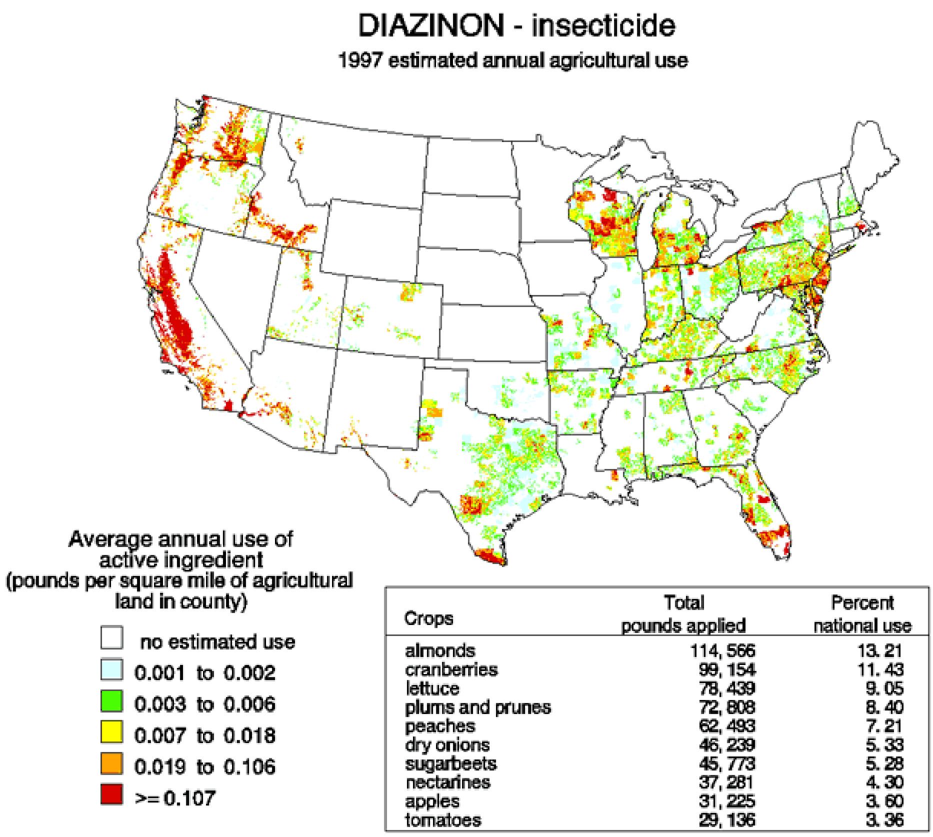
Diazoxon (O,O-diethyl-O-(2-isopropyl-4-methyl-6-pyrimidinyl)phosphonate), an intermediate degradate formed by hydrolysis, retains the organophosphate moiety of the parent compound and is a stronger cholinesterase inhibitor than parent diazinon. Diazoxon hydrolyzes rapidly to oxypyrimidine under most circumstances. Diazoxon was detected at low levels in field dissipation studies, but was not reported to be a major degradate in laboratory studies. Diazoxon has been also reported in air, rain, fog and surface waters. Schomburg *et al.* (1991) reported concentrations of diazinon and diazoxon measured in fog samples taken in California, with concentrations of diazinon and diazoxon ranging 150-4800 and 1900-11000 ng/L, respectively. Ratios of diazoxon to diazinon ranged 0.67-13, with the majority of the samples from 5 fog events indicating that diazoxon concentrations in fog were greater than the parent. The authors indicated that the ratios were greater in non-agricultural areas, when compared to agricultural areas. They indicated that it is possible that degradation of diazinon to diazoxon takes place while diazinon was present in the atmosphere or in the fog. Diazinon and diazoxon are then atmospherically transported from agricultural to non-agricultural areas. Glotfelty *et al.* (1990) also reported measured concentrations of diazinon and diazoxon in fog samples taken in California. The reported range of the diazoxon to diazinon concentrations during 6 fog events was 0.056-7.1, with the majority of the samples indicating that the parent concentration was greater than the degradate. The authors indicated that the degradation of diazinon in the atmosphere could be attributed to oxidation occurring during daylight hours, followed by uptake into the fog. The persistence of diazinon and diazoxon in the atmosphere and in precipitation is unknown.

2.4.2 Mechanism of Action

Organophosphate toxicity is based on the inhibition of the enzyme acetylcholinesterase which cleaves the neurotransmitter acetylcholine. Inhibition of acetylcholinesterase by organophosphate insecticides, such as diazinon, interferes with proper neurotransmission in cholinergic synapses and neuromuscular junctions.

2.4.3 Use Characterization

Nationally diazinon usage has been substantially curtailed since 2004. The pesticide is used to control foliage and soil insects and pests of many fruit, nut, vegetable, and ornamental crops as well as cattle. All residential uses have been cancelled. Approximately 4 million pounds of the active ingredient diazinon are used annually on agricultural sights. Use is highest on almonds and stone fruits. **Figure 1** presents the national distribution of annual diazinon use estimated between 1995 and 1998 (USGS 2007). This historical information is based on estimates that include uses that have been restricted and/or cancelled. Therefore, there has likely been a significant reduction in both the amount and distribution of diazinon use. Indoor residential uses were phased-out in 2002 while outdoor residential uses were phased-out in 2004.



Crops	Total pounds applied	Percent national use
almonds	114, 566	13. 21
cranberries	99, 154	11. 43
lettuce	78, 439	9. 05
plums and prunes	72, 808	8. 40
peaches	62, 493	7. 21
dry onions	46, 239	5. 33
sugarbeets	45, 773	5. 28
nectarines	37, 281	4. 30
apples	31, 225	3. 60
tomatoes	29, 136	3. 36

Figure 1. Historical (1997) Extent of Diazinon Use (lbs).

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

Currently, labeled uses of diazinon include several fruit, nut, and vegetable crops as well as cattle ear tags. There are 14 active Section 3 labels of products containing diazinon. The EPA registration numbers for these labels are 2935-408, 4581-392, 5905-248, 19713-91, 19713-492, 66222-9, 66222-10, 66222-103, 11556-123, 39039-3, 39039-6, 61483-78, 61483-80, and 61483-92. In addition, a SLN (TX-040026) is available for application of diazinon to several crops in TX only. A comprehensive list of these uses is included in **Table 3.**

Table 3. Specific sites on which diazinon is currently registered for use.

Category	Specific Crops
Fruit	Apples, apricots, blueberries, caneberries, cherries, cranberries, figs, nectarines, peaches, pears, pineapple, plums, prunes, strawberries
Nut	Almonds
Vegetable	Beans (succulent), beets (red), broccoli, Brussels sprouts, cabbage, carrots, cauliflower, collards, cucumbers*, endive, ginseng, kale, lettuce, melons, mustard, onions, parsley*, peas (succulent), peppers*, radishes, rutabagas, spinach, tomatoes
Other (non-agricultural)	Cattle ear tag, outdoor ornamentals
*SLN for TX only.	

There is potential use of diazinon contained in cattle ear tags within the action area. Ear tags may contain up to 6 grams of diazinon each (EPA Reg. No. 61483-80). Based on 2006 AgCensus data, the Barton Springs action area may contain 10,500 to 13,000 cows (USDA 2007). With two tags per cow replaced 1-2 times per year, there is the potential of over 1000 pounds of diazinon released into the action area per year (possible gradual release of 2.8 lbs a.i./day) due to this use. However, most of the diazinon released from cattle ear tags is expected to volatilize, adsorb to the cow or to soil, or degrade, such that exposure to water bodies is expected to be minimal. Current exposure modeling methodologies are not available to quantitatively assess exposures of diazinon originating from cattle ear tags. Therefore, this exposure route was not quantitatively assessed for potential risk to the salamander.

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 when 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**), located in Zilker Park near downtown Austin, Texas. Based on salamander survey results conducted by the City of Austin, Barton Springs salamanders appear to prefer areas near the spring outflows, with clean, loose substrate for cover, but may also be found in 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. This species has one of the smallest ranges of any vertebrate species in North America (Chippindale, 1993). The Barton Springs segment of the Edwards Aquifer and its contributing zone supply all of the water in the springs that make up the

Barton Springs complex. Flows of clean spring water are essential to maintaining well-oxygenated water necessary for salamander respiration and survival.

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

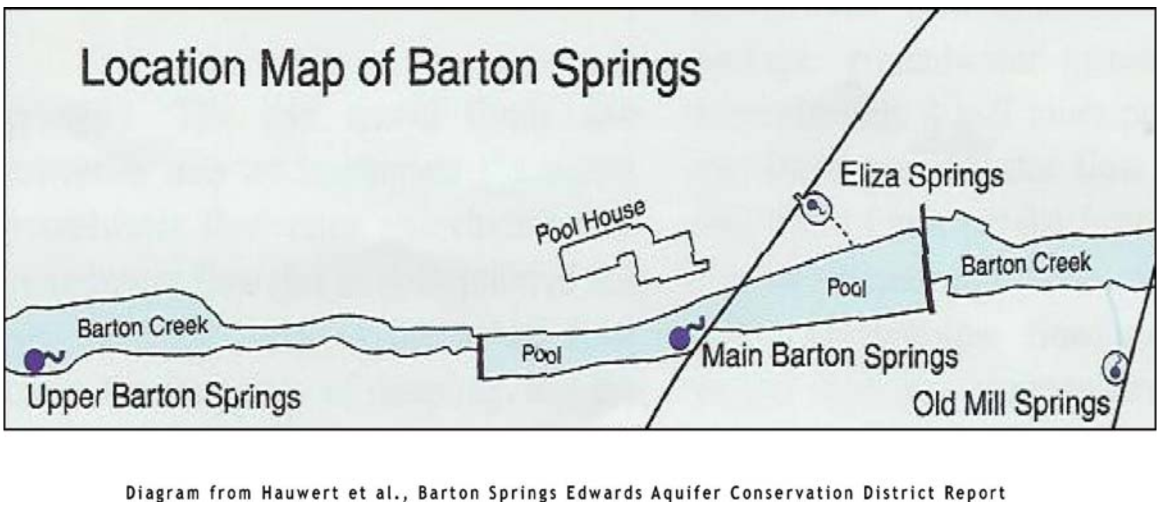


Figure 2. Barton Springs Complex (from Hauwert *et al*., 2005). Circles represent spring locations.

2.6 Action Area

It is recognized that the overall action area for the national registration of diazinon uses is likely to encompass considerable portions of the United States based on the large array of 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 from potential direct and indirect toxic effects of diazinon and from potential adverse effects on its habitat, 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 diazinon may be expected to have on the environment, the diazinon exposure levels that are associated with those effects, and the best available information concerning the use of diazinon and its fate and transport within Barton Springs.

Unlike exposure pathways for most aquatic organisms, where pesticides are potentially transported via surface water to the receptor within a defined watershed, the Barton Springs salamander resides in a somewhat unique environment in which the water and the diazinon reaches the salamander via subsurface flow. The Barton Springs salamander is known to inhabit only four springs and associated pools and subterranean areas in the aquifer itself (USFWS, 2005). Thus, the fate and transport of diazinon is an important factor in defining the action area for the Barton Springs salamander. The fate profile (see **Section 2.4.1**) indicates why runoff from treated fields, transported in ground water that flows 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 primarily defined by those areas within the hydrogeologic "watershed" that discharge to the springs. **Figure 3** depicts the extent of the action area based on this hydrogeologic framework.

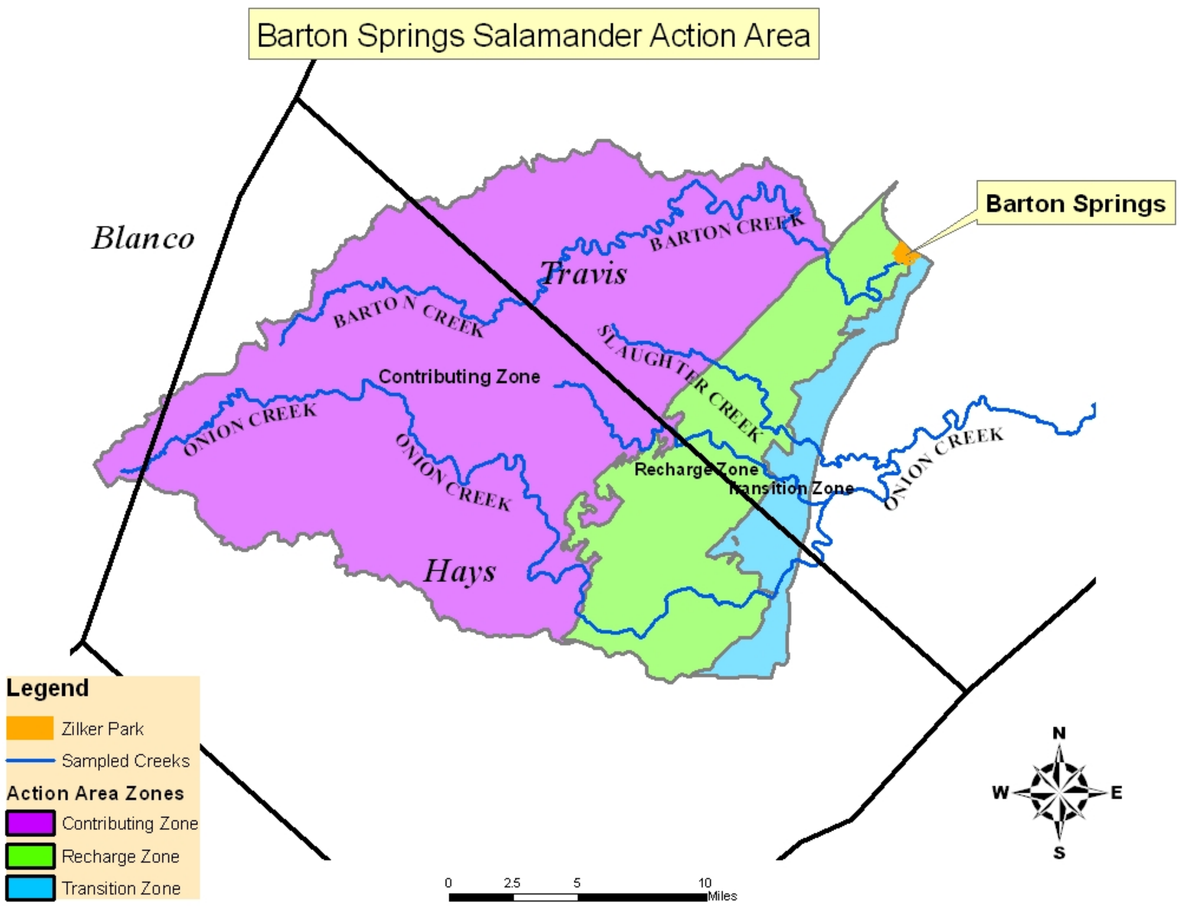


Figure 3. Action Area for Diazinon as it Relates to the Barton Springs Salamander.

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) (See **Figure 2**). Collective flow from this group of springs represents the fourth largest spring system in Texas (Brune, 1981). The springs are fed by the Barton Springs Segment of the Edwards Aquifer (BSSEA). During high flow conditions, the surface water flow from Barton Creek may enter the Barton Springs Pool, if it overtops the dam at the upper end of the pool. However, 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, this is not expected to be a significant source of water in the areas where the salamander resides. Thus, groundwater quality is the primary determinant of exposure for the salamander.

Flow to the Barton Springs is controlled by the geology and hydrogeology of the Barton Springs Watershed, which is divided into three hydrogeologic zones. These are, from west to east, the Contributing Zone (683 km^2), the Recharge Zone (233 km^2), and the Artesian Zone. Some have sub-divided the Recharge Zone further into the Recharge and Transition Zones (**Figure 3**). The BSSEA is comprised of the Recharge and Artesian zones (401 km^2). Of these zones, the Contributing and Recharge Zones have the greatest and most direct influence on Barton Springs. The Artesian Zone does not contribute subsurface flow to the springs (Slade *et al*., 1985, Hauwert *et al*., 2004). A more detailed description of the geology and hydrogeology of these zones is provided in **Section 3.2.2.**

Numerous geological and groundwater studies (Slade *et al.*, 1986, Hauwert *et al.*, 2004, Lindgren et al., 2004)) have been conducted that define the extent of the area contributing water to the Barton Springs. The Contributing Zone includes six creeks (Barton, Williamson, Slaughter, Bear, Little Bear, and Onion Creeks) that drain the watersheds and are maintained by spring flow from the Trinity aquifer. These creeks flow toward the Recharge Zone across the boundary of the Edwards aquifer. In the Recharge Zone, the creeks flow over the surface of the highly fractured and weathered limestone of the Edwards aquifer and rapidly infiltrate through the faults, caves, and sinkholes characteristic of a karst aquifer system. The Trinity aquifer is juxtaposed at depth against the Edwards aquifer and likely discharges into the Edwards aquifer, but this represents a minor portion of overall recharge (Lindgren, 2004).

Within the Recharge Zone of the BSSEA groundwater is rapidly transported toward the Barton Springs with velocities along the dominant flow path of 1-5 miles/day, depending on groundwater flow conditions (USFWS, 2005). Based on dye tracer studies, pesticides present within the recharge zone could potentially be transported to the springs on a time scale of hours to weeks (Hauwert *et al*., 2004).

An evaluation of usage information was completed to determine whether any or all of the area defined by the Barton Springs Watershed should be included in the Action Area. Current labels and local use information were reviewed to determine which diazinon uses could possibly be present within the defined area. These data suggest that limited agricultural and ornamental uses are present within the defined area. Finally, local land cover data (City of Austin, 2003a and b; USGS, 2003) were analyzed and interviews with the local agricultural sector (Davis, 2006; Garcia, 2006; Perez, 2006; see **Appendix B** for more detail) were conducted to refine the characterization of potential diazinon use in the areas defined by Hays, Travis, and Blanco counties. The overall conclusion of this analysis was that while certain agricultural and ornamental uses could not be excluded, the entire urbanized areas of Hays, Travis and Blanco counties could be excluded from the final action area based on usage and land cover data, since no residential uses of diazinon remain.

In addition to diazinon exposures from contaminated surface and groundwater, there is potential that transport of diazinon through spray drift and/or long-range atmospheric transport could contribute to concentrations in the aquatic habitat used by the salamander. The environmental fate profile of diazinon, coupled with available monitoring data, suggest that long range transport of volatilized diazinon cannot be precluded as a possible route of exposure to non-target organisms. The Agency does not currently have quantitative models to address the long range transport of pesticides from application sites. The extent of the Action Area that could hypothetically be influenced by this route of exposure is uncertain.

Based on the available information on potential diazinon use sites, none of the streams in the watersheds that are within the range of the Barton Spring salamander could be excluded from the action area. Therefore, the portion of the diazinon action area assessed here includes the area within the boundaries of the watersheds that contain the Barton Springs salamander. **Figure 3** depicts the action area graphically.

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" (USEPA 1992). Selection of the assessment endpoints is based on valued entities (*i.e*., Barton Springs salamander), the ecosystems potentially at risk (*i.e*, Barton Springs), the migration pathways of diazinon (*i.e*., runoff), and the routes by which ecological receptors are exposed to diazinon-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 guideline tests that are performed on a limited number of organisms. Given that 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. Consequently, fish are used as a surrogate for amphibian/salamanders, in accordance with guidance specified in the Agency's Overview Document (U.S. EPA, 2004). Specific assessment endpoints and measures of ecological effects considered in this assessment are defined in **Table 4**. Additional ecological effects data from the open literature, as identified by ECOTOX, were also considered.

Table 4. Summary of Assessment Endpoints and Measures of Ecological Effect.

Assessment Endpoint	Measures of Ecological Effect
1. Survival, growth, and reproduction of Barton Springs salamander individuals via direct effects	1a. Rainbow trout acute LC_{50} 1b. Brook trout chronic NOAEC
2. Survival, growth, and reproduction of Barton	2a. Waterflea acute EC_{50}

Assessment Endpoint	Measures of Ecological Effect
Springs salamander individuals via indirect effects on prey (*i.e.*, freshwater invertebrates)	2b. Waterflea chronic NOAEC 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 Springs salamander individuals via indirect effects on habitat and/or primary productivity (*i.e.*, aquatic plant community)	3a. Non-vascular plant (freshwater algae) acute EC_{05}

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 diazinon to the environment. Based on the results of the 2002 diazinon IRED (U.S. EPA, 2006), and considering the possibility that diazinon has the potential for long-range transport, the following risk hypotheses are presumed for this endangered species assessment:

- Diazinon in groundwater, runoff, spray drift and/or atmospheric deposition from treated areas may directly affect Barton Springs salamanders by causing mortality or adversely affecting growth or fecundity;
- Diazinon in groundwater, runoff, spray drift and/or atmospheric deposition from treated areas may indirectly affect Barton Springs salamanders by reducing or changing the composition of prey populations; and
- Diazinon in groundwater, runoff, spray drift and/or atmospheric deposition 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 diazinon on the Barton Springs salamander is shown in **Figure 4**.

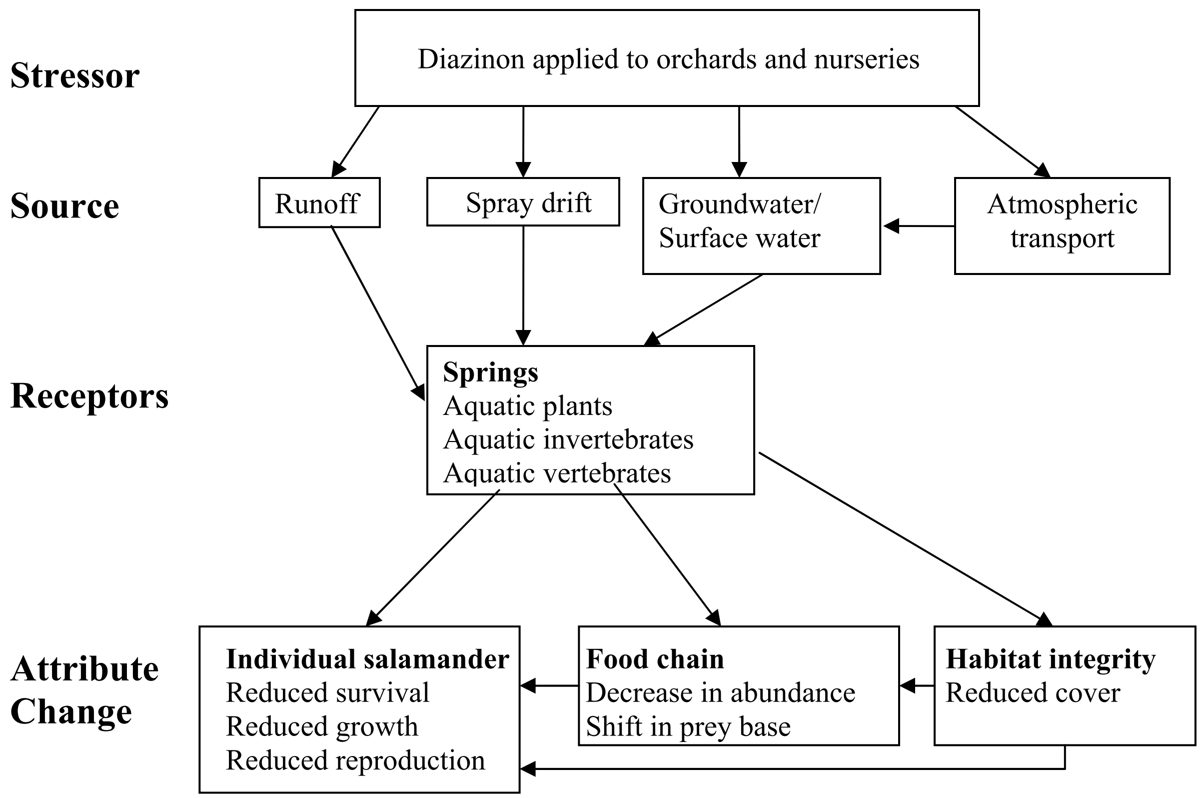


Figure 4. Conceptual Model Depicting Potential Risk from Diazinon Use to the Barton Springs Salamander.

The conceptual model provides an overview of the expected exposure routes for Barton Springs salamander within the action area. In addition to freshwater aquatic vertebrates including Barton Springs salamanders, other aquatic receptors of concern that may be potentially exposed to diazinon 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 diazinon may indirectly affect the Barton Springs salamander via reduction in food and habitat availability. The available data indicate that diazinon is not likely to bioconcentrate in aquatic food items, with fish bioconcentration factors (BCFs) ranging from 542 to 583 and rapid depuration in 7 days (MRID 40660808). Therefore, bioconcentration of diazinon in salamanders via the diet is not likely to be a concern.

Individual Barton Springs salamanders with the greatest potential to experience direct adverse effects from diazinon use are those that occur in surface water and/or groundwater with the highest concentrations of diazinon. Water passing into, and through Barton Springs comes from groundwater in the BSSEA. 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, diazinon has not been used in this area (personal communication with Elizabeth McVeety, pesticide applicator at Zilker Park, April 21, 2006).

The source and mechanism of release of diazinon into surface and groundwater are ground applications via foliar spray to agricultural sites and on ornamentals. Surface water runoff from the areas of diazinon application is assumed to follow topography, resulting in direct runoff to Barton Creek and/or runoff to the recharge area of the BSSEA, where it becomes groundwater that discharges to the Barton Springs. Additional potential exposure routes include spray drift and atmospheric transport as a result of volatilization. However, spray drift is not considered to be a significant route of exposure because the source area for diazinon is generally removed from the spring system where the salamander resides, and the diazinon exposures that reach the springs do so via subsurface flow.

Besides exposures of diazinon resulting from runoff and subsequent aqueous transport to the salamander's habitat, exposure of the salamander to diazinon through atmospheric transport and deposition is possible (Stein and White 1993; Majewski and Baston 2002). As described in the Diazinon IRED, diazinon and its degradate diazoxon can be present in air or precipitation (*e.g.* rain and fog) due to spray drift, volatilization from application sites and/or wind erosion of soil containing residues (Unsworth *et al*. 1999). Wet (precipitation) and dry (particulate matter) deposition could contribute to diazinon and diazoxon loads in aquatic systems (LeNoir *et al*. 1999; USGS 2003a); however, diazinon is most likely to be deposited in wet rather than dry deposition (Majewski et al. 2006).

At this time, EFED does not have an approved model for estimating atmospheric transport of pesticides and resulting exposure to aquatic organisms in areas receiving pesticide deposition from the atmosphere. Potential mechanisms of transport of diazinon to the atmosphere, such as volatilization, wind erosion of soil, and spray drift, can only be discussed qualitatively. Given the presence of diazinon in air and precipitation reported in monitoring data, it is possible that diazinon is present in air and precipitation in the Barton Springs area. However, the majority of monitoring data for diazinon relate to areas with significantly different use patterns than those found in Southern Texas. In particular, available monitoring data are generally relevant to California, which has greater use of diazinon than Texas. Given a lack of appropriate modeling and relevant monitoring data, contributions of atmospheric transport and subsequent deposition of diazinon to the exposure of the salamander are not considered quantitatively in this assessment. Qualitative discussions involving transport mechanisms and national monitoring data for diazinon concentrations in air and precipitation are discussed in the uncertainty section of this document.

3. Exposure Assessment

3.1 Label Application Rates and Intervals

In the 2002 IRED, EPA stipulated numerous changes to the use of diazinon including label restrictions and other mitigation measures designed to reduce risk to human health and the environment (U.S. EPA 2006). Specifically pertinent to this assessment, the Agency terminated all indoor residential uses and phased out all outdoor residential uses. Technical registrants were required to reduce the amount of diazinon they produced by 50% or more by 2003. As of December 31, 2004, it was unlawful to sell outdoor, non-agricultural diazinon products in the United States, including all outdoor home, lawn, and garden products.

Other mitigation measures were identified but not implemented until January 2007, including cancellation of all granular registrations, deletion of aerial applications for all uses except on lettuce, cancellation of all seed treatment uses, and cancellation of foliar applications to all vegetable crops except honeydew melons in California to control leafhoppers. For most uses, only one application per growing season is allowed. Crops with dormant-season and in-season uses (*e.g.* stone fruits) are limited to a single application per season, for a total of two applications per year. On all orchard crops with dormant season uses, label language has been added recommending that applications be made every other year unless pest pressure are such that consecutive annual treatments are necessary.

Diazinon is formulated as granular, liquid, wettable powder, and dry flowable formulations. Application equipment for the agricultural uses include those for ground application (the most common application method), aerial, band treatment, incorporated treatment, and various sprayers (low-volume, hand held, directed), and spreaders for granular applications.

The Use Characterization section (**Section 2.4.3**) of this assessment indicates that the only labeled uses that are expected to potentially result in exposures from runoff to the Barton Springs Salamander are nectarines, peaches, and outdoor ornamentals. **Table 5** lists the pertinent label application information for these uses. Peach uses were used to represent nectarine uses for aquatic exposure modeling because the label application information for each use is the same. As current labels do not provide maximum numbers of applications for outdoor ornamental uses, a practical limit of 26 applications per year (due to the 14-day minimum application interval) was assumed for these uses (EPA Reg. No. 4581-392, 5905-248, 19713-91, 19713-492, 66222-9, 66222-10, 66222-103).

Table 5. Maximum Labeled Use Patterns of Diazinon in the Action Area of the Barton Springs Salamander Endangered Species Assessment.

Use Site	Maximum Application Rate (lbs a.i./acre)	Maximum Number of Applications per Year	Method of Application	Minimum Interval Between Applications (days)
Ornamentals	1.0	26	Foliar spray	14
Peaches/ Nectarines	2.0	2	Ground spray/ Foliar spray	Undefined period between dormancy and pest infestation

3.2 Aquatic Exposure Assessment

This 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, using a combination of simulation modeling and monitoring data collected in the BSSEA action area. The Agency's Pesticide Root Zone Model (PRZM, v3.12beta, May 24, 2001) was used to provide estimates of exposure in the Barton Springs resulting from direct transport in runoff water to streams in the contributing zone and resultant recharge and subsurface flow through the fractured limestone of the Edwards Aquifer. 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.

Available historical monitoring data from the spring systems and groundwater wells in the action area were evaluated. While of high quality, targeted to the Barton Springs system, and in selected instances targeted to pesticide use and single runoff events, the historical monitoring data are likely to miss peak concentrations due to insufficient sample frequency. Therefore, the monitoring data are useful for long duration (annual average) estimates of exposure, but they are not considered robust in terms of estimating acute or intermediate duration (14-day, 21-day, 30-day, 60-day, or 90-day average) exposures.

The highest potential exposure was predicted to occur from use of diazinon on outdoor ornamentals within the recharge zone. The exposure assessment yields modeled peak and annual average 1-in-10-year aggregate exposure estimates that are consistent with concentrations seen in the monitoring data.

3.2.1 Background

The Barton Springs salamander resides in a geographically limited area defined by a set of spring-fed pools within the city of Austin, Texas. These pools represent the total areal 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; approximately 80% of the flow originates from the Main Spring. All of the springs are fed via subsurface flow originating in the fractured limestone of the Edwards Aquifer, 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 in the Barton Springs Watershed. 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 Watershed defines the action area (see **Section 2.6)** of diazinon use for the Barton Springs salamander. Several hydrogeologic zones define the watershed. From west to east, these are the Contributing Zone, the Recharge Zone (which some divide further into Transition and Recharge zones), 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 hydrogeology of the system. The Contributing Zone and the Recharge Zone contribute the majority of the water to the Barton Springs pool systems. Therefore, land use patterns within these zones were considered to determine the potential for diazinon exposure to the Barton Springs salamander. **Figure 5** shows the extent of the Barton Springs Watershed.

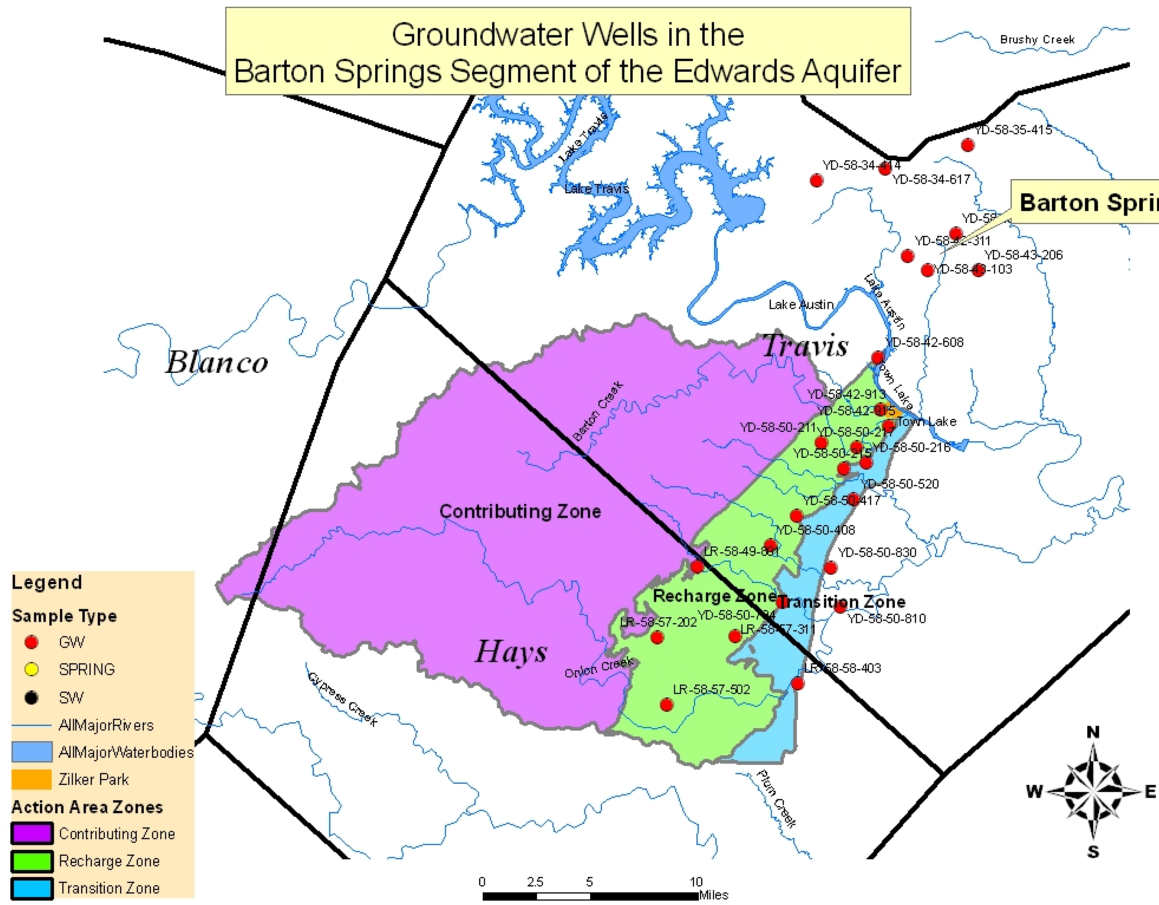


Figure 5. Hydrologic zones of the Barton Springs Watershed.

Groundwater flow within the Recharge Zone is dominated by subsurface flow through fractures and solution features of a portion of the limestone Edwards aquifer known as the BSSEA. Numerous studies have been conducted which document the nature of the subsurface geology and the nature and extent of groundwater flow *(Slade et al.*, 1986; *Hauwert et al.*, 2004; *Mahler*, 2005, *Lindgren et al*, 2004). Ground water flow moves rapidly from various locations within the recharge zone to discharge at the springs, with transit times, measured in dye tracer studies, of hours to weeks following individual precipitation events. The sources of the ground water in the Edwards aquifer that contribute to the Barton Springs are primarily infiltration from streams and creeks that originate in the Contributing Zone, and recharge resulting from precipitation directly in the Recharge Zone. Slade *et al.* (1986) estimated that the streams contribute roughly 85% and direct precipitation roughly 15% of groundwater to the Barton Springs

The Contributing Zone lies due west of the Recharge Zone. In this zone, runoff from sites treated with diazinon may be transported via overland flow to surface water streams and ponds. These streams also derive some component of their total flow, estimated at 30%, from the Trinity aquifer as baseflow (Kuriansky, 1990). Diazinon may then be transported via surface water streams to the Recharge Zone, where it rapidly infiltrates into the network of karst fractures that ultimately feed the Barton Springs system. Unlike pesticides originating within the Recharge Zone, some dilution and degradation is expected during this transport process. Ground water flow across the Trinity-Edwards aquifer boundary is negligible (Lindgren *et al*., 2004)

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, however, the pools are typically fed by groundwater flow through the Recharge Zone of the BSSEA.

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 salamander location is somewhat unique from the other in how exposures are expected to interact with the salamander.

Potential exposures to pesticides for salamanders residing within the fracture system are due to a combination of sources of groundwater: base flow from the Edwards aquifer and groundwater recharge from precipitation events. Thus, salamanders residing within the fracture system of the springs are likely to be exposed to longer-term base flow concentrations of diazinon with occasional shorter duration pulses correlated with precipitation-derived runoff events transported through the fractures.

Figures 6 and 7 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**.

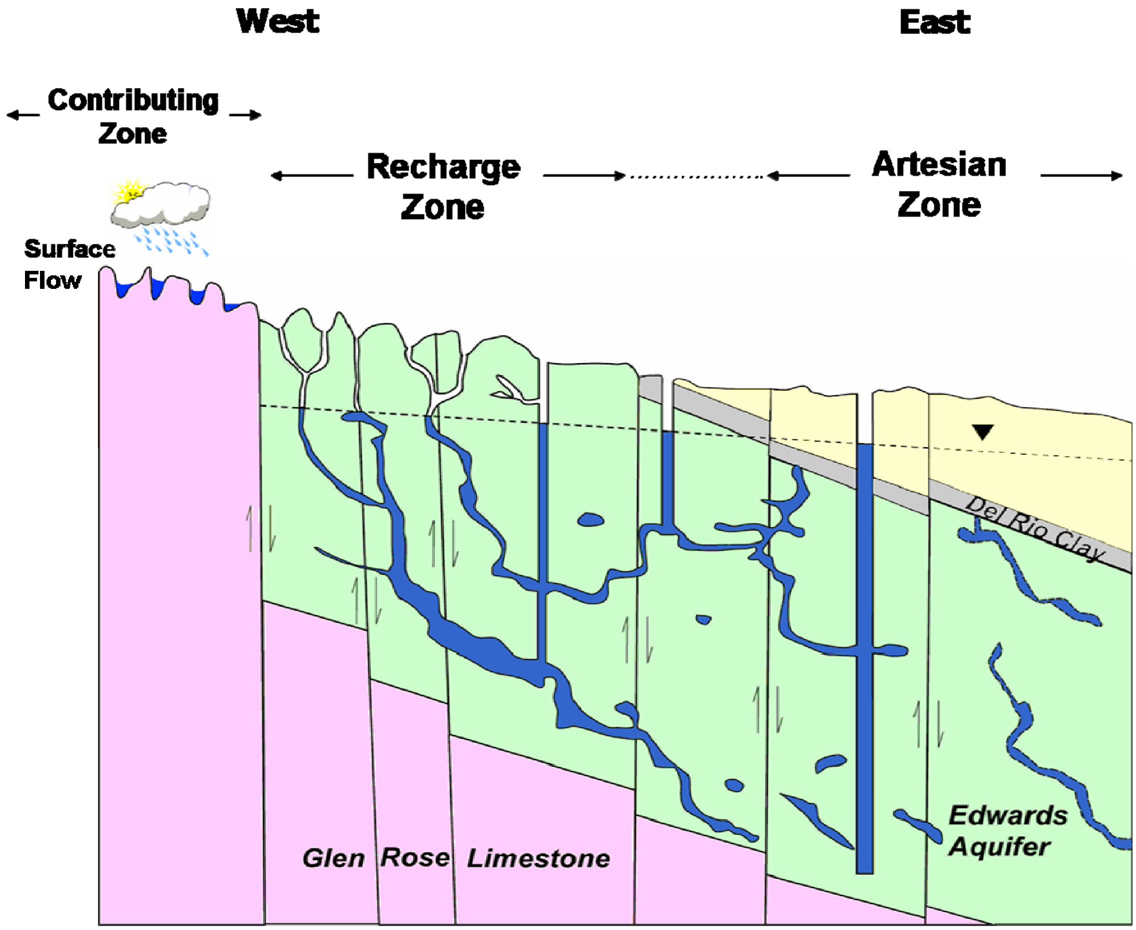


Figure 6. Hydrogeologic Cross Section of the Barton Springs Segment of the Edwards Aquifer and the Contributing Zone Showing Dominant Flow Pathways within Each Hydrozone (Taken from Mahler, 2005).

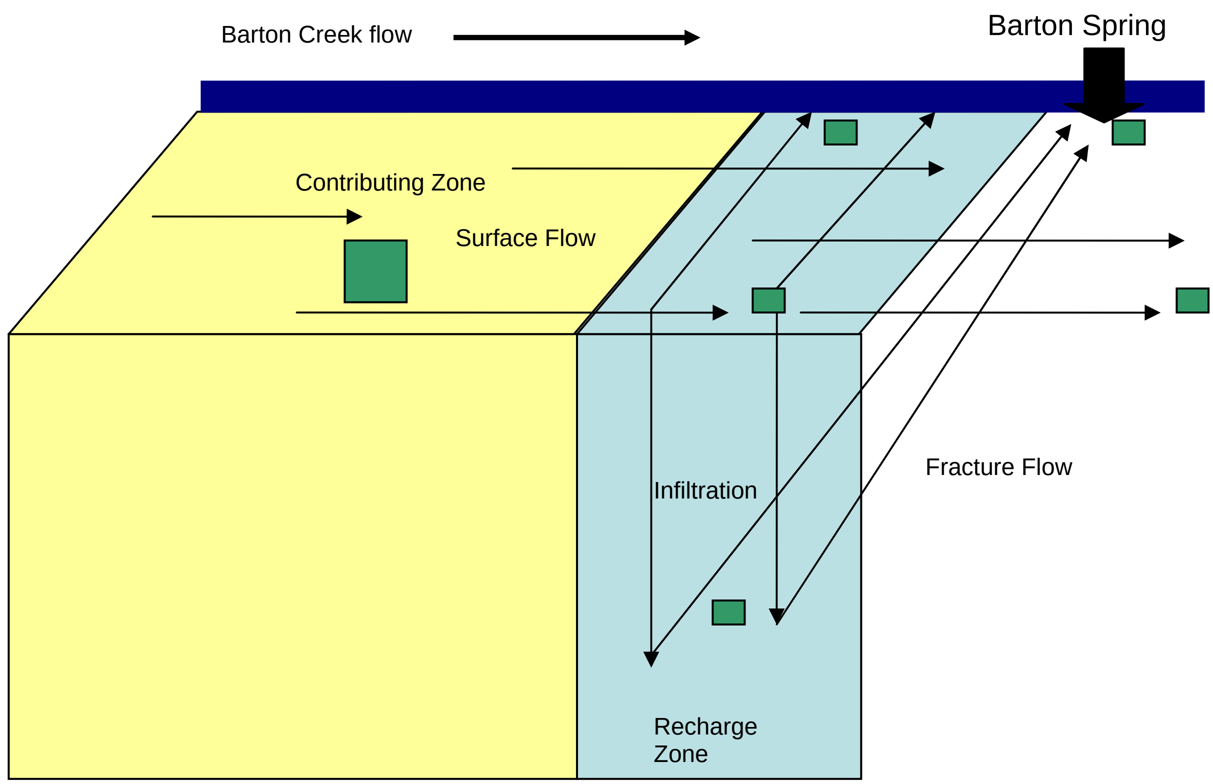


Figure 7. Conceptual Model of Surface and Subsurface Flow within the Barton Springs Watershed. Green Boxes Represent Movement of Dissolved Diazinon Mass.

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 is a major source of groundwater used for drinking water and represents 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).

The Barton Springs Segment of the Edwards aquifer extends from the Colorado River of Texas south roughly 20 miles into Hays County and covers 401 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 of Texas and south of the divide flowing south-southwest. In general, the BSSEA is unconfined in the Recharge Zone and confined (by the Del Rio clay) in the Artesian Zone. It discharges at a number of springs along the Colorado River and Barton Creek. Discharge into Barton Springs is predominantly through the Recharge Zone, and, based on hydrograph data, is typically around 35 cubic feet per second (cfs) during low flow periods (the median annual minimum flow), but can reach above 120 cfs during high flow conditions; the average flow is reported to range between 53 cfs (Hauwert *et al.*, 2004) and 56 cfs (Mahler, 2005). Hydrograph data for Barton Springs from the USGS (**Figure 8**) yields an average flow of 62 cfs. Slade *et al.* (1986) estimated that up to 85% of the recharge reaching the BSSEA was derived from infiltration of the main creeks crossing the Recharge Zone. The remaining recharge is derived from water in inter-stream areas of the Recharge Zone, including from minor tributaries and direct infiltration of precipitation.

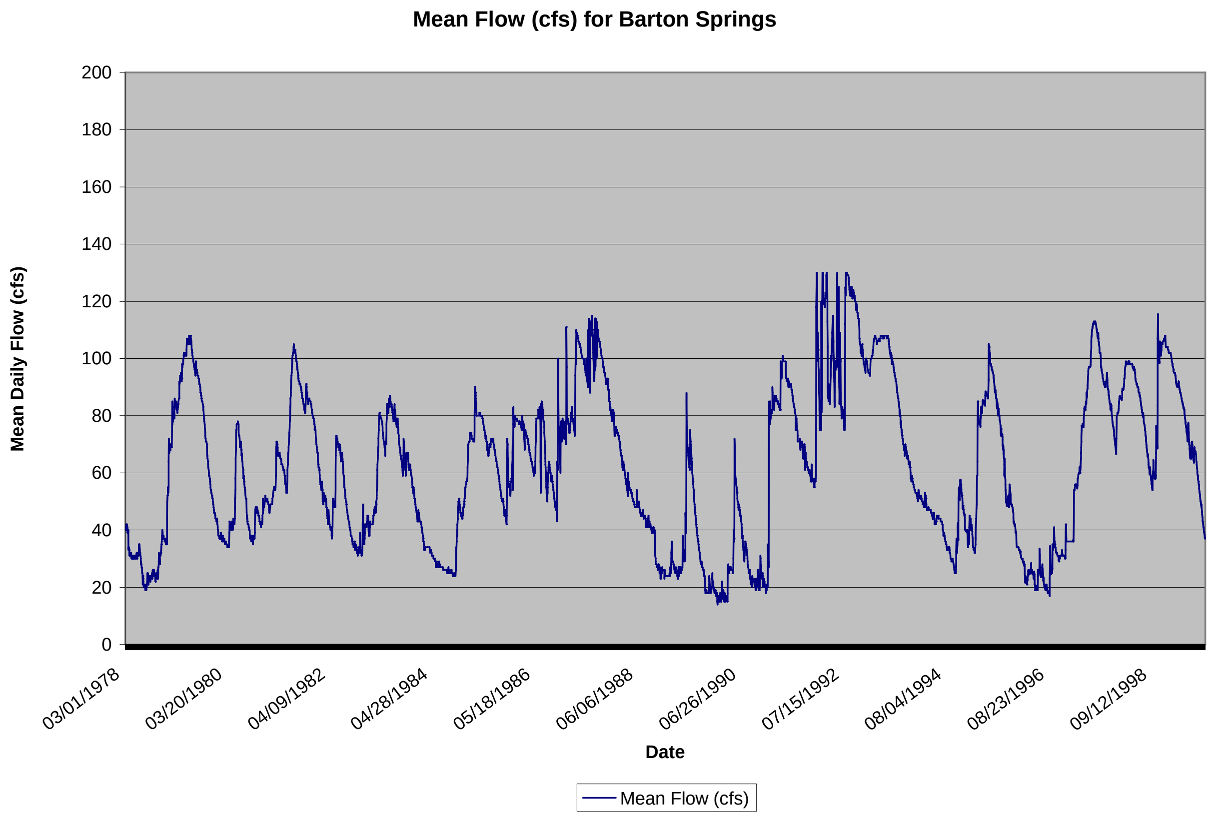


Figure 8. Flow Hydrograph Data for Barton Springs.

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 for locations in close proximity to the springs (defined by Travis County), while farther south and west in the recharge zone, travel times can increase to approximately 4 weeks. **Figure 9** presents a summary of the flow paths defined by this study (Hauwert *et al.*, 2004).

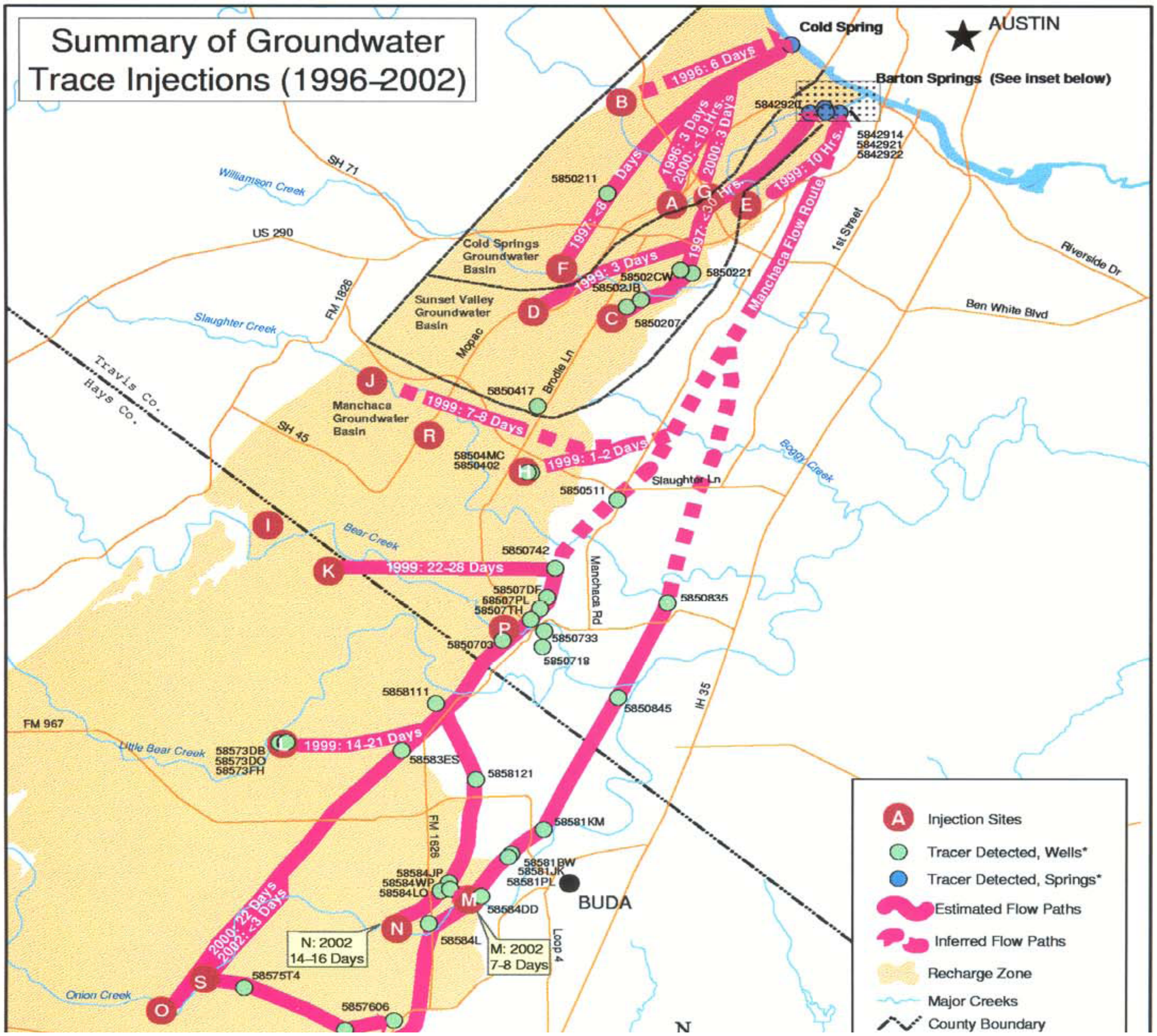


Figure 9. Flow paths within Recharge Zone of the Barton Springs Segment of the Edwards Aquifer (Taken from Mahler, 2005; originally published in Hauwert et al., 2004). Water generally flows from south west to north east.

3.2.3 Conceptual Model of Exposure

Given the understanding of the geology and hydrogeology described above, a combination of modeling and monitoring data is needed to assess the potential exposures from diazinon to the Barton Springs salamander. Routes of exposure are dependent on the location of registered use sites for diazinon within the action area (defined in **Section 2.6** as the Contributing and Recharge Zones), and locations within the pool system (fractures versus pools) where the salamander resides. For instance, uses which are predominantly within the Recharge Zone of the BSSEA result in concentrations in water that are likely to reach the springs via direct transport through the fractures within the karst zone. Uses in the Contributing Zone result in concentrations in water that are transported over longer flowpaths and are subject to both surface and sub-surface transport processes. The interconnected nature of the subsurface network in the BSSEA recharge Zone can have a significant influence on mixing, dilution, storage and degradation of flow (Field, 2004).

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

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

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

3.2.4 Existing Water Monitoring Data

Water monitoring data exist for the springs where the salamander is located as well as creeks and ground water wells located within and near the Barton Springs area of concern (Mahler, 2005). NAWQA data also exist for ground and surface waters throughout the state of Texas (USGS 2006). In addition, creek monitoring data exist for Denton, TX, which is located approximately 200 miles from Austin (Banks *et al.* 2005a). The latter data are particularly interesting to this assessment since they demonstrate that after mitigation resulting from the 2002 IRED, surface water concentrations of diazinon decreased significantly in waters fed by runoff from urban areas.

3.2.4.1 USGS Data Set from Barton Springs Area

Data are available for monitoring of surface water (springs and creeks) and ground water (wells) from the Barton Springs portion of the Edwards Aquifer (Mahler, 2005). Samples were taken at irregular intervals between 1975 and 2005. In total, there were 4 springs sampling locations, 15 creeks sampling locations and 24 well sampling locations. Several of the creek and well locations lie outside of the Barton Springs Aquifer area (**Figures 13 and 14**). 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. 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, 2005a). The comprehensive data set from USGS described in this section is included in **Appendix C**.

3.2.4.1.1 Data from Springs

The most relevant sampling data for this assessment are those collected from the springs. Four springs were included in the USGS analysis, including Main Spring, Eliza Spring, Upper Spring, and the Old Mill Spring (see **Figure 2**). 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. These sampling locations are consistent with the reported locations of the Barton Springs salamander.

Diazinon was detected in samples collected from Main Barton Springs, Upper Barton Springs and Eliza Springs. Diazinon was not detected in any of the 12 samples collected from Old Mill Springs from 2001-2005. The highest detection of diazinon was 0.143 µg/L in the Upper Spring; 91% of samples in this spring and 87% to 100% of samples in the other springs were below the detection limit for diazinon. However, given the nature of the flow regime within the springs, it is unlikely that these sampling events have captured peak exposures. A summary of the available data is located in **Table 6**.

Table 6. Detections of diazinon in 4 spring sampling locations.

Spring Site	# Detections	# Total Samples	Detection Rate	Sampling Dates	Maximum Concentration (µg/L)
Main Barton	10	82	12%	1978-2005	0.03
Upper Barton	5	43	9.3%	2001-2005	0.143
Old Mill	0	12	0%	2001-2005	<0.005
Eliza	2	15	13%	2000-2005	0.00509

Figures 10 - 12 depict the concentrations of diazinon measured in the springs samples from 2000-2005. In these figures, samples which were below the level of detection are depicted as half of the level of detection. These figures also depict the exposure concentrations that would exceed the acute risk LOC for listed invertebrates, *i.e.*, 0.0105 µg/L (parts per billion; ppb), (discussed in greater detail in **Section 5.1.2**) used to determining potential indirect effects to the Barton Springs salamander through reduction of food sources. Exposure concentrations that would exceed the acute and chronic risk LOCs for listed aquatic vertebrates, *i.e.*, 4.5 and <0.55 µg/L, respectively (discussed in greater detail in **Section 5.1.1**) and the chronic risk LOC for listed invertebrates, *i.e.*, 0.17 µg/L, are not exceeded by measured concentrations of diazinon from 2000-2005 in the springs.

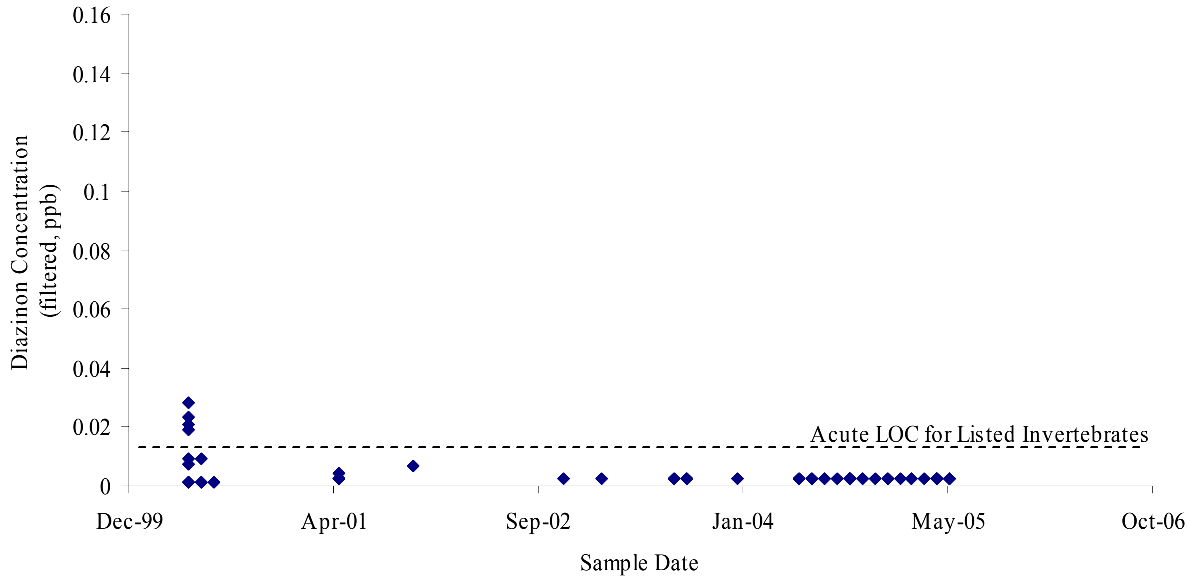


Figure 10. Detections of diazinon in Main Barton Spring from 2000-2005.

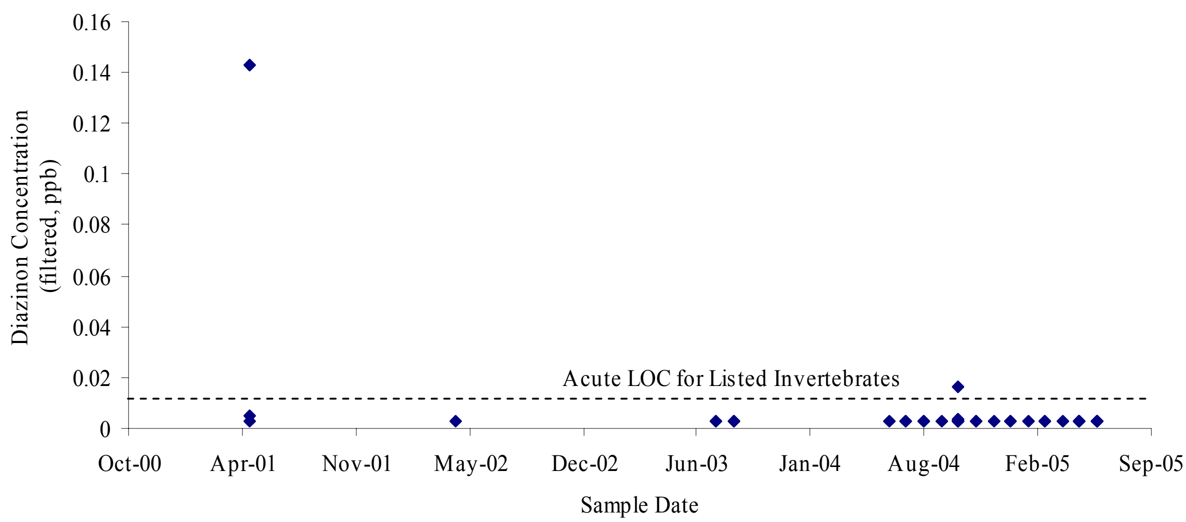


Figure 11. Detections of diazinon in Upper Barton Spring from 2001-2005.

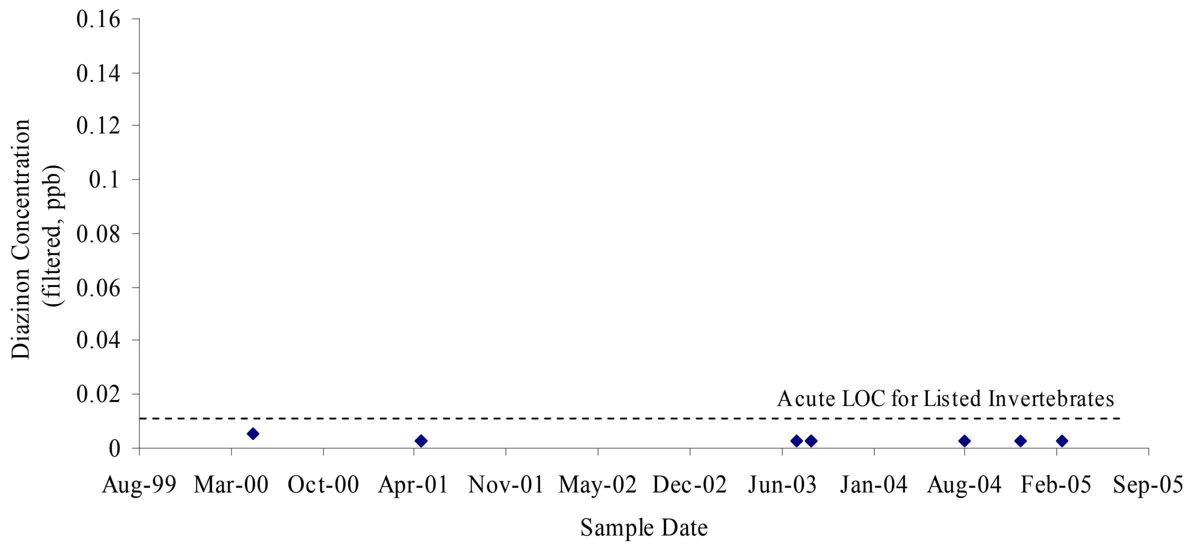


Figure 12. Detections of diazinon in Eliza Spring from 2000-2005.

For Main Barton Springs, the highest diazinon concentration was 0.03 µg/L (unfiltered sample), which was measured in 1978 (not shown in **Figure 10**). During 2000-2001, 20 samples yielded 9 diazinon detections up to 0.0235 µg/L (filtered samples). Only 1 sample was analyzed for diazinon in 2002, which yielded no detection of diazinon. From 2003-2005, 44 filtered samples yielded no detections of diazinon (**Figure 10**).

For Upper Barton Springs, the highest detected concentration of diazinon was 0.143 µg/L (filtered sample), which was measured in 2001. This sample was the only one to exceed the acute risk LOC (RQ>0.05) for listed invertebrates. During 2001-2004, 28 samples yielded 5 diazinon detections. In 2005, 16 samples yielded no detections of diazinon (**Figure 11**).

For Eliza Springs, the highest detected concentration of diazinon was 0.00509 µg/L (filtered sample), which was measured in 2000. During 2000-2001, 7 samples yielded 2 detections of diazinon. No samples were measured for diazinon in 2002. From 2003-2005, diazinon was not detected in 8 samples (**Figure 12**).

3.2.4.1.2 Data from Creeks

There are a total of 15 sites in and near the action area where creeks were sampled and analyzed for diazinon (**Figure 13**). The majority of the sites were sampled only before 2000, prior to the implementation of label mitigations, such as the phase out of urban uses. From 1975-1995, 112 samples were collected from 11 creek sites. Of these samples, 31 contained detectable levels of diazinon at concentrations up to 0.47 µg/L. Five creek sites were sampled during 2000-2005 (**Table 7**). The highest measured concentration of diazinon was 0.26 ppb. Several samples taken from Barton Creek above Barton Springs and the Williamson Creek at Manchaca exceeded the acute and chronic LOCs for listed invertebrates. These exceedances are relevant to the

Barton Springs salamander, as salamanders and their prey are exposed to water from creeks that recharge the Edwards aquifer.

Table 7. Detections of diazinon in 5 creek sampling locations from 2000 to 2005. Samples are filtered.

Creek Site	# Detections	# Total Samples	Detection Rate	Sampling Dates	Maximum Conc. (µg/L)
Barton 71	3	8	38%	2002-2004	0.0099
Barton Creek above Barton Springs	9	13	69%	2000-2004	0.179
Williamson Creek at Manchaca	9	9	100%	2000-2005	0.26
Onion Creek at Driftwood	0	5	0%	2003-2005	<0.005
Onion Creek at Twin Creeks Road	0	3	0%	2004-2005	<0.005

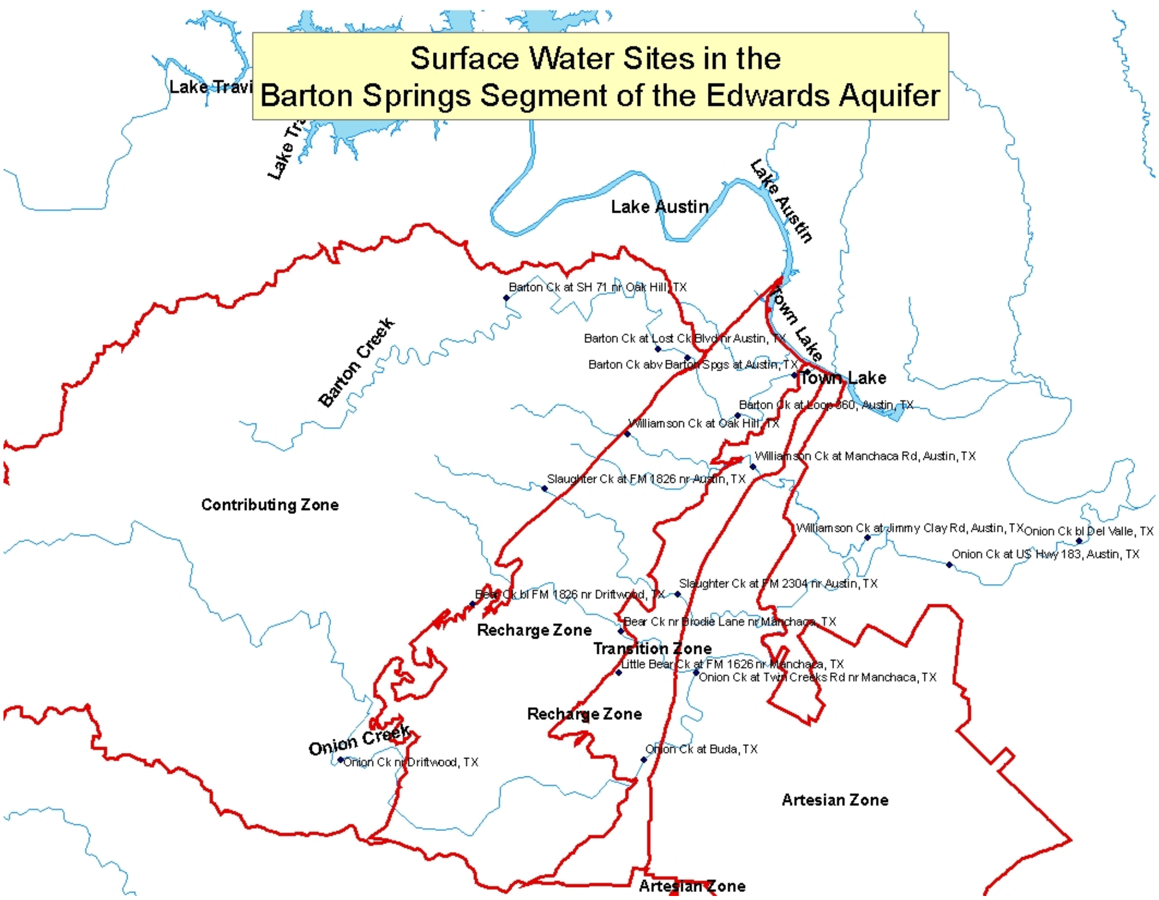


Figure 13. Location of Surface Water Monitoring Sites within the Barton Springs Watershed.

3.2.4.1.3 Data from ground water wells

There are a total of 24 sites in and near the action area where wells were sampled for diazinon (**Figure 14**). Of a total of 71 samples taken during 2000-2005 from 16 wells, 2 contained detectable levels of diazinon, both reported as approximately 0.0017 µg/L (below the limit of quantitation, 0.005 µg/L). From 1977-1993, 4 of 22 samples (from 11 wells) contained detections of diazinon, up to 0.04 µg/L.

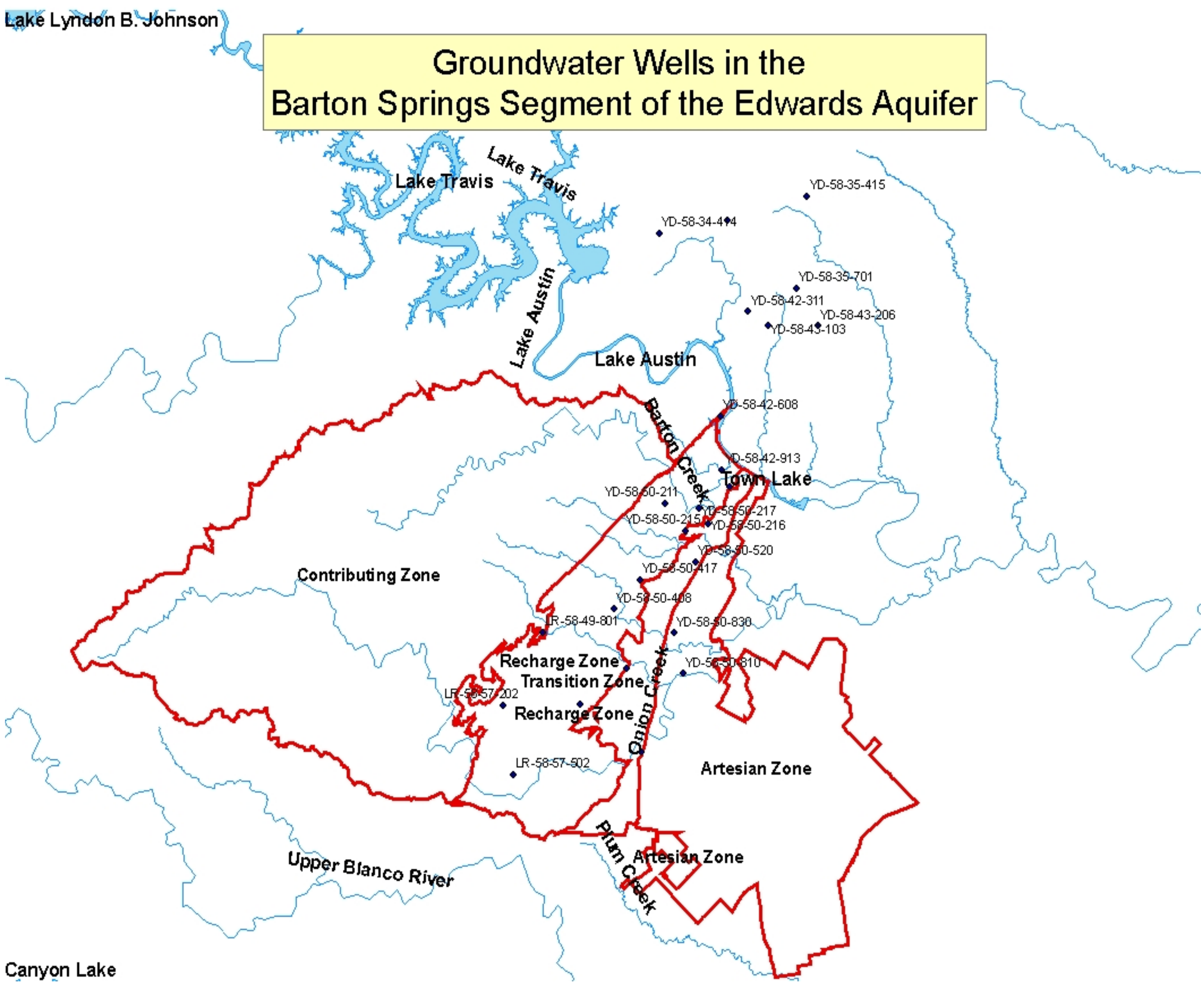


Figure 14. Location of Groundwater Monitoring Sites within the Barton Springs Segment.

3.2.4.2 NAWQA data

Monitoring data of surface water and ground water are available from the United States Geological Survey (USGS) National Water Quality Assessment (NAWQA) program conducted since 1991 (USGS 2006). Data are available through Sep. 30, 2005. During the program, diazinon was analyzed for 19,003 times in surface water and 7048 times in ground water, nationally. The oxygen analog of diazinon (diazoxon) was not analyzed in either surface water or ground water. The monitoring data for diazinon are summarized in **Table 8** at three scales, that for the United States, Texas State, and the three Texas counties of the Barton Springs area: Blanco, Hays, and Travis. Across the United States, diazinon was detected 7,048 times in surface water, with concentrations up to 3.8 µg/L, and 674 times in ground water with concentrations up to 19 µg/L.

Table 8. Detections of diazinon at NAWQA stations in the United States, Texas State, and the Barton Springs area.1

Source	# Detections	# Total Samples	Detection Rate	Sampling Dates	Maximum Concentration (µg/L)
United States					
Surface water	7,048	19003	37%	1991 – 2005	3.8
Ground water	674	53964	1.2%	1992 – 2005	19
Texas State					
Surface water	481	791	61%	1993 – 2005	0.69
Ground water	174	2836	6.1%	1994 – 2002	0.089
Barton Springs area					
Surface water	1	2	50%	1996 – 1998	0.003
Ground water	0	30	0%	1996 – 1997	<0.002

[1]. Concentrations reported in the NAWQA database as indiscrete values (*e.g.* data had a Remark Code indicating that the actual value was less than a set value) are considered as non-detects.

In Texas state, surface water samples (N=791) were collected from June 2, 1993 to Sep. 7, 2005 yielding 481 diazinon detections (61% detection rate) at a maximum concentration of 0.69 µg/L. After a high detection (0.56 µg/L) in an urban area of Dallas County on May 21, 2003, concentrations tended to be less than those of previous years. Ground water in Texas was analyzed for diazinon 2,836 times from Mar. 4, 1994 to Aug. 19, 2002 yielding 174 detections (6.1% detection rate) at a maximum concentration of 0.089 µg/L.

In the three Texas counties of the Barton Springs area, *i.e.*, Blanco, Hays, and Travis counties, only two surface water samples (from Hays County on Dec. 26, 1996 and Jun. 16, 1998) were analyzed for diazinon and 30 ground water samples. Diazinon was below the limit of quantitation (0.002 µg/L) in these surface water samples in 1996 and estimated near the limit of quantitation (est. at 0.003 µg/L) in 1998. Nine of the 30 ground water analyses occurred from June to July in 1996 in Blanco County and 21 analyses occurred from June to August in both 1996 and 1997 in Hays County. None of the ground water analyses in either county detected diazinon above the limit of quantitation (0.002 µg/L).

These NAWQA monitoring data indicate that the detection frequency of diazinon has been higher in surface water than in ground water. In NAWQA monitoring in the Barton Springs area, diazinon has not been observed above the limit of quantitation in ground water and has been estimated near the limit of quantitation in surface water.

3.4.2.3 Denton, Texas data

A network of 70 monitoring stations in rural and urban streams was monitored during periods of normal flow for diazinon concentrations in the City of Denton, Texas, which is located roughly 200 miles north of the Barton Springs area, near Dallas. Sampling was conducted on a monthly basis from March through August during the years 2001 through 2004 (Banks *et al.* 2005a). Collected samples (1243 total) were analyzed by enzyme-linked immunosorbent assays (ELISA) specific for diazinon, with a limit of detection (LOD) of 0.022 μg/L.

The proportion of samples per year that were above the LOD significantly decreased from 2001 through 2004 ($p<0.0001$) (**Table 9**). The proportion of monitoring stations where at least one sample above the LOD was collected per year significantly decreased from 2001 through 2004 as well ($p<0.0007$). Variability in specific conductance and atrazine concentrations from 2001 through 2004 did not indicate any significant trends in this time period, suggesting that environmental factors such as precipitation did not cause these trends of decreasing diazinon concentrations. These results show that a significant reduction in diazinon surface water exposure followed the release of the 2002 IRED, in which label mitigations were recommended to reduce and eventually eliminate diazinon production for residential uses by 2004.

Table 9. Diazinon surface water monitoring data summary from the City of Denton, Texas from 2001 through 2004.

	2001	2002	2003	2004
Maximum concentration (μg/L)	2.58	1.67	1.91	0.85
Proportion of samples above the LOD[1]	100.0	87.8	46.8	44.9
Number of samples	308	311	252	372
Proportion of stations with at least one detect above the LOD[1]	100.0	98.6	79.4	91.2
Number of stations	70	70	68	68

[1] LOD = limit of detection (0.022 μg/L)

3.2.5 Modeling Approach

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

The Approach for Barton Springs Modeling. Because of the unique geology and location-specific focus of the Barton Springs assessment, an approach was taken that incorporated the specific hydrology of the area in an effort to make the modeling approach more relevant than the standard modeling approach that the Agency uses for more generic national-type assessments. A brief description of the Spring's salient features are given here.

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

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

Masses and volumes of runoff were determined for this assessment from modeling scenarios developed specifically for the orchards, nurseries, and other areas found in the Barton Springs Salamander action area (see **Section 3.2.6** and **Appendix B**). Outdoor ornamental uses were modeled with the nursery scenario. Use on peaches was modeled with the orchard scenario. Similar to the Agency's standard ecological risk assessment methodology described above, 30 years of meteorological data for the Austin area were used in these specific scenarios to estimate 1-in-10-year exposure in the Barton Springs.

A summary of the potential diazinon use areas is presented in **Table 10**. Only one orchard was determined through investigation to operate in the action area. Its area (7 acres) was reported online (http://barsanaorchards.com/news8article.html; Mar. 1, 2007). The area of nurseries (3.25 acres) in the action area was investigated using a variety of sources (see p. 11 of **Appendix B**). The use areas are shown to be much smaller than the area where no use occurs (non-use area), the latter of which accounts for roughly 100% of the action area.

Table 10. Extent of Potential Diazinon Use Areas in the Action Area of the Barton Springs Segment of the Edwards Aquifer (BSSEA).

Use Scenario	Area (acres)	Area in Contributing Zone (acres)	Area in Recharge Zone (acres)
Nursery	3.25 (0.00144%)	0.5 (0.0003%)	2.75 (0.00477%)
Orchard	7 (0.003%)	7 (0.004%)	0
Non-use area	226,000 (100%)	169,000 (100%)	57,600 (100%)

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

Contributing Zone. This assessment uses the long-term average stream flow information to calculate an approximate average daily stream flow in the contributing zone. Because the ratio of runoff flow to base stream flow was estimated to be 70:30, knowing the long-term runoff flow enables an estimate of the long-term average streamflow. The long-term (30 years simulated) runoff volume was calculated for each of the scenarios in **Table 10** using PRZM and the respective areas within the contributing zone. The cumulative runoff volume for the contributing zone was calculated according to

$$V_{CZ} = \sum_{t=1}^{n} \left(V_{CZorchard,t} + V_{CZnursery,t} + V_{CZnon-use,t} \right) \qquad \textbf{(3.1)}$$

where V_{CZ} = 30 year simulated cumulative runoff volume [volume]
$V_{CZorchard,t}$ = orchard runoff volume on day t in the contributing zone [volume]
$V_{CZnursery,t}$ = nursery runoff volume on day t in the contributing zone [volume]

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

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

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

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

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

$$V_{CZ,t} = V_{CZorchard,t} + V_{CZnursery,t} + V_{CZnon-use,t} \tag{3.3}$$

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

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

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

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

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

$$C_{CZ,t} = \frac{\left(M_{CZorchard,t} + M_{CZnursery,t}\right)}{\left(V_{CZ,t}\right)} \tag{3.5}$$

where $C_{CZ,t}$ = the concentration in runoff across the contributing zone on any day t [mass/volume]
$M_{CZi,t}$ = the mass of diazinon in runoff in the contributing zone for scenario i on any day t [mass]

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

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

where $C_{stream,t}$ = the concentration in contributing zone streams on any day t [mass/volume]
C_{base} = the average concentration monitored in base flow [mass/volume]

Note that the background concentration in base flow was assumed to be negligible. This is supported by monitoring data in which there were only 2 detections of diazinon out of 71 groundwater samples in this region, and both detections were estimated to be less than the limit of quantitation (<0.005 µg/L). Also, diazinon is expected to hydrolyze moderately in matrix flow under karst conditions (half-life of 77 days at pH 9), further supporting the assumption of negligible background concentrations.

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

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

$$V_{RZ,t} = V_{RZorchard,t} + V_{RZnursery,t} + V_{RZnon-use,t} \qquad \mathbf{(3.7)}$$

where V_{RZ} = runoff volume on day t in the recharge zone [volume]
$V_{RZorchard,t}$ = orchard runoff volume on day t in the recharge zone [volume]
$V_{RZnursery}$ = nursery runoff volume on day t in the recharge zone [volume]
$V_{RZnon-use}$ = non-use runoff volume on day t in the recharge zone [volume]

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

$$C_{RZ,t} = \frac{\left(M_{RZorchard,t} + M_{RZnursery,t} + M_{RZnon-use,t}\right)}{V_{RZ,t}} \qquad \mathbf{(3.8)}$$

where $C_{RZ,t}$ = the concentration in runoff across the recharge zone on any day t [mass/volume]
$M_{RZi,t}$ = the mass of diazinon in runoff in the recharge zone for scenario i on any day t [mass]

Barton Springs Daily Concentrations. It is assumed that the stream flow from the contributing area and the runoff from the recharge area mix and flow through the karst and into the Barton Springs. Stream flow that does not ultimately pass through the Barton Springs is assumed not important because of the assumption of instant mixing of diazinon residues in flow volumes prior to potential diversion. The discharge in streams that leave the action area as a result of large precipitation events is assumed negligible. Therefore, the total discharge produced is determined as:

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

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

Using these calculations, runoff from the recharge zone provides 11% of discharge through the Barton Springs, on average. This is similar to the approximation by Slade *et al*. (1986) and Barrett and Charbeneau (1996) that 15% of recharge to the Barton Springs originates in the recharge zone and 85% originates in the contributing zone.

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

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

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

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

3.2.5.1 Model Inputs

The appropriate PRZM input parameters were selected from environmental fate data submitted by the registrant and in accordance with EFED water model input parameter selection guidance (U.S. EPA 2002). The input parameters selected are similar to those used in the 2002 diazinon IRED (U.S. EPA, 2006); no new environmental fate data were incorporated into this assessment. A summary of the model inputs used in this assessment are provided in **Table 11**. Input parameters for the PE4 shell relating to the EXAMS model were unnecessary for this assessment. Model input reports and the stepwise approach for processing model output are provided in **Appendix E**.

Table 11. PRZM Input Parameters. Source Data are in Tables 2 and 3.

Input Parameter	Value	Source
Application Rate in lbs a.i./A (kg a.i./ha)	Ornamentals: 1.0 (1.1) Peaches: 2.0 (2.2)	Active labels
Applications per Year	Ornamentals: 26 Peaches: 2	Active labels
Application Interval (days)	Ornamentals: 14 Peaches: 120	Active labels
Date of Initial Application	Ornamentals: Jan 2nd Peaches: Jan 15th	Active labels
Application Efficiency[1]	99 % for ground	Input Parameter Guidance[2]
CAM Input	2	Active labels

Input Parameter	Value	Source
IPSCND Input	Ornamentals: 2 Peaches: 3	USDA Crop Profiles[3]
Aerobic Soil Metabolism Half-life (days)	38.7	MRID 40028701 MRID 44746001
Koc (L/kg$_{OC}$)	616	MRID 00118032

1 – Spray drift not included in final EEC due to proximity of use areas to Barton Springs.
2 – Inputs determined in accordance with EFED water model input parameter selection guidance (U.S. EPA 2002).
3 – USDA Crop Profiles information is located at: http://pestdata.ncsu.edu/cropprofiles.

Each use scenario was modeled with ground-based foliar spray application because aerial application to the use sites is no longer allowed. Regardless of the application method, spray drift is not considered to be a significant route of exposure because the source area for diazinon is generally removed from the spring system where the salamander resides, and the diazinon exposures that reach the springs do so via subsurface flow. Therefore, spray drift is assumed to be negligible.

The deposition of diazinon in the post-season (termed "IPSCND" for PRZM modeling) is modeled as complete removal during harvest for ornamentals. For orchards, this parameter is modeled as partial removal during harvest, with the remaining surface residue undergoing decay on plant surfaces.

Since the coefficient of variation for the organic carbon partition coefficient, *i.e.*, $K_{OC.}$ (CV = 25) is less than the coefficient of variation for K_f (CV = 159) in the submitted study, the average K_{OC} of 616 L/kg$_{OC}$ was used to represent binding to soil and sediment.

There are two studies available to estimate the aerobic soil metabolism rate for diazinon, each on one soil. Because the half-lives from these studies are similar (37.4 days and 38.0 days), the upper confidence bound on the mean is similar as well (38.7 days), as calculated according to current EFED guidance for selecting water model input parameters (U.S. EPA 2002).

3.2.6 PRZM Scenarios

A total of three use scenarios were developed for this assessment: nursery, orchard and residential. The residential scenario was not used to model applications of diazinon; it was simply used to provide runoff estimates representative of the action area. Each scenario used meteorological data from a weather station located in Austin, Texas. No weather station closer to the action area provides the data required for exposure modeling. A discussion of each assessed exposure scenario is provided below.

3.2.6.1 Nursery

NASS data for 2002 indicate that *outside* acreage for reported ornamental crops in Hays and Travis Counties is negligible relative to indoor acreage (< 0.1% total indoor and outdoor acreage). The majority of acreage for nursery, greenhouse, floriculture, mushrooms, sod, and vegetable seeds in both years and both counties was grown under glass or other protection. Three confirmed outdoor nursery operations reside within the BSSEA (Kathy Shay, personal communication; Andrea DeLong-Amaya, personal communication); all three are within the Travis county portion of the BSSEA. Total outside wholesale nursery production in the BSSEA is approximately three acres.

For the purposes of modeling a nursery operation in the BSSEA, one of the nurseries was used to conceptualize a facility that is representative of one located within the BSSEA. This nursery was chosen because it had the largest acreage of the three identified nurseries in the action area. Communications with a staff member were used to parameterize the model. The nursery of interest has indoor and outdoor areas for growing and maintaining plants. Outdoor plants include cacti, annuals, perennials, shrubs, and trees. Outdoor plants are maintained on either weed control mats or on gravel. Plants are kept in pots of various sizes, ranging from 4" to multiple gallons, depending upon the type of plant kept within. Irrigation is carried out daily with either hose or sprinkler systems. Plants are maintained outside year-round, with some becoming dormant in the winter and some remaining green. Spring and fall represent the busiest times for plant production and sales for this nursery (personal communication with nursery employee).

3.2.6.2 Orchard

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

3.2.6.3 Residential (for runoff estimation)

Non-use areas of the action area were represented by this scenario for runoff estimation because residential land use (43.4% of action area) is more prevalent than any other type (COA, 2003b). This scenario is intended to represent pervious urban/suburban home and residential areas in the Barton Springs watershed. Brackett soils were chosen to represent residential areas, as they are found in both the contributing and recharge zones and are the most common soil on which residential dwellings are located, accounting for 35% of all soils in residential areas (USDA 2006; USGS 2003). Brackett is a Hydrologic Group C soil, which accounts for approximately 47% of residential soils in drainage.

3.2.6 Aquatic Modeling Results

Table 12 presents the aggregate 1-in-10-year exposure estimates in the Barton Springs from both relevant use scenarios. The modeled 1-in-10-year aggregate peak and average exposure estimates are consistent with concentrations seen in the monitoring data (up to 0.143 µg/L). Due to the conservative assumptions made in the conceptual model (*e.g.* no degradation after runoff) and the modeling of maximum application practices, these estimates may overestimate exposure. Monitored concentrations sampled before the implementation of label mitigations are expected to surpass or be consistent with these modeled values that reflect current labeled uses.

Table 12. 1-in-10-year Barton Springs EECs for Modeled PRZM Scenarios.

Use Pattern	Scenario	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)
Ornamentals	Nursery	0.058	0.007	0.006	0.004	0.003	0.003
Peach	Orchard	0.009	0.001	0.001	0.0006	0.0004	0.0003
Aggregate	NA	0.060	0.007	0.006	0.005	0.003	0.003

4. Effects Assessment

This assessment evaluates the potential for diazinon 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 amphibian 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 diazinon. A summary of the available freshwater ecotoxicity information, use of the probit dose response relationship, and the incident information for diazinon are provided in **Sections 4.1** through **4.4**, respectively. A detailed summary of the available ecotoxicity information for diazinon formulated products is presented in **Appendix A.**

The available information also indicates that aquatic organisms are more sensitive to the technical grade (TGAI) than the formulated products of diazinon; therefore, the focus of this assessment is on the TGAI of diazinon.

4.1 Evaluation of Aquatic Ecotoxicity Studies for Diazinon

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 2000 diazinon IRED (U.S. EPA, 2000a) as well as information obtained on December 14, 2006. The December 2006 ECOTOX search included all open literature data for diazinon and diazoxon (*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 2000 IRED for diazinon, potential adverse effects on sensitive aquatic organisms were identified. 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 the Agency's Overview Document (U.S. EPA, 2004), the most sensitive endpoint for each taxa are 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 13** 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**

Table 13. Aquatic Toxicity Profile for Diazinon.

Assessment Endpoint	Species	Toxicity Value Used in Risk Assessment	Probit Slope	Citation MRID # (Author & Date)	Comment
Acute Direct Toxicity to Salamander	Rainbow trout[1]	96-hour LC_{50} = 90 µg/L	4.5	400946-02 (Johnson and Finley 1980)	Acceptable
Chronic Direct Toxicity to	Brook trout[1]	NOAEC <0.55 µg/L LOAEC = 0.55	N/A	ROODI007 (Allison and	Acceptable: reduced growth

Assessment Endpoint	Species	Toxicity Value Used in Risk Assessment	Probit Slope	Citation MRID # (Author & Date)	Comment
Salamander		µg/L		Hermanutz 1977)	
Indirect Toxicity to Salamander via Acute Toxicity to Freshwater Invertebrates (*i.e.* prey items)	Water flea (*Ceriodaphnia dubia*)	48-hour EC_{50} = 0.21 µg/L	4.5	Banks *et al.* 2005	Supplemental:
Indirect Toxicity to Salamander via Chronic Toxicity to Freshwater Invertebrates (*i.e.* prey items)	Water flea (*D. magna*)	NOAEC = 0.17 µg/L LOAEC = <0.32 µg/L	N/A	407823-02 (Supernant 1988)	Mortality
Indirect Toxicity to Salamander via Acute Toxicity to Non-vascular aquatic plants	Green algae	EC_{50} = 3,700 µg/L EC05= 66 µg/L	0.90	405098-06	Acceptable Decreased growth

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

Acute toxicity to aquatic fish and invertebrates is categorized using the system shown in **Table 14** (U.S. EPA, 2004). Toxicity categories for aquatic plants have not been defined. Based on these categories, at most, diazinon is classified very highly toxic to freshwater fish and invertebrates on an acute exposure basis.

Table 14. Categories of Acute Toxicity for Aquatic Organisms.

LC_{50} (ppb)	Toxicity Category
< 100	Very highly toxic
> 100 – 1,000	Highly toxic
> 1,000 – 10,000	Moderately toxic
> 10,000 – 100,000	Slightly toxic
> 100,000	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 diazinon 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 amphibians, 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. Diazinon toxicity has been evaluated in numerous freshwater fish species, including rainbow trout, brook trout, bluegill sunfish, fathead minnow, tilapia, zebrafish, goldfish, and carp. The results of these studies demonstrate a wide range of sensitivity to diazinon. The range of acute freshwater fish LC_{50} values for diazinon spans one order of magnitude, from 90 to 7,800 µg/L; therefore, diazinon is categorized as very highly (< 100 µg/L) to moderately (>1,000 to 10,000 µg/L) toxic to freshwater fish on an acute exposure basis. The freshwater fish acute LC_{50} value of 90 µg/L is based on a static 96-hour toxicity test using rainbow trout (*Oncorhynchus mykiss*) (MRID # 400946-02). No sublethal effects were reported as part of this study. A complete list of all the acute freshwater fish toxicity data for diazinon 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 study for diazinon is available and summarized in **Table A-12** of **Appendix A**. The chronic effects of diazinon on fathead minnows (*Pimephales promelas*) and brook trout (*Salvelinus fontinalis)* were determined in flow-through systems with constant toxicant concentrations (Allison and Hermanutz 1977). Fathead minnows exposed to the lowest concentration tested (3.2 µg/L) from 5 days after hatch through spawning had a significantly higher incidence of scoliosis than the control ($p=0.05$). Hatch of their progeny was reduced by 30% at this concentration. Yearling brook trout exposed to 4.8 µg/L and above began developing scoliosis and lordosis within a few weeks. Growth of brook trout was substantially inhibited during the first 3 months at 4.8 μg/L and above. Neurological symptoms were evident in brook trout at 2.4 ug/L and above early in the tests, but were rarely observed after 4 or 5 months of exposure. Exposure of mature brook trout for 6 to 8 months to concentrations ranging from 9.6 μg/L to the lowest tested (0.55 μg/L) resulted in equally reduced growth rates for their progeny. Transfer of progeny between concentrations indicated that effects noted for progeny of both species at lower concentrations were the result of parental exposure alone and not the exposure of progeny following fertilization. Decreased growth of progeny relative to controls was roughly similar for both the highest and lowest treatment concentrations with a 16% decrease in body length and 40% decrease in body weight relative to controls; thus, the fish exhibited a non- monotonic dose response. Although offspring were reduced in size, survival of the young was not statistically different from controls in diazinon-treated groups. It is possible though that the reduced size of the young as well as the skeletal deformities of the adults would render the animals more susceptible to predation. At this time, there are no data for diazinon that meet guidelines testing requirements for establishing a chronic NOAEC in freshwater fish. However, the registrant is in the process of completing these studies in response to a data call-in since the original study failed to establish a NOEC. Based on the information discussed above, the NOAEC is less than the lowest concentration tested using brook trout (NOAEC <0.55 μg/L).

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.

In Atlantic salmon (*Salmo salar*), neuroendocrine-mediated olfactory functions were affected at 1.0 μg/L diazinon (Moore and Waring, 1996). 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 diazinon at 0.5 μg/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 a study of chinook salmon (*Oncorhynchus tshawytscha*) antipredator behavior by Scholz *et al* (2000), diazinon exposure resulted in significant effects of swimming and feeding behavior at concentrations of 1 μg/L; fish remained more active and fed more frequently in the presence of an alarm stimulus (skin extract) relative to controls. The effect of diazinon on chinook salmon homing success was also examined in the Scholz *et al* (2000) study. Significantly fewer salmon returned after exposure to 10 μg/L diazinon; however, chinook salmon survival was not reported as impaired. This study has been more thoroughly reviewed (**Appendix A**) and there is considerable uncertainty regarding the extent to which diminished olfactory response as it relates to predator avoidance and homing behavior will affect the survival and reproduction of fish.

In addition, EPA did not use these data in development of the aquatic life water quality criteria for diazinon because population level effects of specific chemicals on the olfactory system of aquatic organisms can only be hypothesized at this time and not substantiated (no articles were obtained that evaluated this issue satisfactorily). The primary unanswered question is how serious of an impact does the temporary loss of olfactory function and associated altered behavior have on the homing, migratory patterns, feeding activity and avoidance of predators for the exposed organisms, and more importantly, on the ability of the exposed population to reproduce, grow and ultimately survive in the wild. Thus, the impact of sublethal effects on the long-term survival of an exposed aquatic population is very difficult to determine from laboratory studies, and therefore complex long-term field studies are needed to address this issue.

Although these studies raise concern about the effects of diazinon 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 diazinon-related mortality and sublethal effects to aquatic-phase amphibians 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 mountain yellow-legged frog (*Rana. boylii*) indicate that they are relatively insensitive to diazinon [compared to fish] with acute LC_{50} values 7,500 µg/L (Sparling and Fellars 2006). Acute toxicity data are not available for salamanders. No chronic toxicity data are available for aquatic-phase amphibians.

4.1.2.2 Amphibians: Open Literature Data on Sublethal Effects

Frogs (Anurans)

Very few data are available to evaluate the toxicity of diazinon to either aquatic or terrestrial-phase amphibians. The data that do exist indicate that freshwater fish are many orders of magnitude more sensitive to diazinon than aquatic and/or terrestrial phase amphibians. In a study of mountain yellow-legged frog larvae (*Rana boylii*), the nominal 96-hr LC_{50} for diazinon and diazoxon were 7,500 and 760 µg/L, respectively (Sparling and Fellers 2006). Although actual concentrations were not measured, the study is useful for demonstrating that diazoxon is roughly an order of magnitude more toxic than the parent compound.

Addtionally, the EFED exotoxicity database reports an LD_{50} of greater than 2000 mg/kg for terrestrial-phase bullfrogs (*R. catesbiana*).

4.1.3 Toxicity to Freshwater Invertebrates

Freshwater aquatic invertebrate toxicity data were used to assess potential indirect effects of diazinon to the Barton Springs salamander. Direct effects to freshwater invertebrates resulting from exposure to diazinon 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, 2006), is provided below in **Sections 4.1.3.1 through 4.1.3.3**.

4.1.3.1 Freshwater Invertebrates: Acute Exposure Studies

Diazinon is classified as very highly toxic to aquatic invertebrates. Toxicity estimates, EC_{50} and LC_{50} values, for freshwater invertebrates ranged from 0.8 to 35 µg/L. Although the original ecological risk assessment of diazinon reported a 96-hr LC_{50} as low as 0.2 µg/L for scuds (*Gammarus fasciatus*), a reanalysis of the raw data indicated that the 96-hr LC_{50} value was off by an order of magnitude and that the correct value is 2 µg/L (U.S. EPA Memo to SRRD dated 10/05/2005). Data were located through ECOTOX indicating that diazinon is very highly toxic to *Ceriodaphnia dubia* (48-hr EC_{50}=0.21 µg/L) (Banks *et al*. 2005). All of the available acute toxicity data for freshwater invertebrates are provided in **Section A.2.5 and Table A-18** of **Appendix A**.

Several years ago, OPP conducted an analysis of U.S.G.S. data used to support the Mayer and Ellerseick data set. The analysis (**Appendix I**) included 48-hr acute toxicity data for freshwater aquatic invertebrates including *Simocephalus serrulatus*, *Daphnia pulex*, *Gammarus fasciatus* and *Pteronarcys californica*. Across the four species, the 48-hr probit dose response slope ranged from 5.74 to 6.90; the mean slope and standard error of the mean were 6.34 and 0.21, respectively. Since a probit dose-response slope is not available for the most the most sensitive species, *i.e*., *Ceriodaphnia dubia*, the mean slope of 6.34 will be used in the analysis of potential individual effects discussed below.

4.1.3.2 Freshwater Invertebrates: Chronic Exposure Studies

The most sensitive chronic endpoint for freshwater invertebrates is based on a 21-day flow-through study on waterfleas (*Daphnia magna*), which showed significant effects on survival (100% mortality) at diazinon concentrations greater than 0.17 µg/L; the NOAEC and LOAEC for this study are 0.17 and 0.32 µg/L, respectively (MRID # 407823-02).

4.1.4 Toxicity to Aquatic Plants

Aquatic plant toxicity studies were used as one of the measures of effect to evaluate whether diazinon 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 diazinon to affect primary productivity. Laboratory studies were used to determine whether diazinon 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

A single aquatic plant study is available for determining the toxicity of diazinon to nonvascular aquatic plants. Toxicity testing with green algae (*Pseudokirchneriella subcapitata*) resulted in a 7-day EC_{50} of 3,700 µg/L (MRID 405098-06). A reanalysis of the data to estimate an EC_{05} was conducted using the Probit procedure of the Statistical Analysis System (Release 9.1; SAS Institute, Inc., Cary, NC); the probit-estimated EC_{05} is 66 µg/L; the probit dose-response slope is relatively shallow at 0.90. Relative to other aquatic organisms tested, green algae are not particularly sensitive to diazinon given the chemical's primary mode of action as an acetylcholine esterase inhibitor.

Although no acceptable data are available for aquatic vascular plants, the data on nonvascular plants suggests that the aquatic plants are not as sensitive to diazinon as aquatic animals. Additionally, Tier II vegetative vigor testing of vascular terrestrial plants reported in the IRED (USEPA 2002), indicates EC_{25} values in excess of the highest rates tested (EC_{25}>7 lbs a.i.) for the majority of species tested; however, the most sensitive species, *i.e,* cucumbers (*Cucumis* *sativis*) had an EC_{25} and EC_{05} at exposure levels equivalent to application rates of 3.2 and 1.3 lbs a.i./A. Tier II seedling emergence studies indicated that the most sensitive species tested, *i.e.*, oats (*Avena sativa*) had a EC_{25} and an EC_{05} at exposure levels equivalent to application rates of 5.3 and 0.17 lbs a.i./A, respectively.

4.1.5 Freshwater Field Studies

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

Diazinon has been the subject of a mesocosm study where 450 m^2 ponds were monitored following 6 applications of diazinon, alternating between spray drift events and simulated runoff events separated by 1-wk intervals (MRID 425639-01). Nominal treatment concentrations were equivalent to 5.7, 11.4, 22.9, 45.8 and 91.5 µg a.i./L of pond water. Diazinon was shown to have strongly affected the zooplankton taxon Cladocera, where abundance was significantly reduced in all treatments in 5 (36%) of 14 sample periods. Tricoptera abundance was also significantly reduced in all treatments for 29% of the sample periods. Dipterans were also significantly affected. The overall impact of diazinon on the aquatic community was that many aquatic invertebrates were affected at treatment concentrations greater than 11 µg a.i./L; however, most taxa recovered after treatment. Although significant reductions were observed in macroinvertebrate abundance throughout the study period, fish and plants were generally unaffected by the diazinon treatments. Under the study conditions tested, mesocosms treated with multiple applications of diazinon did not reveal any statistically significant direct or indirect effects on fish even though there were significant fluctuations in aquatic macroinvertebrates due to diazinon. A more complete description of this study is located in **Appendix A**.

4.2 Discussion of Degradate Toxicities

With respect to the diazinon degradate oxypyrimidine, it is assumed that it is of lesser toxicity as compared to the parent compound. Comparison of available toxicity information for oxypyrimidine indicates lesser aquatic toxicity than the parent for freshwater fish, invertebrates, and aquatic plants. Specifically, the available degradate toxicity data for oxypyrimadine indicate that it is practically nontoxic to freshwater fish (rainbow trout 96-hr LC_{50}>101 mg a.i./L) (MRID 463643-12; Grade 1993a) and invertebrates (48-hr EC50>102 mg a.i./L) (MRID 463643-13; Grade 1993b) with no mortality at the maximum concentrations tested. In addition, available aquatic plant degradate toxicity data for oxypyrimidine indicate that oxypyrimidine is practically nontoxic to nonvascular aquatic plants (green algae) with non-definitive EC_{50} values (EC_{50}>109 mg a.i./L) (Grade 1993c; MRID 463643-14) at concentrations 29 times higher than the lowest reported aquatic plant EC_{50} value for parent diazinon. Therefore, given the lesser toxicity of oxypyrimidine, as compared to the parent, concentrations of this degradate are not assessed.

With respect to the intermediate degradate diazoxon, acute and subacute toxicity testing with birds indicate that the compound is minimally as toxic (LD_{50}=5 mg a.i./kg bw) (Rodgers 2005a ;MRID 465796-04) as the parent (LD_{50}=10 mg a.i./kg bw) on an acute oral exposure basis and is more toxic (LC_{50} = 72 mg a.i./kg diet) (Rodgers 2005b; MRID 465796-02) than the parent (LC_{50}=245 mg a.i./kg diet) on a subacute dietey exposure basis. Toxicity testing with aquatic-phase amphibians indicates that diazoxon is an order of magnitude more toxic than the parent compound (Sparling and Fellars 2007). However, as discussed in the screening-level ecological risk assessment of diazinon (USEPA 2002), the formation of diazoxon was not observed in any of the laboratory biotic or abiotic degradation studies of diazinon. None of the monitoring data collected in the Barton Springs area targeted the oxygen analog of diazinon. Therefore, it is uncertain what conditions favor its formation and/or persistence in the environment. At this point there is no reasonable way to document the potential risk from diazoxon other than to recognize that the oxon is more toxic than the parent and that the extent to which it may form under conditions present in the BSSEA is uncertain.

Appendix A contains more detailed descriptions of studies assessing the toxicities of oxypyrimidine and diazoxon to aquatic and terrestrial organisms.

5. Risk Characterization

Risk characterization is the integration of the exposure and effects characterizations to determine the potential ecological risk from varying diazinon 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 is estimated by calculating the ratio of exposure to toxicity using 1-in-10 year estimated environmental concentrations (EECs; **Table 12**) and the appropriate toxicity endpoint (see **Table 13**). 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**). For acute exposures to the salamander and invertebrates, the LOC is 0.05. The LOC for chronic exposures to fish and invertebrates, as well as acute exposures to aquatic plants is 1.0.

RQs were based on the most sensitive endpoints and modeled surface water concentrations from the following scenarios for diazinon:

- outdoor ornamental use @ 1 lbs a.i./A; 26 applications with 14 days between applications
- peach and nectarine use @ 2 lbs a.i./A; 2 applications, once at dormancy and once in-season

In addition, RQs were derived based on the aggregate exposure of the two uses listed above.

5.1.1 Direct Effects

For assessing risks of direct effects to the salamander, 1-in-10 year peak EECs are used with the lowest acute toxicity value for fish in order to derive acute risk quotients for the salamander. For chronic risks, 1-in-10 year peak 60-day EECs and the lowest chronic toxicity value for fish are used to derive RQ values for the salamander.

Based on RQ values calculated using individual 1-in-10 year EECs for waters within the Barton Springs proper, for acute exposures, the acute risk LOC is not exceeded for any individual uses. Additionally, acute exposure of the salamander to diazinon from all uses (aggregate) does not result in an exceedance of the acute risk LOC for listed species. For chronic exposures, the LOC is possibly exceeded for all uses (**Table 15**). The uncertainty results from the fact that the chronic risk estimate is based on a LOEC and the actual NOEC from the study is less than the lowest concentration tested.

Table 15. Direct Effect RQs for the Barton Springs Salamander based on refined EECs.

Duration of Exposure	Toxicity Value (µg/L)	Use	EEC (µg/L)[3]	RQ	LOC Exceedance? [4]
Acute	90[1]	Ornamentals	0.058	0.001	No
		Peach	0.009	0.0001	No
		Aggregate[5]	0.060	0.001	No
Chronic	<0.55[2]	Ornamentals	0.003	>0.006[6]	Possibly
		Peach	0.0004	>0.0007[6]	Possibly
		Aggregate[5]	0.003	>0.006[6]	Possibly

[1] 96-h LC_{50} value from toxicity study with Rainbow Trout (MRID 400946-02).
[2]NOAEC value from chronic toxicity study with brook trout (MRID Allison and Hermanutz 1977).
[3]EECs are from **Table 12**. RQs for acute exposures utilize peak EECs, while RQs for chronic exposures utilize 60-day EECs.
[4]For acute exposures, the LOC is 0.05. For chronic exposures, the LOC is 1.0.
[5]Aggregate use represents the sum of diazinon from all uses.
[6] Potentially exceeds chronic risk level of concern (RQ≥1.0)

5.1.2 Indirect Effects

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

For assessing risks of indirect effects to the salamander due to effects to its prey, RQs were derived for freshwater invertebrates based on EECs representative of concentrations of diazinon in the springs. Peak 1-in-10 year EECs for the Barton Springs are used with the lowest acute toxicity value for invertebrates in order to derive acute risk quotients for invertebrates. For chronic risks, 1-in-10 year peak EECs over a 21-day period and the lowest chronic toxicity value for freshwater invertebrates are used to derive RQ values.

For acute exposures, the acute risk to listed species LOC is exceeded for use on ornamentals and for aggregated uses. Chronic exposures of invertebrates to diazinon from individual and aggregated uses do not exceed the chronic risk LOC (**Table 16**).

Table 16. Invertebrate RQs relevant to indirect effects to the Barton Springs Salamander.

Duration of Exposure	Toxicity Value (µg/L)	Use	EEC (µg/L)[3]	RQ	LOC Exceedance?[4]
Acute	0.21[1]	Ornamentals	0.058	0.28[6]	Yes
		Peach	0.009	0.03	No
		Aggregate[5]	0.060	0.29[6]	Yes
Chronic	0.17[2]	Ornamentals	0.006	0.04	No
		Peach	0.001	0.01	No
		Aggregate[5]	0.006	0.04	No

[1] 48-h EC_{50} value from toxicity study with *Ceriodaphnia dubia* (Banks *et al.* 2005).
[2]NOAEC value from chronic toxicity study with *Daphnia magna* (MRID 407823-02).
[3]EECs are from **Table 12**. RQs for acute exposures utilize peak EECs, while RQs for chronic exposures utilize 21-day EECs.
[4]For acute exposures, the LOC is 0.05. For chronic exposures, the LOC is 1.0.
[5]Aggregate use represents the sum of diazinon from all uses.
[6]Exceeds the acute risk to endangered species LOC (RQ>0.05)

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

For assessing risks of indirect effects to the salamander due to effects to its habitat, RQs were derived for aquatic plants based on EECs representative of concentrations of diazinon in the springs. Peak 1-in-10 year EECs are used with the lowest acute toxicity value for aquatic plants in order to derive acute risk quotients for plants.

For all exposures, including the aggregate of all exposures, the LOC is not exceeded by RQs for aquatic plants (**Table 17**). Although there are no data to assess the risk to vascular aquatic plants, the available data of nonvascular aquatic plants and for terrestrial vascular plants suggest that plants are not particularly sensitive to diazinon. Additionally, there are no reported field incidents related to the use of diazinon. Therefore, at the application rates modeled and based on the available data, the use of diazinon in the action area is not likely to indirectly affect the Barton Springs salamander based on reductions in aquatic vascular plants.

Table 17. . Aquatic plant RQs relevant to indirect effects to the Barton Springs Salamander.

Plant Type	Toxicity Value (µg/L)	Use	EEC (µg/L)[2]	RQ	LOC Exceedance?[3]
Unicellular	66[1]	Ornamentals	0.058	0.01	No
		Peach	0.009	0.0001	No
		Aggregate[4]	0.060	0.001	No

[1] EC_{05} value from toxicity study with *green algae* (MRID 405098-06).
[2]EECs are from **Table 12**. RQs utilize peak EECs.
[3]For exposures to plants, the LOC is 1.0.
[4]Aggregate use represents the sum of diazinon from all uses.

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 diazinon'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

Based on exposure estimates for use of diazinon on individual uses alone and for the aggregate exposure from use on ornamentals and orchards within the action area, the acute risk to endangered species LOC is not exceeded for direct effects to the salamander.

Chronic risk RQ values (RQ>0.006) for direct effects to the Barton Springs salamander are several orders of magnitude below the chronic risk LOC; however, there is uncertainty regarding the absence of a discrete NOEC value (NOEC<0.55 µg/L). In the fathead minnow full life cycle study for which the NOEC/LOEC is based, there was a 16% decrease in progeny length and a 40% decrease in progeny body weight at the lowest concentration tested (0.55 µg/L). However, no other measurement endpoint was affected at this concentration. While none of the chronic toxicity tests reported in the original risk assessment for freshwater fish established a NOEC, there is nothing available in either registrant-submitted studies or open literature to suggest that freshwater vertebrates exhibit chronic effects at diazinon concentrations that would be necessary (NOEC=0.006) to exceed the chronic risk LOC based on estimated environmental concentrations for Barton Springs. Therefore, the likelihood of direct chronic effects of diazinon at the concentrations estimated to occur in the BSSEA is considered low.

Therefore, diazinon use in the action area is not likely to affect the Barton Springs salamander through direct acute effects on the salamander. Although there is uncertainty regarding the chronic effects threshold value (NOEC) for freshwater vertebrates, the preponderance of data [and lack of any data to the contrary] that effects thresholds are orders of magnitude higher than what would be required to exceed the chronic risk LOC. Additionally, monitoring data collected subsequent to the cancellation of all residential uses and the reduction in the number and type of agricultural uses indicate that diazinon in the Barton Springs is below the level of detection. These data suggest that the underlying assumption of 26 applications/year used to model ornamental/nursery uses in the BSSEA is very conservative. Therefore, diazinon use in the action area is deemed a may affect but not likely to adversely affect the Barton Springs salamander via direct chronic effects since the potential chronic effects are considered discountable.

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

Consistent with the toxicity data indicating that diazinon is very highly toxic to freshwater invertebrates, exposure estimates for each of the evaluated uses exceed the acute risk to endangered species LOC by a factor of roughly 5.5X. Based on a presumed probit dose-response slope of 6.3 discussed previously and an RQ value of 0.27, the likelihood of acute mortality for individual invertebrates following use of diazinon on ornamentals in the action area is 1 out of 5870 (0.02%) (**Appendix I**). Use on ornamentals and aggregated uses are expected to result in diazinon concentrations in runoff that will result in acute mortality of aquatic invertebrates. Even a single application of diazinon to ornamentals would result in an exceedance (RQ~0.08) of the acute risk to listed species LOC although the likelihood of an individual invertebrate mortality would be low at 1 out of 4.1×10^{11}. Although the risk assessment for effects to invertebrates is based on the most sensitive species (*Ceriodaphnia*), cladocerans as a whole (**Figure 17**) are sensitive to diazinon and RQ values for less sensitive species within the taxon, *e.g. Daphnia magna* EC_{50}=0.87 µg/L, would exceed the acute risk to listed species LOC (RQ=0.07). Addtitionally, the potential effects of diazinon on specific taxa has been demonstrated in mesocosm data (MRID 425639-01) where cladocerans were effectively eliminated from the invertebrate community at higher exposure concentrations.

The data on cladocerans represent information on the sensitivity of zooplankton to diazinon as the remaining taxa for which there are data are more representative of macroinvertebrates. The zooplankton serve as prey for aquatic macroinvertebrates and the apparent sensitivity of zooplankton to diazinon suggests that macroinvertebrates could be affected through reduction in their forage base.

As discussed in greater detail in **Appendix D**, although the Barton Springs salamander is considered an opportunistic feeder, the most prevalent invertebrates found in stomach content analyses were macroinvertebrates consisting of ostracods, amphipods, and chironomids (USFWS, 2005). These are relatively large invertebrates (macroinvertebrates) and it is not clear as to the extent that smaller invertebrates (zooplankton) like cladocerans make up the diet of the salamander. Additionally, it is uncertain as to the extent that the most sensitive species used in this assessment reflect the sensitivities of the larger prey items; however, the sensitivity distribution depicted in **Figure 17** suggests that larger invertebrates tend to be less sensitive than smaller invertebrates. To the extent that larger invertebrates are less sensitive and to the extent that Barton Springs salamanders preferentially feed on the less sensitive taxa would markedly affect risk estimates for indirect effects to the salamander.

Based on the likelihood of individual effect analysis where only 0.02% of the most sensitive species are expected to experience acute mortality at the estimated environmental concentrations for diazinon in the BSSEA, it does not appear likely that this loss would substantially affect the forage base for macroinvertebrates. Also, although it is not likely that Barton Springs salamanders depend exclusively on macroinvertebrates as a forage base, the information provided through the U. S. Fish and Wildlife Service on stomach content analysis and based on toxicity data showing that macroinvertebrates are not as sensitive to diazinon as zooplankton, it does not appear likely that the forage base for Barton Springs salamanders will be adversely affected. Therefore, the likelihood of indirect effects on the Barton Springs salamander from the use of diazinon is viewed as a may affect but not likely to adversely affect since the potential effects are considered discountable.

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

With an EC_{50} of greater than 3,700 μg/L, aquatic plants were some of the least sensitive aquatic organism tested with diazinon. Based on the available data for freshwater nonvascular plants, estimated diazinon concentrations have no affect on aquatic [nonvascular] plants.

There is uncertainty regarding the potential effect of diazinon on aquatic vascular plants since the habitat of the salamander is composed of moss and vascular plants (See **Appendix D**). However, the risk of diazinon to the salamander through reduction of habitat is considered to be low based on the data available for aquatic nonvascular plants, vascular terrestrial plants and the lack of any reported field incidents involving plants.

5.2.4. Incident reports

The original IRED contained a relatively thorough discussion of ecological incidents associated with the use of diazinon up to 2002. The IRED indicates that approximately 239 (IRED Table 86) incidents were reported for diazinon across the United States in the Ecological Incident Information System (EIIS) and that from 1979 until 1998. During this time period, the number of reported incidents per year was increasing and the majority of reported incidents [where use was known] during this period was associated with diazinon use on turf.

As discussed earlier, a number of use restrictions have been imposed on diazinon subsequent to the interim reregistration eligibility decision. Although currently there is a total of 492 incidents associated with the use of diazinon, of which 79% are associated with effects on terrestrial animals [reported in the EIIS database] there has been a downward trend in the number of reported incidents since risk mitigation measures were imposed beginning in 2003. However, the lack of incident reports cannot be interpreted to mean the lack of incidents. **Figure 15** depicts the yearly number of reported incidents by incident type and illustrates that terrestrial incidents predominated while aquatic incidents, representing roughly 4% of the total reported incidents, were considerably less frequent. As indicated in the IRED, terrestrial incidents, primarily involving bird deaths, continued to show an increasing trend until 2002, after which time the number of reported incidents dropped precipitously. Since 2003 only 3 incidents have been reported, all of which have involved birds. Of the 163 terrestrial incidents where the treatment site is reported, the majority (80%) occurred from residential and turf uses, both of which are now cancelled. The last reported incident involving aquatic animals took place in 2003 and involved the death of 12 fish (I014322-001). For aquatic incidents where the treatment site is reported, roughly 45% have been associated with residential uses while 27% have been associated with orchard uses. The aquatic incident reported in 2003 did not report the treatment area.

No incidents involving the loss of Barton Springs salamanders, associated with the use of diazinon, are captured in the EIIS. The incident data as a whole suggest that mitigation efforts for diazinon have been effective in reducing the number of non-target mortality events. Where residential diazinon uses have been historically associated with a large number of incidents, those uses have been eliminated. While orchards have also been associated with a number of incidents and there are orchards in the BSSEA, aquatic exposure estimates from those uses result in RQ values well below acute risk LOCs for direct effects (acute mortality) in the Barton Springs salamander and the lack of incident data is consistent with the low risk estimates.

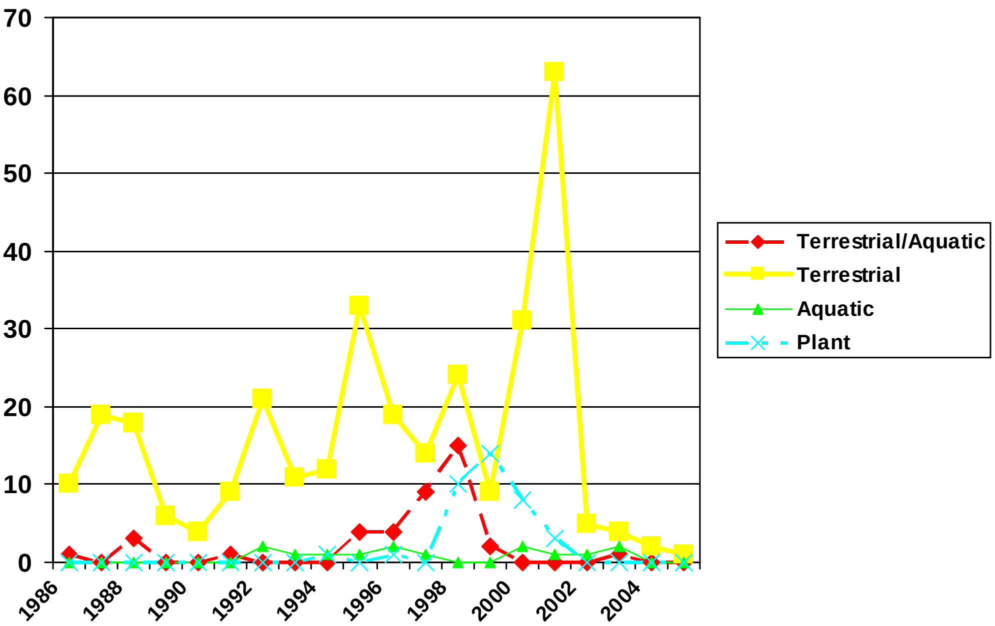


Figure 15. Total number of reported ecological incidents per year involving plants, aquatic animals, terrestrial animals and terrestrial/aquatic animals combined associated with the use of diazinon.

5.2.5 Description of Assumptions, Limitations, Uncertainties, Strengths and Data Gaps

5.2.5.1. Exposure Assessment

5.2.5.1.1 Aquatic exposure modeling of diazinon

Exposure modeling is characterized by the use of simplifying assumptions that allow complex systems to be described in manageable terms. The complexity of the Karst hydrology of the BSSEA increases the number of assumptions and uncertainties that usually characterize exposure modeling. For this assessment, all precipitation and applied diazinon in the contributing zone are assumed to have an equal chance of arriving at the recharge zone and all precipitation, applied diazinon, and discharge from the contributing zone are assumed to have an equal chance of arriving at the Barton Springs. All runoff and baseflow in the action area is assumed to recharge the Barton Springs and be available to dilute all diazinon concentrations in runoff. All four Barton Springs are assumed to receive recharge from the same sources.

Ground water baseflow from the Trinity aquifer is assumed to contribute 30% of the average flow from the contributing zone, although baseflow is likely to vary over time. All transit times across zones are assumed equal and instantaneous with negligible degradation between the edge-of-field and the Barton Springs. Losses from evaporation, transpiration, aquifer storage, stream flow that doesn't pass through the Springs, and withdrawal for drinking water are neglected.

Contributions from eroded sediment containing bound diazinon are assumed negligible. Contributions from overflow of Barton Creek during large stormflow are also assumed negligible. Spray drift contributions for applications in the action area are assumed negligible as well because of the conceptual model that assumes all runoff from treated areas that occurs in the recharge zone is instantaneously recharged and that applications are at sufficient distances from the Barton springs such that the exposed water in the springs is not directly impacted by spray drift.

The modeled use scenarios are assumed to represent actual use sites in the action area. The modeled runoff scenario is assumed to represent the entire action area where use does not occur, although the action area is approximately 43% residential.

Modeled exposure estimates were generated to reflect the maximum application practices allowed on current labels. Because actual diazinon usage may be less than that allowed on current labels, both in application practices and in percent of the action area where applied, modeled EECs may over-estimate exposure.

In this assessment, exposures are estimated for salamanders residing within the fracture system. Thus, salamanders residing within the fracture system of the springs are likely to be exposed to longer-term base flow concentrations of diazinon with occasional shorter duration pulses 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.

5.2.5.1.2 Other routes of exposure

5.2.5.1.2.1 Cattle ear tag exposure

As mentioned in the Problem Formulation, there is potential use of diazinon contained in cattle ear tags within the action area. The maximum potential release of diazinon from cattle ear tags in the action area is approximately 1000 lbs a.i. per year (2.8 lbs a.i./day). Most of the diazinon released from cattle ear tags is expected to volatilize, adsorb to the cow or to soil, or degrade, such that exposure to water bodies is expected to be minimal. Uncertainty in this assumption is based on uncertainty in the extent of cattle ear tag use in the action area, including the number of tagged cattle in the action area and the rate of tag replacement; the rate of diazinon emission from the tags; the magnitude of dissipation from the tags; and the likelihood of direct aquatic exposure when cattle are in close proximity to water bodies.

5.2.5.1.2.2 Atmospheric transport and deposition from sources outside of the action area

Diazinon is one of the most frequently detected of the organophosphate pesticides in air and in precipitation (USGS 1997). The majority of monitoring studies involving diazinon have been in CA; however, diazinon has been detected throughout the U.S. (**Table 18**). Magnitude of detected concentrations of diazinon in air and in precipitation can vary based on several factors, including proximity to use areas and timing of applications. In air, diazinon has been detected at concentrations 0.001-306.5 ng/m^3. Measured concentrations of diazinon in rain have ranged from 1.3 to 2,000 ng/. In fog, diazinon has been detected at 140-76,300 ng/L (Majewski and Capel, 1995). At this time, no air or precipitation monitoring data relevant to Texas have been located.

Potential diazinon use areas (*e.g.* agricultural lands) are located upwind of the Barton Springs. Available data indicate that prevailing winds in the Austin area originate from the south, with annual speeds of 9 miles per hour (NOAA 1998). Analysis of National Land Cover Data (NLCD 1992) from areas south of the action area indicate that agricultural lands (landcovers classified: row crops, small grains and fallow) are located within 30 miles upwind of Barton Springs (**Figure 16**). Ranges of diazinon transport in the Barton Springs area are unknown. Muir *et al*. (2004) estimated a half-distance (representing the distance traveled to reach a 50% decline in air concentration) of 440 (±153) miles for diazinon, based on empirical data from Canada. This group also estimated characteristic travel distances for diazinon of 1 to163 miles, depending upon model assumptions (*e.g.* related to precipitation, and degradation). Therefore, we cannot preclude that atmospheric transport of diazinon applied to areas that are 30 miles, or more, to the south of the Barton Springs action area could be deposited on the BSSEA. The extent to which this could reasonably result in potential exposure of the salamander to diazinon has not been assessed and remains an uncertainty.

Table 18. Diazinon detections in air and precipitation samples taken in the U.S.

Location	Year	Sample type	Maximum Conc.*	Detection frequency	Source
CA, MD	1970s-1990s	Air	306.5	N/A	Reported in Majewski and Capel, 1995
Mississippi River, from LA to MN	1994	Air	0.36	100%	Majewski *et al.* 1998
Solomons, MD	1995	Air	0.180	20 %	Harman-Fetcho *et al.* 2000
Sequoia National Park, CA	1996	Air	0.24	41.7%	LeNoir *et al.* 1999
Sacramento, CA (Franklin Field Airport)	1996-1997	Air	19.11	37.1 %	Majewski and Baston 2002
Sacramento, CA (Sacramento Metropolitan Area)	1996-1997	Air	12.25	46.5 %	Majewski and Baston 2002
Sacramento, CA (Sacramento International Airport)	1996-1997	Air	112.16	38.5 %	Majewski and Baston 2002
Fresno County, CA	1997	Air	290	N/A	State of California, 1998 a
Fresno County, CA	1998	Air	160	N/A	State of California,

Location	Year	Sample type	Maximum Conc.*	Detection frequency	Source
					1998 b
IA	2000-2002	Air	59.1	10 %	Peck and Hornbuckle 2005
Throughout US (including AR, CA IL, KS, KY, LA, ME, MS, MT, NM, NC, OH, OK)	1970s-1990s	Rain	2000	N/A	Reported in Majewski and Capel, 1995
Sequoia national Park, CA	1995-1996	Rain	19	57 %	McConnell *et al.* 1998
San Joaquin River Basin, CA	2001	Rain	908	100%	USGS 2003a
CA, MD	1970s-1990s	Fog	76300	N/A	Reported in Majewski and Capel, 1995
Parlier, CA	1986	Fog	18000	N/A	Glotfelty *et al.* 1990
Monterey, CA	1987	Fog	4800	N/A	Schomburg *et al.* 1991
Sequoia national Park, CA	1995-1996	Snow	14	62.5 %	McConnell *et al.* 1998

*For Air, ng/m^3, for rain, snow and fog, ng/L

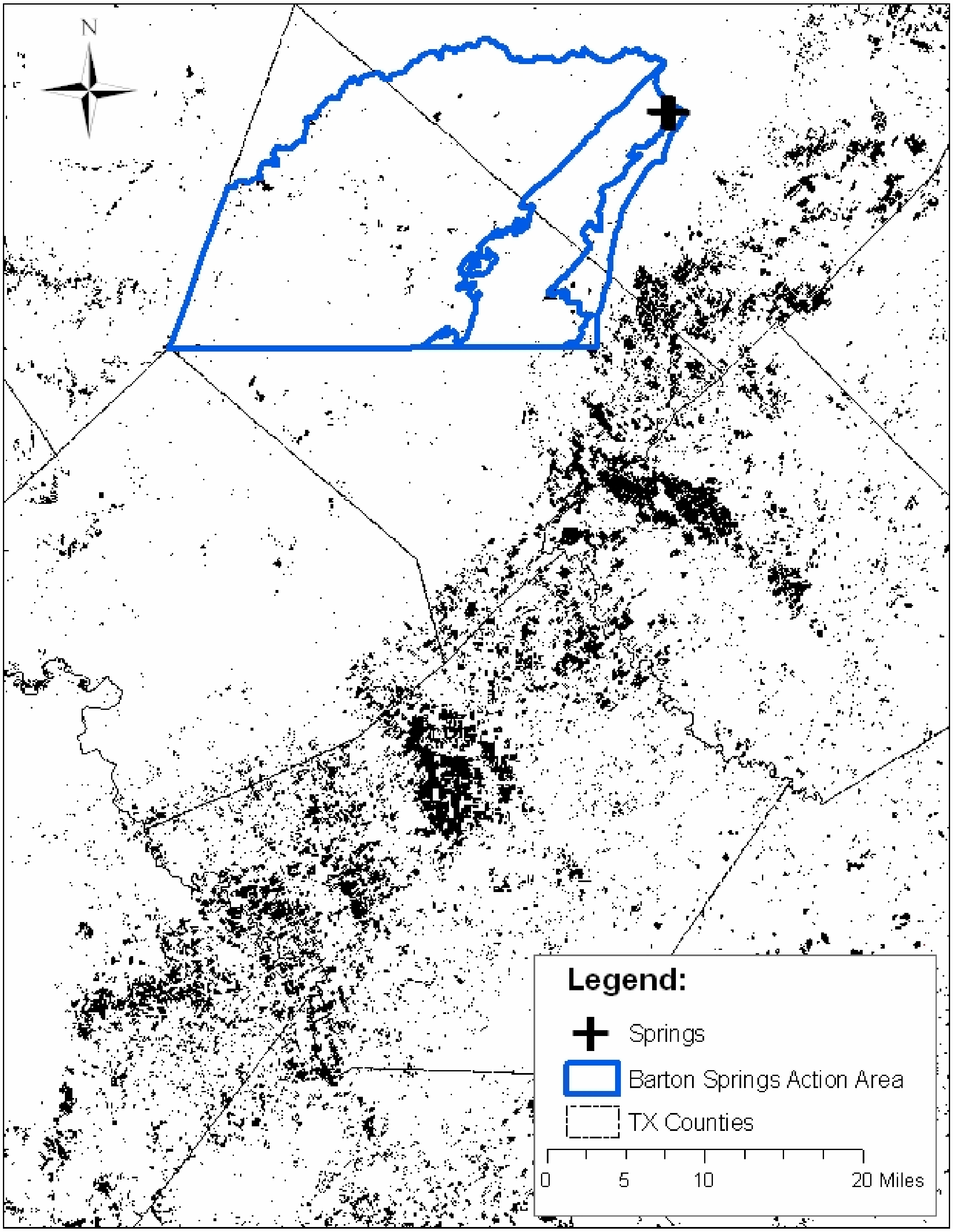


Figure 16. Map depicting agricultural land cover (black polygons) in relation to action area.

There are several potential mechanisms that can result in transport of diazinon from an application area to the atmosphere. These mechanisms include 1) volatilization from soil and plant surfaces in treated areas, 2) wind erosion of soil containing sorbed diazinon and 3) drift of diazinon during spray treatments of fields.

There are several factors which can influence volatilization of diazinon from a treated area, including: vapor pressure, adsorption to soil, incorporation depth, Henry's law constant, diffusion coefficients (Woodrow *et al*. 1997). Diazinon has a vapor pressure of 1.40×10^{-4} mm Hg @ 20°C. The vapor pressure and reported Henry's law constant of 1.40×10^{-6} atm m^3/mol would indicate that diazinon would volatilize from soil and water.

In a study involving diazinon, evaporation rates were estimated for 6 days after applying the pesticide to a fallow field at a rate of 1.5 kg a.i./ha (Majewski *et al*. 1990). Observations indicated that evaporation occurred at different rates throughout the first 4 days after application, with no evaporation observed on the 5th and 6th days after application. Reported evaporation rates at different time steps over the 4 days following the application ranged from <0.1 to 38 µg/m^2-h. These rates represent an hourly loss of <0.000067 to 0.025% of the total diazinon applied to the field. Average evaporation rates over the 4-day period after the application (which were calculated with no consideration of time weight) were 1.69-6.84 µg/m^2-h, which translate to an evaporation of 2.8-11.3% of the total mass of diazinon which was applied to the field.

As discussed in the environmental fate and transport assessment section, batch equilibrium studies indicated that diazinon is relatively mobile and not expected to adsorb to soils of low organic carbon content to a significant degree. Therefore, wind erosion of soils containing bound diazinon is expected to contribute little to the overall mass of diazinon that is transported atmospherically. In addition, it is assumed in this assessment that transport of diazinon through spray drift is negligible. Therefore, this route of transport is not considered.

Several studies are available involving monitoring of diazinon concentrations in lakes which are removed from agricultural areas and are presumed to receive inputs of diazinon from atmospheric deposition only. In a 1999-2001 study of several current use pesticides in Canada, diazinon was detected in lakes receiving runoff from agricultural areas (<0.003-2.8 ng/L), as well as remote lakes (≥50 km from agricultural areas) with no known inputs from agricultural runoff (<0.003-9.7 ng/L). No difference was detected between diazinon concentrations in the two types of lakes (Muir *et al.* 2004). Two 1997 studies (Fellers *et al.* 2004; LeNoir *et al.* 1999) measured diazinon concentrations in lake water in Kings Canyon and Sequoia National Parks (located in the Sierra Nevada Mountains in CA). The authors attributed these detections to atmospheric deposition from dry deposition and/or gas exchange from air samples of diazinon originating from agricultural sites located in California's Central Valley, which is up wind of the lakes. These studies indicate that atmospheric transport could represent a significant source of diazinon exposure to organisms in aquatic organisms. This exposure route alone could potentially pose a risk to invertebrates for acute exposures to invertebrates in these environments.

5.2.5.1.3 Degradates

As previously discussed in the effects assessment, the toxicity of the primary degradate of diazinon, oxypyrimidine, is assumed to be less than the parent compound; therefore, RQ values were not derived for exposures to this degradate.

Although data indicate that the toxicity of diazoxon is greater than that of the parent, RQ values were not quantified due to a lack of data useful for characterizing the persistence and transport properties of this degradate. It is possible that applications of diazinon could result in exposures of the salamander, its prey and its habitat to diazoxon. Given that this degradate is an order of magnitude more toxic to amphibians than the parent (Fellars and Sparling 2007), the degradate and parent combined could result in greater risk to the salamander than through direct or indirect effects from the parent compound alone. However, the effect endpoint (rainbow trout LC_{50}=90 μg/L) used to assess potential direct effects to the salamander is an order of magnitude more sensitive than the estimated toxicity of diazoxon to aquatic-phase amphibians (96-hr LC_{50}=760 μg/L) and is two orders of magnitude more sensitive that the estimated toxicity of the parent diazinon (96-hr LC_{50}=7488 μg/L) to aquatic-phase amphibians. Therefore, this assessment is considered protective for the potential increased toxicity of the diazoxon degradate to aquatic-phase amphibians.

Monitoring studies in CA have also detected diazoxon in air and precipitation samples (**Table 19**). In studies of diazinon and diazoxone concentrations in fog, diazoxone has been observed at greater concentrations than the parent (Schomburg *et al*. 1991). If diazinon and diazoxon are atmospherically transported and deposited within the Barton Springs, it is possible that the deposition of the degradate is greater than that of the parent. However, as indicated earlier, neither abiotic or biotic degradation studies of the parent conducted in the laboratory have demonstrated the formation of diazoxon; therefore, the conditions under which the oxygen analog may form is uncertain and at this point there are insufficient data with which to model exposure. Additionally, there are no monitoring data from the BSSEA that provide any information on diazoxon concentrations.

Table 19. Diazoxon detections in air and precipitation samples taken in the U.S.

Location	Year	Sample type	Maximum Conc.*	Source
CA	1980s-1990s	Air	10.8	Reported in Majewski and Capel, 1995
CA	1980s-1990s	Rain	115.8	Reported in Majewski and Capel, 1995
CA	1980s-1990s	Fog	28000	Reported in Majewski and Capel, 1995
Parlier, CA	1986	Fog	4800	Glotfelty *et al.* 1990
Monterey, CA	1987	Fog	11000	Schomburg *et al.* 1991

*For Air, ng/m^3, for rain, snow and fog, ng/L

5.2.5.1.4 Mixture Effects

This assessment considered only the single active ingredient of diazinon. However, the assessed species and their environments may be exposed to multiple pesticides simultaneously. Interactions of other toxic agents with diazinon could result in additive effects ($1/LC_{50mix} = 1/LC_{50Pesticide\_A} + 1/LC_{50Pesticide\_B}\ldots$), synergistic effects ($1/LC_{50mix} = 1/LC_{50Pesticide\_A} + 1/LC_{50Pesticide\_B}\ldots x\ Y$; where $Y > 1$) or antagonistic effects ($1/LC_{50mix} = 1/LC_{50Pesticide\_A} + 1/LC_{50Pesticide\_B}\ldots\ x\ Y$; where $Y < 1$). Conceptually, the combined effect of the mixture is equal to the sum of the effects of each stressor (1 + 1 = 2) for additive toxicity. Synergistic effects occur when the combined effect of the mixture is greater than the sum of each stressor (1 + 1 >2), and antagonistic effects occur when the combined effect of the mixture is less than the sum of each stressor (1 + 1 <2).

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

5.2.5.2 Effects Assessment

5.2.5.2.1 Direct Effects

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. While a limited amount of amphibian data are available, these studies either failed to establish an LC_{50} value or did not report measured concentration values. The available data suggest that amphibians are considerably less sensitive to diazinon than fish; however, these data also demonstrated that frogs are 10-times more sensitive to diazoxon than to the parent. To the extent to which amphibians are less sensitive than the surrogate species used in this assessment, the assessment is overly conservative. By the same token though, to the extent to which diazoxon is present in runoff from treated area, the assessment is less conservative in estimating potential effects. This assessment though is considered to be conservative since the effects endpoint, *i.e.*, rainbow trout 96-hr LC_{50}=90 μg/L, used to assess potential acute effects to the salamander is two orders of magnitude more sensitive than similar estimates for the toxicity of diazinon to aquatic-phase amphibians and is an order of magnitude more sensitive than the estimate of the toxicity of diazoxon to aquatic-phase amphibians..

5.2.5.2.2 Sublethal Effects

Open literature was useful in identifying sublethal effects associated with exposure to diazinon. These effects included but were not limited to decreased response from olfactory epithelium, effects on heat shock proteins, decreased acetylcholine esterase activity, and effects on endocrine-mediated processes. However, no data are available to link the sublethal measurement endpoints to direct mortality or diminished reproduction, growth and survival that are used by OPP as assessment endpoints. While the study by Scholz *et al*. 2003 attempted to relate the results of olfactory perfusion assays to decreased predator avoidance and homing response in salmon, the study results are not sufficiently vetted to establish a clear dose-dependent relationship. OPP acknowledges that a number of sublethal effects have been associated with diazinon exposure; however, at this point there are insufficient data to definitively link the measurement endpoints to assessment endpoints. To the extent to which sublethal effects are not considered in this assessment, the potential direct and indirect effects of diazinon on Barton Springs salamanders may be underestimated.

5.2.5.2.3 Indirect Effects

Indirect effects on the Barton Springs salamander are estimated based on the most sensitive invertebrate tested, *i.e.*, *Ceriodaphnia dubia*. While this is a relatively common invertebrate, cladocerans do not appear to be a major food source for Barton springs salamanders based on stomach content analyses. However, while ostracod exoskeletons have been identified in the stomachs Barton Springs salamanders, these invertebrates would be relatively easy to discern whereas cladocerans may not. Thus, the extent to which the most sensitive species used in this analysis is representative of the diet of Barton Springs salamanders is uncertain. However, it should be noted that the toxicity endpoints for surrogate organisms are not intended to represent specific taxa but rather they serve as indicators of the potential sensitivity of invertebrates as a whole.

5.2.5.2.4 Species Sensitivity Distributions

In order to characterize the conservativeness of the endpoints selected to represent direct effects to the salamander (*e.g.* rainbow trout LC_{50} = 90 μg/L), and indirect effects to the salamander through direct effects to its prey (*e.g. Ceriodaphnia dubia* EC_{50} = 0.21 μg/L) species sensitivity distributions were derived using the available acute toxicity data for freshwater fish and invertebrates, respectively.

Two sets of distributions were established for each group: quantitative and qualitative. Data were considered useful for the quantitative distributions if they were classified acceptable or supplemental. Data included in the qualitative distributions were those considered qualitative as well as additional data identified in ECOTOX. Data available in ECOTOX were taken directly from the database, not from their original citations. Once a data set was assembled, the average of the Log10 values of the LC_{50} values for a species was calculated. Then, the average of the Log10 values of the genera was estimated. A normal distribution was used to estimate the species sensitivity distribution by considering the mean and standard deviation of all genus mean values. A full description of the data and results used to derive these distributions is included in **Appendix F**.

In order to consider the distribution in context of the exposure and the LOC, the maximum aggregate peak exposure (0.058 µg/L) was divided by the LOC (0.05) for acute exposures. This concentration of 1.16 µg/L represents the maximum value of the EC_{50} that would result in an exceedance of the LOC. In other words, an EC_{50} greater than 1.16 µg/L would not result in direct or indirect effects to the salamander.

The number of data points, species and genera incorporated into each of the four species sensitivity distributions are identified in **Table 20**. The curves of the species sensitivity distributions are represented by **Figures 17 - 20**. In the figures, each point represents the genus mean value for the respective species and the solid line represents the sensitivity distribution based on these data. The distributions include a dashed line, which represents the adjusted exposure concentration of 1.16 µg/L.

Table 20. Numbers of data points, species and geneses incorporated into each of the four species sensitivity distributions.

Taxa	Quantitative/qualitative	Number of Data Values	Number of Species	Number of Genera	Lower 95th Percentile (µg/L)
Fish	Quantitative	11	9	7	139
	Qualitative	41	17	14	126
Invertebrates	Quantitative	9	7	6	0.13
	Qualitative	49	14	12	0.31

The lower 95th percentile of the quantitative fish distribution (139 µg/L) indicates that the use of the lowest available toxicity value (90 µg/L) is likely a conservative estimate of the toxicity of diazinon to freshwater vertebrates. When considering the weighted exposure value, there is risk to sensitive species below the 5th percentile of the distribution.

The lower 95th percentile of the quantitative invertebrate distribution (0.13 µg/L) indicates that the use of the lowest available toxicity value (0.21 µg/L) is not as conservative as the value used for fish. It is however, within the lower 90th percentile of sensitive species (<0.26 µg/L). When considering the adjusted exposure value, there is risk to approximately 30% of invertebrate species for which there are quantitative data.

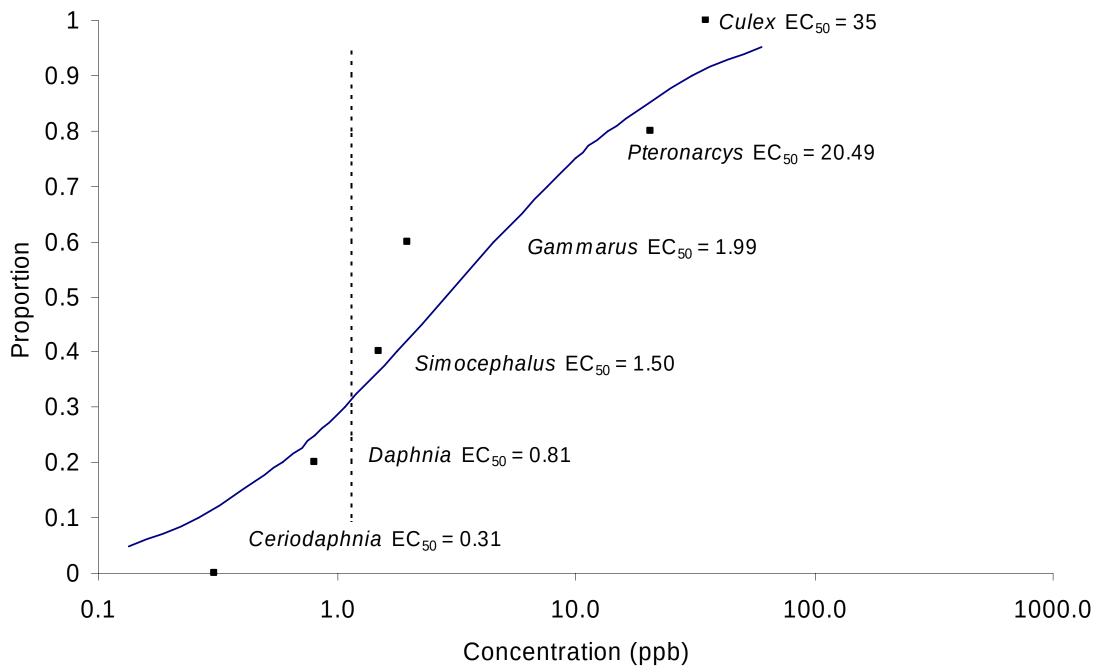


Figure 17. Invertebrate species sensitivity distribution of toxicity data considered useful for quantitative purposes. The dashed line represents the adjusted exposure (peak EEC/LOC).

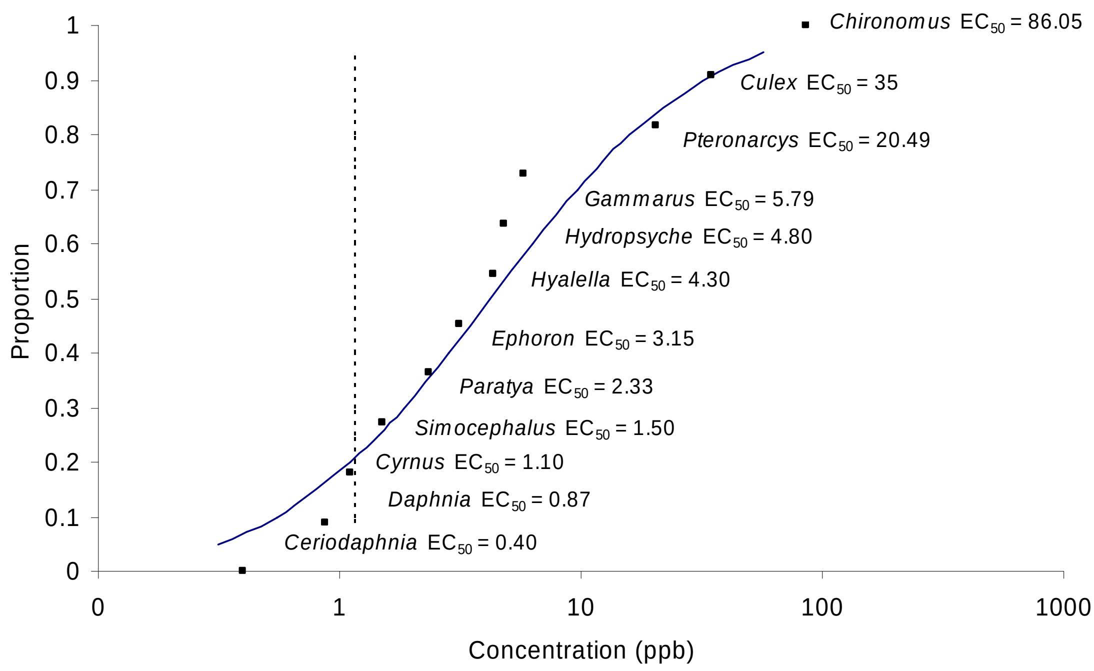


Figure 18. Invetebrate species sensitivity distribution of toxicity data considered useful for qualitative purposes. The dashed line represents the adjusted exposure (peak EEC/LOC).

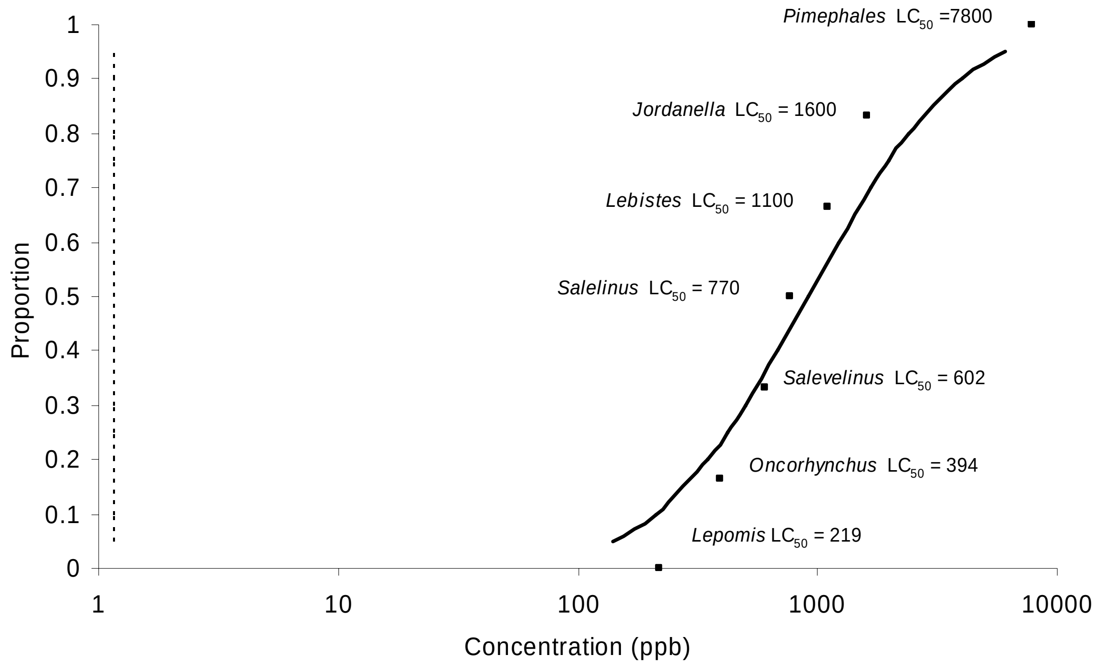


Figure 19. Fish species sensitivity distribution of toxicity data considered useful for quantitative purposes. The dashed line represents the adjusted exposure (peak EEC/LOC).

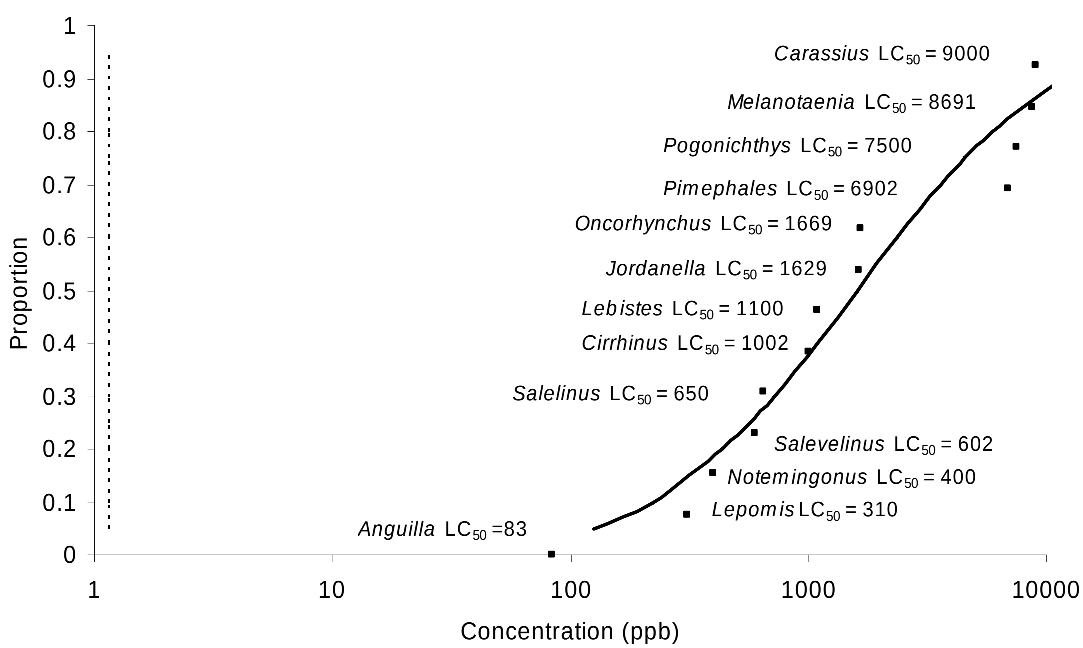


Figure 20. Fish species sensitivity distribution of toxicity data considered useful for qualitative purposes. The dashed line represents the adjusted exposure (peak EEC/LOC).

5.3. Conclusions

The conceptual model for potential risks of diazinon use to Barton Springs salamanders (**Figure 4**) depicts direct and indirect changes in receptor attributes. Biological receptors included the Barton Springs salamander, aquatic invertebrates that serve as the salamanders' forage base for the salamander, and aquatic plants that serve as habitat/cover for the species and its prey. Potential attribute changes for these receptors included decreased survival, reproduction and growth. An assessment of potential sources (routes of exposure) for diazinon estimates peak exposure concentrations in the Barton Springs at 0.06 μg/L and chronic 1-in-10 year average 60-day chronic exposure is estimated at 0.003 μg/L. These exposure estimates combined with acute (90 μg/L) and chronic (<0.55 μg/L) toxicity estimates for the most sensitive species result in a no effect determination for direct acute effects on the salamander and a may affect but not likely to adversely affect determination for chronic effects to the salamander since the potential effects are discountable (**Table 21**). Potential chronic effects were considered discountable since the measurement endpoint (NOEC) would have to decrease by roughly three orders and magnitude in order to exceed the Agency's chronic risk LOC for endangered species. The available chronic toxicity data indicate that while growth appeared to be impaired in the chronic toxicity study, survival was not impaired. Additionally, monitoring data collected subsequent to the cancellation of all residential uses and many of the agricultural uses of diazinon indicate diazinon [within Barton Springs] is below the level of detection. These data suggest that remaining uses of diazinon in the BSSEA are likely lower than the conservative assumptions (26 applications/year) made for ornamental/nursery uses and that the potential for chronic exposure is low.

For indirect effects on the salamander's forage base, the estimated peak concentration (0.06 μg/L) was compared to the most sensitive invertebrate toxicity estimate (0.21 μg/L). Although the resulting risk quotients for the use of diazinon on ornamental plants/nurseries exceeded the endangered species level of concern, the likelihood of individual effect (0.02%) and the availability of less sensitive species that are known to be forage items for the salamander resulted in a may affect but not likely to adversely affect determination since the effect is considered discountable (**Table 21**).

For indirect effects to habitat, the peak estimated environmental concentration (0.06 μg/L) was compared to the most sensitive aquatic plant species (66 μg/L) and the resulting risk quotient was below the acute risk LOC. The result is a no effect determination for habitat (**Table 21**).

Although there are a number of uncertainties in this assessment, the approaches used to estimate potential exposure and effects are considered relatively conservative and protective for the species. Based on the may affect but not likely to adversely effect determinations for direct chronic effects and indirect effects, an informal consultation with the U. S. Fish and Wildlife Service under Section 7 of the Endangered Species Act is warranted.

Table 21. Diazinon Effects Determination Summary for the Barton Springs Salamander.

Assessment Endpoint	Effects Determination	Basis for Determination
Acute mortality	No effect	Acute LOC is not exceeded based on the most sensitive surrogate freshwater vertebrate data.
Chronic survival, growth, and reproduction effects on Barton Springs salamander individuals via direct effects	May affect but not likely to adversely affect	Although there is uncertainty regarding the potential for chronic effects on growth since available chronic toxicity data fail to establish a definitive chronic NOEC, estimated environmental concentrations are sufficiently low to render the likelihood of chronic effects low and as such are considered discountable.
Indirect effects to Barton Springs salamander via reduction of prey (*i.e.*, freshwater invertebrates)	May affect but not likely to adversely affect	Acute risk to endangered species LOCs are exceeded based on the most sensitive aquatic invertebrates evaluated; however, the likelihood of individual effects is low and as such are considered discountable.
Indirect effects to Barton Springs salamander via reduction of habitat and/or primary productivity (*i.e.*, aquatic plants)	No effect	Diazinon use does not directly affect individual non-vascular aquatic plants in Barton Springs. Estimated peak EECs for all modeled diazinon use scenarios within the action area are well below the threshold concentration for aquatic, non-vascular plants. Although there are no toxicity data for aquatic vascular plants, the data for nonvascular aquatic plants and vascular terrestrial plants and the lack of any reported field incidents involving plants indicate that plants are less sensitive to diazinon than animals.

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Appendix A. ECOTOX Open Literature Reviews.

A total of 2,335 references were identified for diazinon in a search of ECOTOX conducted in September 2006 Of these, approximately 27 studies contained toxicity endpoints that were more sensitive than those listed in the 2002 IRED. Reprints for each of these studies were reviewed to determine whether the studies could be used either quantitatively or qualitatively to describe the potential effects of diazinon on aquatic organisms. Below is a brief description of each of the studies along with any uncertainties that were identified during the review. The bolded number preceding each of the citations represents the ECOTOX reference number.

ECOTOX Record Number and Citation: 18129 Werner, I. and R. Nagel. 1997. Stress Proteins HSP60 and HSP70 in three Species of Amphipods Exposed to Cadmium, Diazinon, Dieldrin and Fluoranthene. Environmental Toxicology and Chemistry. 16(11): 2393 – 2403.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Article reports 24-hr LC_{50} value determined as part of a range finding test for measuring response of heat shock proteins. Diazinon concentrations determined using immunoassay (EnviroGard test kit; Millipore, Bedford, MA). Three replicate test containers each containing 150 mL. Control and solvent controls run; no solvent used for diazinon. Ten test species (freshwater *Hyalella azteca* and the marine *Rhepoxynius abronius*); 20 estuarine *Ampelisca abdita* because of smaller size. Filtered (0.22 µm) dilution water obtained from Bodega and San Francisco bays for saltwater and freswater studies. Dissolved oxygen 6.9 – 9.0 mg/L; pH ranged from 7.7 to 8.4.

	24-hr	48-hr
H. azteca	30 µg/L	19 µg/L
A abdita	21 µg/L	10 µg/L
R. abronius	9.2 µg/L	--

Remainder of study examines heat shock protein responses; the relevancy of these data to assessment endpoints is not determined quantitatively.

Description of Use in Document (QUAL, QUAN, INV): Qualitative

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ECOTOX Record Number and Citation: 15687 Sancho, E., M. D. Ferrando, M. Gamon and E. Andreu-Moliner. 1994. Uptake and Clearance of Diazinon in Different Tissues of the European Eel (*Anguilla anguilla* L.) Biomedical and Environmental Sciences 7: 41 – 49.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Study is deemed to be of low utility:

Wild-caught eels
Test animals did not respond to food and therefore may have been fasted for 2 weeks before the study and during the 96-hr study.
Tap water is used.
No mention is made whether concentrations are measured therefore, the concentrations are presumed to be nominal; the accumulation study did measure concentrations though.
Aquaria are aerated.

	24-hr	48-hr	72-hr	96-hr
A. anguilla	164 µg/L	114 µg/L	92 µg/L	85 µg/L

Description of Use in Document (QUAL, QUAN, INV): Qualitative

Rationale for Use: Eels are not the most sensitive speices tested with diazinon. The study provides useful information for qualitative species sensitivity distribution.

Limitations of Study: The fact that the test animals were essentially fasted for at least 2 weeks prior to test initiation raises serious concerns regarding the utility of these data. Extensive fasting would likely mobilize the animal's fat reserves. Given the uncertain chemical exposure history for the eels, it is uncertain what effect the fasting may have on the study's ability to detect treatment effects.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist
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ECOTOX Record Number and Citation: 1055 Ferrando, M. D., E. Sancho, and E. Andreu-Moliner. 1991. Comparative Acute Toxicities of Selected Pesticides to *Anguilla anguilla*. Journal of Environmental Science and Health B26: 491 – 498.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Wild-caught eels (Albufera Lake, Valencia, Spain)

Acclimatized for 2 weeks; however, animals did not respond to feeding attempts.
Glass aquaria (40 L) containing 35 L test solution; 4 replicates with10 fish per replicate per treatment. (Diazinon 92% a.i.) Controls run. No mention of whether concentrations were measured.

	24-hr	48-hr	72-hr	96-hr
A. anguilla	160 µg/L	110 µg/L	90µg/L	80µg/L

The results of this study are strikingly similar to results reported in the 1994 publication by Sancho. It is unclear whether this is the same study.

Description of Use in Document (QUAL, QUAN, INV): Qualitative

Rationale for Use: Eels are not the most sensitive speices tested with diazinon. The study provides useful information for qualitative species sensitivity distribution.

Limitations of Study: The fact that the test animals were essentially fasted for at least 2 weeks prior to test initiation raises serious concerns regarding the utility of these data. Extensive fasting would likely mobilize the animal's fat reserves. Given the uncertain chemical exposure history for the eels, it is uncertain what effect the fasting may have on the study's ability to detect treatment effects.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist

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ECOTOX Record Number and Citation: 16043 Norberg-King, T. J. 1987. Toxicity Data on Diazinon, Aniline and 2, 4-Dimethylphenol. Memo to Charles Stephan, ERL Duluth from the U.S. EPA Environmental Research Laboratory in Duluth.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Summary of diazinon (85% ai) acute (48-hr) toxicity tests with *Ceriodaphnia dubia* (in-house culture; <24 hrs old) using water from various sources: Lake Superior water (LSW), reconstituted water (RCW), diluted mineral artificial water (DMW) and Lake Superior culture water (water enriched by previous goldfish use). Daphnia in most of the studies were fed using green algae *Pseudokirchneriella subcapitata* (formerly *Selenastrum capricornutum*) and yeast concentrate. Test volumes of 12.5 ml in replicate with two replicates per test concentration. Diazinon dissolved in methanol

	48-hr
DMV	0.57 µg/L
LSW	0.66 µg/L
RCW	0.57 µg/L
LSCW	>1.0 µg/L

Limitations of Study: Concentration of methanol is not reported. It is unclear whether the control is a solvent control or neat control. Some studies had concentrations measured in the treatment units while others measured diazinon in the stock solutions.

A 7-day chronic toxicity study is also reported using one daphnid (<6-hr old) in 15 ml of test solution (DMW) with 10 reps per treatment concentration; solutions renewed daily and all concentrations were measured.

NOEC = 0.22 μg/L; LOEC = 0.34 μg/L (mean number of young/female).

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of freshwater nonvascular aquatic plants to diazinon.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist

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ECOTOX Record Number and Citation: 16547 Oh, H. S., S. K. Lee, Y. H. Kim and J. K. Roh. 1991. Mechanism of Selective Toxicity of Diazinon to Killifish (*Oryzias latipes*) and Loach (*Misgurnus anguillicaudatus*). Aquatic Toxicology and Risk Assessment: Fourteenth Volume, ASTM STP 124. M. A. Mayes and M G. Barron (editors), American Society for Testing and Materials. Pp 343 – 353.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Study reports a 96-hr LC_{50} value for killifish (LC_{50}= 3,910 μg/L) and loach (LC_{50}=270 μg/L); however, the methods section does not indicate that any such test was undertaken.

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of fish to diazinon.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist

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ECOTOX Record Number and Citation: 821 Ankley, G. T., J. R. Dierkes, D. A. Jensen, and G. S. Peterson. 1991. Piperonyl Butoxide as a Tool in Aquatic Toxicologicl Research with Organophosphate Insecticides. Ecotoxicology and Environmental Safety 21 (3): 266 – 274.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: *Ceriodaphnia dubia*, *Daphnia magna* and *Daphnia pulex* obtained from in-house cultures; all test organisms $\leq$48 hrs old. Five organisms per test replicate, two replicates per treatment with 10 mL per treatment container. Tests conducted at 25oC; control used 10% mineral water (Perrier, Vergeze, France) diluted in high purity water from a Millipore system.

	48-hr LC_{50}
C. dubia	0.50 µg/L
D. magna	0.80 µg/L
D. pulex	0.65 µg/L

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of freshwater invertebrates to diazinon.

Limitations of Study: Specific purity of diazinon is not provided; report simply cites purities ranging from 95 to 99%. Test concentrations are nominal. Methanol is used as a co-solvent; report states that concentration did not exceed 1.5% and this is "well below" the 48-hr LC_{50} for methanol. However, no solvent control is run and it is unclear why the control contained 10% mineral water.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist

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ECOTOX Record Number and Citation: 4009 Fernández-Caladerrey, A., M. D. Ferrando and E. Andreu-Moliner. 1994. Effect of Sublethal Concentrations of Pesticides on the Feeding Behavior of *Daphnia magna*. Ecotoxicology and Environmental Safety 27: 82 – 89.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: *Daphnia magna* from the Laboratory for Biological Research in Aquatic Pollution (Gent, Belgium) and cultured in laboratory. Diazinon 92% ai was dissolved in acetone. Study procedure according to EEC standard. Six concentrations plus a control acetone (0.06 mg/L) consisting of 3 replicates with 10 neonates (<24 hr old) placed in 30 ml glass beaker containing 25 ml test solution. Animals were fasted and study was conducted under static conditions.

	24-hr LC_{50}
D. magna	0.9 µg/L diazinon
	0.62 mg/L endosulfan

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of freshwater invertebrates to diazinon and endosulfan.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist
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ECOTOX Record Number and Citation: 5311 Dennis, W. H., A. B. Rosencrance and W. F. Randall. 1980. Acid Hydrolysis of Military Standard Formulations of Diazinon. Journal of Environmental Science Health, Part B. Pestic Food Contam. Acric. Wastes, B15(1): 47 – 60.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Young-of-the-year bluegill sunfish (*Lepomis macrochirus*; 0.8 g) from an unspecified source were exposed to diazinon (88.1% ai) for 96 hrs in a static system. Five-gallon glass jars containing 15 L treatment solution and contained 10 fish per rep and three reps per treatment. Mortality and treatment concentrations were measured every 24 hours. Well water used in study with alkalinity of 138 mg/L as $CaCO_3$.; temperature 20 $\pm$ 1oC

	96-hr LC_{50}
Bluegill	120 µg a.i./L

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of freswater fish to diazinon.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist

Limitations of Study: In this study technical grade diazinon is more toxic than the formulated products tested (Diazinon EC; LC50 530 µg a.i/L
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ECOTOX Record Number and Citation: 885 Sanders, H. O. 1969. Toxicity of Pesticides to the Crustacean *Gammarus lacustris*. Technical Papers of the Bureau of Sport Fisheries and Wildlife. U. S. Department of the Interior, Fish and Wildlife Service, Bureau of Sport Fisheries and Wildlife, Washington DC.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Laboratory stock cultured from scuds (Gammarus lacustris) collected at pond near the Fish-Pesticide Research Laboratory (Denver, CO). Reconstituted water (pH = 7.1; alkalinity = 30 ppm). Glass aquariums (5.7 L) containing 4 L of tests water. Ten 2-month old scuds placed in each aquarium; then 2 hours later, test material was added to aquaria. Test conducted at 21oC (70oF) Appears that only neat control and not a solvent (ethanol) control was run. Procedure indicates that emulsifiable concentrates and wettable powders were dissolved in deionized water while technical grade pesticides were dissolved in ethanol; however the article does not discuss what form the diazinon was in. Ethanol concentration never exceeded 1 mL per liter; however, 1 ml/l is a very high concentration of co-solvent. The endpoints reported in the study are no more sensitive than what is already reported for aquatic invertebrates..

	24-hr	48-hr	96-hr
Scud	800 µg/L	500 µg/L	200 µg/L

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of freshwater invertebrates to diazinon.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist

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ECOTOX Record Number and Citation: 18190 Bailey, H. C., J. L. Miller, M. J. Miller, L. C. Wiborg, L. Deanovic and T. Shed. 1997. Joint Acute Toxicity of Diazinon and Chlorpyrifos to *Ceriodaphnia dubia*. Journal of Environmental Toxicology and Chemistry. 16(11): 2304-2308.
Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Diazinon (99% ai) dissolved in 100% methanol. Dilution water obtained from everse osmosis-treated well water brought to moderately hard standard. Nominal test concentrations of 0.05, 0.10, 0.20, 0.40 and 0.80 µg/L. *Ceriodaphnia dubia* (<24 hr old) obtained from in-house laboratory culture. Exposures conducted in 20-l glass scintillation vials containing 18 ml of solution. Four replicates containing five neonates in each used at each of the five test concentrations; studies were static tests as 25 + 1oC with a 16 hr day and 8 hr night photoperiod. Initial concentrations of diazinon determined through ELISA. Animals fasted through study period.

	24-hr	48-hr	72-hr	96-hr
Ceriodaphnia	0.75 µg/L	0.48 µg/L	0.40 µg/L	0.35 µg/L
	0.58 µg/L	0.58 µg/L	0.35 µg/L	0.32 µg/L

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of fish to diazinon.

Limitations of Study: This study has a relatively good methodology; however, diazinon was dissolved in methanol and the final concentration of methanol is not reported. Also, a solvent control is not reported.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist

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ECOTOX Record Number and Citation: 19300 Harris, M. L., C. A. Bishop, J. Struger, B. Ripley and J. P. Bogart. 1998. The Functional Integrity of Northern Leopard Frog (*Rana pipiens*) and Green Frog (*Rana clamitans*) Populations in Orchard Wetland. II. Effects of Pesticides and Eutrophic Conditions on Early Life Stage Development. Environmental Toxicology and Chemistry 17(7): 1351 – 1363.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Leopard frog adults obtained from R. Elinson (Hazen Frog Farms, Alburg, VT) and from wild-caught adults. Green frog adults were wild-caught. Animals were induced with 0.1 µg luteninzing hormone-releasing hormone or with whole frog or toad pituitary extracts.

Laboratory assays conducted in 250-ml beakers maintained at 19.5 $\pm$ 1.5oC for leopard frogs and 19.5 $\pm$ 0.6 and 18.6 $\pm$ 0.6oC for green frog assays. Photoperiod of 12:12 hr light:dark maintained. Beakers contained 10 individuals with 2 or 3 replicates per treatment. Tests initiatated at 9 hours post-fertilization (Gosner developmental stage 8/9). Larvae fed boilded lettuce (0.5 g) every other day; rations were increased to 1 g after approximately 1 week. Tests continued for 2 weeks (1993) for both species and for 3 weeks (1994) with green frogs. At test temination, survival, hatching success and tadpole growth rates determined.

Green frogs (Gosner stage 8 embryos through stage 25 tadpoles) were also continuously exposed for 13-day static renewal (4 day) toxicity tests to Basudin® 500 EC and technical grade diazinon. After 4 days, treatment solutions were replaced with reference pond water and embryos hatched and began feeding in "uncontaminated" conditions. After 7.5 day in reference water (with renewal every second day) treatment solutions were reintroducted. Treatment concentrations of Basudin® 500EC were 0.001, 0.01, 0.1, 1.0, 10 and 25 µg/L; treatment concentrations for technical grade diazinon were: 0.5, 5 and 50 µg/L. Results presented below are for technical grade diazinon; formulated end-product appears to be less toxic than the technical grade.

	96-hr LC_{50}	16-day LC_{50}
Green Frog	>50 µg/L	5 µg/L

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of aquatic-phase amphibians to diazinon.

Limitations of Study: Laboratory studies appeared to be conducted using reference pond water; however, background pesticide residues were not analyzed at the time of the study. It is also unclear whether controls were run. The 16-day study was with feeding.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist

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ECOTOX Record Number and Citation: 3664 Culley, D. D. and D. E. Ferguson. 1969. Patterns of Insecticide Resistance in the Mosquitofish, *Gambusia affinis*. J. Fish. Res. Board Can 26(9): 2395-2401.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Wild-caught fish from a drainage canal near Belzoni, MS, acclimatized for 1 – 5 days. Fish apparently had fungal infection prior to use and required treatment with malachite green and noniodized table salt. Fish fasted 24-hr prior to testing. Diazinon dissolved in acetone. Test containers were 1-gal jars containing 2.5 l of treatment solution in replicate with 6 fish in each jar (approximately 0.5 g fish/liter).

Limitations of Study: None of the pesticides tested appear to be diazinon or its degradate (diazoxon).

Primary Reviewer: Thomas Steeger, Ph.D., Senior Scientist

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ECOTOX Record Number(s) and Citation: 6221.and **11219** Sancho, E., M. D. Ferrando, E. Andreau and M. Gamon. 1992. Acute Toxicity, Uptake and Clearance of Diazinon by the European Eel, *Anguilla anguilla* (L). J. Envion. Sci. Health. B27(2): 209 – 221.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Wild-caught eels (20 – 30 g; 16 – 20 cm) obtained from Albufera Lake (Valencia, Spain) and acclimated to laboratory conditions for 2 weeks. Eels did not respond to feeding attempts but appeared healthy. Animals were not fed during the 96-hr toxicity study. Diazinon (95% ai) prepared in acetone and presumably diluted with tap water. Glass aquaria (40 l) containing 35 l of test solution; solvent control run with 65 µl acetone/l. Ten eels per replicate and four replicates per treatment were tested.

	24-hr	48-hr	72-hr	96-hr
European eel	160 µg/L	110 µg/L	90 µg/L	80 µg/L

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of eels to diazinon.

Limitations of Study: Prior chemical exposure (other than diazinon) history is unknown; animals would have been fasted for roughly 3 weeks and likely have mobilized fat reserves where chemical residues may have been present although study claims that diazinon was not detected in the eel prior to exposure.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist

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ECOTOX Record Number and Citation: 7004 and 11438 Sancho, E., M. D. Ferrando, E. Andreu and M. Gamon. 1993. Bioconcentration and Excretion of Diazinon by Eel. Bull. Enviorn. Contam. Toxicol. 50: 578 – 585.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Wild-caught eels (20 – 30 g; 16 – 20 cm) obtained from Albufera Lake (Valencia, Spain) and acclimated to laboratory conditions for 2 weeks. Eels did not respond to feeding attempts but appeared healthy. Animals were not fed during the 96-hr toxicity study. Diazinon (95% ai) prepared in acetone and presumably diluted with tap water. Glass aquaria (40 l) containing 35 l of test solution; solvent control run with 66 µl acetone/l. Ten eels per replicate and four replicates per treatment were tested.

	24-hr	48-hr	72-hr	96-hr
European eel	160 µg/L	110 µg/L	90 µg/L	80 µg/L

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of eels to diazinon.

Limitations of Study: Prior chemical exposure (other than diazinon) history is unknown; animals would have been fasted for roughly 3 weeks and likely have mobilized fat reserves where chemical residues may have been present although study claims that diazinon was not detected in the eel prior to exposure. Essentially the same reference/study as #**6221** and #**11055.**

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist

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ECOTOX Record Number and Citation: 66119. Parkhurst, M A., G. Whelan, Y. Onishi and A. R. Olsen. 1981. Simulation of the Migration, Fate and Effects of Diazinon in two Monticello Stream Channels. Battelle, Pacific Northwest Laboratories Report to the U. S. Army Medical Bioengineering Laoboratory, Fort Dietrick, Frederick, MD. Contract 2311104483.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Only secondary data are cited in the document (Table 3.14). According to the document, the Monticello Experimental Research Station (MERS) borrowed "extensively" from data they had gathered. The primary sources of data are

ECOTOX Record Number and Citation: Sparling, D. W. and G. Fellers. 2006 Comparative toxicity of chlorpyrifos, diazinon, malathion and their oxon derivatives to larval *Rana boylii*. Environmental Pollution (Article in Press; available online at www.sciencedirect.com).

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Wild-caught foothill yellow-legged frog (*Rana boylii*) egg masses (3) collected from a Coast Range stream. Eggs hatched under laboratory conditions in 78 L aquaria for several weeks prior to test initiation. During acclimation, larvae fed boiled organic romaine lettuce and high-protein fish flakes *ad libitum*.

Chloropyrifos, diazinon and malathion and their respective oxons were reagent grade (99% pure) and purchased from Arco Organics (Morris Plains, NJ). Chemicals were dissolved in acetone. Aquaria (8 L) filled with 7 L of reconstituted water; treatment concentrations are nominal. To each aquarium, 9 "same-aged" *R. boylii* tadposles ranging in developmental stage from Gosner 32 to 44. After the first 24 hr of exposure, tadpoles were fed a small amount of organic romaine lettuce.

Total cholinesterase activity determined via a colormetric method of Ellmann *et al* (1961)[1]. Cholinesterase levels were normalized to that of a metamorph by multiplying by 2.4, 1.9 and 1.6 for tadpoles falling into stages 32 – 36, 37 – 39 and 40 – 45, respectively, to account for what the authors claim is an increase in chloinesterase activity with developmental stage of tadpoles.

Probit dose-response curve results for chlorpyrifos, diazinon, malathion and their respective oxygen analogs (oxons) in *R. boylii*.

Chemical	Period	Slope	LC_{50}	95% Confidence Interval
Chlorpyrifos	24	17.018	3.005	0.993 – 157
Diazinon	96	3.374ns	7.488	NA
Diazoxon	96	14.077	0.760	0.336 – 3.212
Malathion	96	31.477 ns	2.137	NA
Maloxon	96	133.659	0.023	0.014 – 0.180

ns not significant
NA – not available

Regression results of normalized cholinesterase acitivty against concentration for chlorpyrifos, diazinon, malathion and their respective oxygen analogs (oxons) in *R. boylii*.

Chemical	N	Slope	Intercept	R2
Chlorpyrifos	46	-0.0330	0.8499	0.1383
Chloroxon	9	-26.8088	1.2525	0.2547
Diazinon	20	-0.0796	1.2169	0.1729
Diazoxon	45	-0.0511	0.8504	0.0908
Malathion	28	-0.1028	1.0534	0.2244
Maloxon	27	-24.5409	1.0193	0.1557

The study concludes that each pesticide and their respective oxons significantly depressed normalized cholinesterase activity compared to controls. Regressions of normalized cholinesterase activity over exposure concentration indicated that the oxon forms had steeper declines in AchE activity by concentration than their respective parental forms. Maloxon and chloroxon had steeper negative slopes than diazoxon. For the parent compounds, chlorpyrifos decreased AchE activity more rapidly than did malathion (p=0.0201).

The median 96-hr lethal concentrations for each of pesticides studied along with their respective oxons are reported in Table XX. The median 96-hr LC_{50} value for diazinon and diazoxon are 7.49 and 0.76 mg/l, respectively, based on nominal concentrations.

Table 96-hr median lethal concentrations and 95% confidence intervals for organophosphate insecticides and their respective oxygen analogs (oxons); probit dose response slopes and associated probability levels are also reported

	Slope	P of slope	LC_{50} (mg/l)	95% Confidence Interval (mg/l)
Chlorpyrifos	17.02	0.0339	3.005	0.993 – 157
Diazinon	3.374	NS	7.488	NA
Diazinon oxon	14.08	0.001	0.760	0.336 – 3.212

[1] Ellman, G. I. , K. D. Coutney, F. Andres, and R. M. Featherstone. 1961. A new and rapid colorimetric determination of actrylcholinesterase activity. Biochemistry and Pharmacology 7: 88 – 95.

Malathion	31.48	NS	2.137	NA
Malathion oxon	133.7	0.011	0.023	0.014 – 0.180

Description of Use in Document (QUAL, QUAN, INV): Qualitative

Rationale for Use: Study provides useful information on the relative sensitivity of amphibians to diazonon compared to surrogate fish species. Also, the study provides useful information on the toxicitiy of the diazoxon degradate relative to the parent compound.

Limitations of Study: Study relies on nominal concentrations rather than measured; wild-caught animals are used and prior chemical exposure history is unknown.

Peer Reviewer: Thomas Steeger, Ph.D., Senior Biologist

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ECOTOX Record Number and Citation: 84407 Lower, N. and A. Moore. 2003. Exposure to insecticides inhibits embryo development and emergence in Atlantic salmon (*Salmo salar* L.). Fish Physiology and Biochemistry 28: 431 – 432.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Six groups of 600 unfertilized eggs placed in 500 ml glass containers and mixed with milt from six male salmon and 200 ml solution with 0.05 and 0.1 µg/L of either cypermethrin or diazinon as well as one group with cypermethrin and diazinon combined at 0.05 µg/L was added. After 2 minutes, the eggs were rinsed in clean water and placed in separate artificial redds.

Fewer fry successfully hatched following exposure to 0.05 and 0.10 µg/L cypermethrin and 0.05 µg/L diazinon compared to other treatment groups. Exposure to 0.05 µg/L cypermethrin caused fry to emerge earlier and exposure to 0.05 µg/L diazinon caused dry to emerge later compared to controls. Disruption of the normal pattern of emergence was greater ($p<0.01$) when embryos were exposed to the pesticides separately, rather than in combination.

Description of Use in Document: Qualitative

Rationale for Use: Study not used quantitatively since exposure concentrations are [presumably] based on nominal and the purity of the test compound is not stated

Limitations for Use: The source of the eggs and male fish used for milt is not specified.; purity of the pesticides is not stated. Concentrations presumed to be nominal since there is no discussion on whether concentrations were measured. No raw data are provided; data are plotted on a graph; however, it is not possible to accurately distinguish treatment groups from the graph.

Percent changes in hatch and emergence cannot be determined from the information presented in the paper.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist.

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ECOTOX Record Number and Citation: 53845 Sánchez, M., M. D. Ferrando, E. Sancho and E. Andreu. 1999. Assessment of the toxicity of a pesticide with a two-generation reproduction test using *Daphnia magna*. Comparative Biochemstry and Physiology Part C 124: 247 – 252.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Waterfleas, *Daphnia magna*, obtained from in-house culture. Diazinon (96%) dissolved in acetone. Daphnids (<24 hrs old) exposed during 21 days to 5 diazinon concentrations (0.05, 0.1, 0.5, 0.75 and 1.0 ng/L plus an acetone control (10-4 µl/l). Daphnids housed individually in 60-ml glasss beakers containing 50 ml test solution under static-renewal (24 hr) conditions. Dilution water was dechlorinated tap water. Test animals fed with algae (*N. oculata*). A total of 15 replicates per each treatment. From the first brood (F_1), 15 neonates (<24 hrs old) individually transferred to 60-ml beakers containing clean, untreated water plus solvent control plus negative control and exposed to same concentrations of diazinon as the parents. Afterward, 15 neonates from the third brood (24 hr old) of the parental generation (F_0) from each pesticide exposure concentration individually transferred to 60-ml beakers containing 50 ml toxicant-free solution, plus the controls; the offspring from this third brood were not exposed to diazinon.

Size (body length), fecundity and survival of each generation determined after 21 days of exposure. Longevity, time to the first reproduction, total number of neonates per female, number of broods and brood size, were the criteria used. Neonates were counted daily and then discarded. The intrinsic rate of natural increase (r) was calculated using the following equation: $\Sigma l_x\ m_x\ e\text{-}r_x=l$ where lx is the proportion of individuals surviving to age x,; m_x is the age-specific number of neonates produced per surviving female at age x (fecundity) and x is days.

Report cites a 24-hr LC_{50} value of 0.86 (0.76 – 0.96) µg/l; however, no data are provided to support this conclusion.

Acccording to the study results summarized in Table XX, length, longevity and number of young per females were significantly different than controls in all of the diazinon treatments. Based on information contained in study tables, longevity of parental generation significantly decreased by 20% in the 0.05 ng/l treatment while number of young decreased by 21% compared to the neat control.

Similarly, brood size, number of young per female and number of broods per female also declined significantly in the F1 generation. Survival decreased by 15% while number of young per female and number of broods per females both declined by 36% and 22%, respectively, relative to controls. These data indicate that the chronic NOAEC for diazinon is less than the lowest concentration tested (<0.05 ng/l) following a 21-day exposure for both parental and F1 generations.

No-observed adverse effect concentration in ng/l for parental (F_0), first brood (F_1 first) and third brood (F_1 third). F_0 exposed to diazinon continuously for 21 days.

Generation	Carapace Length	Longevity	Days to 1st brood	Number of young per female	Brood size	Number of broods per female	r
F_0	<0.05	<0.05	0.1	<0.05	0.05	0.05	0.5
F_1 (first)	<0.05	0.5	0.75	0.1	0.1	0.5	0.5
F_1 (third)	<0.05	0.5	0.5	0.5	0.5	0.5	0.1

Description of Use in Document: Qualitative

Rationale for Use.: Study provides useful information on the sensitivity of freshwater invertebrates to diazinon on a chronic exposure basis.

Limitations of Study: presumably the results are reported in terms of active ingredient. Although the study reports that analytical analyses were conducted, the results of those analyses are not presented and the report simply states that mean measured concentrations were >90% of nominal. It is also uncertain whether statistical analyses were conducted relative to the neat control, the solvent control or the pooled controls. Direct comparisons are made between treated groups and the neat (blank) control so presumably controls were not pooled. In the comparisons for various parameters from the third brood of the first generation daphnia, carapace length, number of young per female and brood size were all significantly different for the solvent control versus the negative control. For number of young per female, the acetone control was 37% larger than the negative control and indicates that the solvent may be having an effect. The study is of questionable utility given that the solvent is having a significant effect. Additionally, the study alludes to the fact that diazinon concentrations are measured; however, the level of detection is not stated. The treatment concentrations of as low as 0.05 ng/L are relatively challenging to detect.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist

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ECOTOX Record Number and Citation: 22702. Sánchez, M., M. D. Ferrando, E. Sancho and E. Andreu. 2000. Physiological Perturbations in Several Generations of *Daphnia magna* Straus Exposed to Diazinon. Ecotoxicology and Environmental Safety 46: 87 – 94

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: This study appears to be identical to Sánchez *et al*. 1999 (**53845)**

Description of Use in Document: Qualitative

Rationale for Use.: Study provides useful information on the sensitivity of freshwater invertebrates to diazinon on a chronic exposure basis.

Limitations of Study: presumably the results are reported in terms of active ingredient. Although the study reports that analytical analyses were conducted, the results of those analyses are not presented and the report simply states that mean measured concentrations were >90% of nominal. It is also uncertain whether statistical analyses were conducted relative to the neat control, the solvent control or the pooled controls. Direct comparisons are made between treated groups and the neat (blank) control so presumably controls were not pooled. In the comparisons for various parameters from the third brood of the first generation daphnia, carapace length, number of young per female and brood size were all significantly different for the solvent control versus the negative control. For number of young per female, the acetone control was 37% larger than the negative control and indicates that the solvent may be having an effect. The study is of questionable utility given that the solvent is having a significant effect. Additionally, the study alludes to the fact that diazinon concentrations are measured; however, the level of detection is not stated. The treatment concentrations of as low as 0.05 ng/L are relatively challenging to detect.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist

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ECOTOX Record Number and Citation: 71888 Banks, K. E., S. H. Wood, C. Matthews, K. A. Thuesen. 2003. Joint acute toxicity of diazinon and copper to *Ceriodaphnia dubia*. Environmental Toxicology and Chemistry 22(7): 1562 – 1567.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Diazinon (99.8% ai) prepared in reconstituted hard water. Ceriodaphnia dubia neonates (<24 hr old) obtained from cultures maintained at the University of North Texas (Denton, TX). Cultures maintained in hard water and fed green algae (*Pseudokirchneriella subcapitata*), blended trout chow and Cerophyll® (Ward's Natural Science Establishment, Rochester, NY) and were exposed to a 16:8 light:dark photoperiod. Nominal diazinon test concentrations were 0.05, 0.10, 0.20, 0.40 and 0.80 µg/L.

Toxicity tests are reported to have followed procedures recommented by U.S. EPA. Exposures conducted in 30-ml plastic containers filled with 15 ml of test solution. Four replicates each containing 5 neonates used for each treatment. The test was conducted under static conditions and no food was provided to the organisms during the 48-hr test duration. All tests conducted at 25 + 1^{o}C.

The initial concentration of diazinon in the stock solution determined with ELISA (EnviroGard 96 Well Plate Kit.

Control survival was >90% and water quality remained within the guidelines established by EPA (temperature 25±1^{o}C; DO 8.27±0.06 mg/L; pH 8.35 – 8.36; alkalinity 136±9.5 mg/L. The measured concentration of diazinon was within 90% of nominal at test initiation. The 48-hr LC_{50} value was 0.45 µg/L (95% CI: 0.36 – 0.57 µg/L).

Description of Use in Document: Qualitative

Rationale for Use: Study provides useful information on the sensitivity of freshwater invertebrates to diazinon.

Limitations of Study: study appears to be scientifically sound; however, it relies on nominal concentrations beyond the single measured concentration on the stock solution.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist

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ECOTOX Record Number and Citation: Dutta, H. M. and H. J. M. Meijer. 2003. Sublethal effects of diazinon on the structure of the testis of bluegill, *Lepomis macrochirus*: a microscopic analysis. Environmental Pollution 125: 355 – 360.

Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 2, 2007

Summary of Study Findings: Male adult bluegills were obtained from a fish hatchery near Baltic, OH.; fish were acclimated in the lab for 4 months prior to the study in dechlorinated tap water. Test water quality consisted of 21±1oC, pH 7 ± 0.16; DO 8.27 ± 0.33 mg/L; alkalinity 41.78 ±1.48 mg/L. Fish were fed daily using Tetra Doro Min (Tetra Werke, Germany). Fish were exposed to 60 µg/L for 24, 48, 72 and 96 h and 1 and 2 wk intervals using formulated end-product (25% a,i. 57% aromatic petroleum derivative solvent and 18% inerts. Exposures conducted in 180-l glass tanks under static renewal conditions with water changes every 24 hours. Ten fish were used in a control tank and presumably the same number was in the treatment tank.

After exposure to 24, 48, 72 96 h and 1 and 2 weeks, treated and control fish were euthanized with 100 mg methyltricaine sulfonate/L buffered with 100 mg sodium bicarbonate/L. Average length, body weight and testicular weight recorded. Testes were fixed in Bouin solution for 24 hrs. Diameter measurement (40) were made of seminiferous tubules, the lumen within the tubules and of the spermatogonia and spermatozoa randomly from the control group and the diazinon-treated group at the different exposure periods using an ocular micrometer.

Table 1 summarizes the results of the study. The authors concluded that in the 96 hr group there were significant reductions in both the lumen and seminferous tubule size in comparison with controls and 24, 48 and 72 hr exposures. After 2 weeks of exposure hardly any lumen was seen. The change in the diameter of the seminiferous tubules was very irregular and there was no correlation between the size of the fish, body weight and weight of the testes after different expsoure periods to diazinon. The authors note significant changes in germ cell diameter; however, they do not appear to be consistently correlated with exposure period.

Description of Use in Document: Invalid

Rationale for Use: Potential solvent effect not accounted for**.**

Limitations of Study: the study only tested a single concentration of diazinon. The study measured the response from a formulated product; however, the study cannot distinguish between the effects that may have been due to the organic solvent/inerts co-formulated with the active ingredient.

Table Summary of mean lumen diameter, mean seminiferous tubule lumen diameter, mean germ cell diameter and mean spermatozoa diameter in mm following 24, 48, 72, 96 hr and 1 and 2 week exposures to diazinon formulated endproduct at 60 µg/L.

Treatment	Mean lumen diameter (mm)	Mean seminiferous tubule lumen (mm)	Mean germ cell diameter (mm)	Mean spermatozoa diameter (mm)
Control	0.01878	0.0647	0.0129	0.001994
24 hr	0.0343 b	0.0836 b	0.0134	0.001875
48 hr	0.0142 a	0.058	0.0112 a	0.001769 a
72 hr	0.0485 b	0.0849 b	0.0126	0.001694 b
96 hr	0.0072 b	0.0514 a	0.0104 b	0.00124 b
1 week	0.0218 a	0.0692	0.0095 b	0.001575 b
2 week	0.0081 b	0.0528 b	0.0094 b	0.001638 b

a Significant
b Highly Significant

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist

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ECOTOX Record Number and Citation: Banks, K. E., P. K. Turner, S. H. Wood, and C. Matthews. 2005. Increased toxicity to *Ceriodaphnia dubia* in mixtures of atrazine and diazinon at environmentally realistic concentrations. Ecotoxicology and Environmental Safety 60: 28 – 36.
Purpose of Review (DP Barcode or Litigation): Litigation

Date of Review: March 30, 2007

Summary of Study Findings: Diazinon (99.8% ai) prepared in reconstituted hard water. Ceriodaphnia dubia neonates (<24 hr old) obtained from cultures maintained at the University of North Texas (Denton, TX). Cultures maintained in hard water and fed green algae (*Pseudokirchneriella subcapitata*), blended trout chow and Cerophyll® (Ward's Natural Science Establishment, Rochester, NY) and were exposed to a 16:8 light:dark photoperiod. Nominal diazinon test concentrations were 0.10, 0.20, 0.40, 0.6, 5, 10, 20 and 40 µg/L.

Toxicity tests are reported to have followed procedures recommented by U.S. EPA. Exposures conducted in 30-ml plastic containers filled with 15 ml of test solution. Four replicates each containing 5 neonates used for each treatment. The test was conducted under static conditions and no food was provided to the organisms during the 48-hr test duration. All tests conducted at 25 + 1°C.

The initial concentration of diazinon in the stock solution determined with ELISA (EnviroGard 96 Well Plate Kit.

Control survival was ≥90% and water quality remained within the guidelines established by EPA (temperature 25±1°C; DO 8.27±0.06 mg/L; pH 8.35 – 8.36; alkalinity 136±9.5 mg/L. The measured concentration of diazinon was within 90% of nominal at test initiation. The 48-hr LC_{50} value was 0.21 µg/L (95% CI: 0.17 – 0.25 µg/L).

The study also notes that in combination with atrazine ranging from 5 to 40 µg/L, diazinon 48-hr LC_{50} values were lower (more sensitive) than with diazinon alone.

Table Median lethal concentrations for diazinon alone and in combination with increasing concentrations of atrazine.

	LC_{50} and 95% Confidence Interval (µg/L)
Diazinon alone	0.21 (0.17 – 0.25)
Diazinon + 5 µg/L atrazine	0.16 (0.14 – 0.19)
Diazinon + 10 µg/L atrazine	0.12 (0.11 – 0..15)
Diazinon + 20 µg/L atrazine	0.14 (0.12 – 0.16)
Diazinon + 40 µg/L atrazne	0.13 (0.11 – 0.16)

Description of Use in Document: Quantitative
Rationale for Use: Study is appears to be scientifically sound and provides a more sensitive endpoint on acute diazinon toxicity to freshwater invertebrates than is available through registrant-submitted data.

Limitations of Use: study appears to be scientifically sound; however, it relies on nominal concentrations beyond the single measured concentration on the stock solution. The depression in median lethal concentrations for diazinon when in combination with atrazine does not appear to be concentration dependent.

Primary Reviewer: Thomas Steeger, Ph.D., Senior Biologist

Secondary Reviewer: Kristina Garber, Biologist

ECOTOX Record Number and Citation: 62247. Scholz, N. L., N. K. Truelove, G. L. French, B. A. Berejikian, T. P. Quinn, E. Casillas and T. K. Collier. 2000. Diazinon disrupts antipredator and homing behaviors in chinook salmon (*Oncorhynchus tshawytscha*). Canadian Journal of Fisheries and Aquatic Science 57: 1911 – 1918.

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OFFICE OF
RESEARCH AND DEVELOPMENT
February 22, 2001

MEMORANDUM

SUBJECT: Review of papers on diazinon effects on salmon olfaction

FROM: Dave Mount ORD/NHEERL/MED

TO: Tom Steeger OPPTS/OPP/EFED

At your request, I have reviewed two manuscripts regarding the effects of diazinon on olfaction in salmon. These are:

Scholz, N.L., N.K. Truelove, B.L. French, B.A. Berejikian, T.P. Quinn, E. Casillas, and T.K. Collier. 2000. Diazinon disrupts antipredator and homing behaviors in chinook salmon (*Oncorhynchus tshawytscha*). Can. J. Fish. Aquat. Sci. 57:1911-1918.

Moore, A., and C.P. Waring. 1996. Sublethal effects of the pesticide diazinon on olfactory function in the mature male Atlantic salmon parr. J. Fish. Biol. 48:758-775.

The Moore and Waring paper deals with electrophysiological measurements on the olfactory epithelium of salmon and on olfactory-stimulated hormone production in salmon, both after exposure to waterborne diazinon. In general I found no obvious faults with the experimental procedures. The electrophysiological experiments used repeated measures on the same fish and I didn't see any data in the paper to show that this is not an issue, although the text indicates reference measurements were made to determine the effect of this procedure. The olfactory responses were made relative to a standard exposure to L-serine; I'm not familiar with this procedure so I can't comment on how to interpret the absolute values of the responses. Some of the graphs also don't make clear what the control response was (e.g., Figure 1), leaving unclear what effect the lowest exposures had relative to control.

Details aside, the overall package does seem to suggest that olfactory responses of salmon measured in this way (electrophysiogram of perfused olfactory rosettes) are changed by exposure to increasing concentrations of diazinon. The interpretation of these effects is discussed farther below.

The second portion of the Moore and Waring paper evaluates the stimulation of several hormones in male parr exposed to female salmon urine with or without pre-exposure to diazinon. Again, I have some minor quibbles with the procedures and data presentation. An exposure to industrial methylated spirits (IMS) alone, without urine, would have been useful. Also, the data analysis seems confused (figs 4 and 5); rather than determining whether the response was significantly greater than the negative control (no urine), in seems much more logical to determine whether the response with diazinon exposure was significantly reduced from the positive control. On balance, however, it does not seem unreasonable to conclude that exposure to diazinon at some concentration changes response to priming with female salmon urine when measured in this way.

The Scholz et al. paper also contains experiments of two types: 1) effects of diazinon pre-exposure on responses to an "alarm" stimulus (a water extract of homogenized salmon skin); and 2) return of salmon to the source hatchery after pre-exposure to varying concentrations of diazinon. In the first set of experiments, individual young salmon are exposed to one of several concentrations of waterborne diazinon for 2 hours, then returned to an observation tank where their activity and feeding behavior (on live daphnids) is monitored for 8 minutes, then a standard aliquot of skin extract is introduced, followed by another 8 minutes of observation. The negative control response is for an approximately 80% reduction in activity and about 90% reduction in food strikes following introduction of the skin extract, presumably indicating a natural response to predation occuring in the field. Following on the work of Moore and Waring, if diazinon affects olfaction, then this "alarm response" would be reduced following diazinon exposure.

The data from these experiments indicate that the 2-hour diazinon pre-exposure did not have an effect on activity or feeding behavior <u>prior</u> to introduction of the skin extract. After introduction of the skin extract, activity and feeding behavior was reduced in all treatments and control; however, the magnitude of the response was significantly reduced (or nearly so) in fish pre-exposed to diazinon at 1 ug/L or 10 ug/L. It should be noted that this "alarm" response was not eliminated, only reduced. For example, in control fish, the post-extract activity was reduced by about 82% from pre-extract activity, while after 10 ug/L pre-exposure, post-extract activity was reduced by about 68%.

The homing study evaluated the effect of diazinon on the ability of fish that had already returned to their natal hatchery to return after being transplanted from the hatchery back to a downstream (2 km) location. At total of 40 fish in each of four treatment groups (control and 0.1, 1.0, and 10 ug/L diazinon pre-exposure) were released downstream; of these, a total of 16, 12, 12, and 6 fish, respectively, returned to the hatchery and were recaptured. The statistical tests applied by the authors find that the return of 6 fish in the highest diazinon treatment was significantly different from the solvent control. The design of this experiment causes some discomfort; one could argue that treating the individual fish as the sampling unit is a form of pseudoreplication. Furthermore, the fish were actually released in a series of small groups, but the details are vague and the results are only given in "lump" form. It seems possible that the individual release dates could be used as an experimental unit instead of the individual fish, but this was not done for some reason. The design in general is not very robust; it would be strengthened greatly if the entire experiment would be repeated. The authors also note that the return rate for the control fish was inexplicably lower than has been observed for similar releases in previous years, although the impact of that on the findings is not immediately obvious. Overall, it seems more likely than not that there may be some effect here, but this is by far the weakest of the experiments in terms of experimental design and interpretation. This is unfortunate, since it is the study that most closely links to assessment endpoints likely to be of concern for ecological risk assessments for this species.

In summary then, all of these experiments (with the possible exception of the last) seem to demonstrate a statistically significant change in physiology or behavior that can be at least theoretically tied to effects of diazinon on olfaction in salmon. The primary issue is how to interpret this information in the context of ecological risk assessment, which is the focus of the remaining discussion. For expediency, I'll refer to the four sets of experiments as the "epithelial", "priming", "alarm", and "homing" studies (in the order described above).

I presume that Agency risk assessments to which these data might be applied would have as their assessment endpoint something like, "protection of balanced, indigenous aquatic communities," or perhaps, "maintenance of naturally reproducing salmon populations." The basic difficulty in interpreting these studies in the context of ecological risk is that the measurements that are made (particularly in the epithelial, priming, and alarm studies) are not clearly tied to these assessment endpoints. One can easily develop scenarios where it is plausible that these measures might affect salmon at the population level, but it is also possible that these changes might be compensated for in other ways that would result in no effect on the population. There is no quantitative link established between these responses and changes in a field population. The Agency's *Framework for Ecological Risk Assessment* (1992) identifies this problem:

> In many cases, measurement endpoints at lower levels of biological organization may be more sensitive than those at higher levels. However, because of compensatory mechanisms and other factors, a change in a measurement endpoint at a lower organizational level (e.g., a biochemical alteration) may not necessarily be reflected in changes at a higher level (e.g., population effects). (p. 14)

And later on:

> Ideally, the stressor-response evaluation quantifies the relationship between the stressor and the assessment endpoint. When the assessment endpoint can be measured, this analysis is straightforward. When it cannot be measured, the relationship between the stressor and measurement endpoint is established first, then additional extrapolations, analyses, and assumptions are used to predict or infer changes in the assessment endpoint. (p. 23)
>
> Measurement endpoints are related to assessment endpoints using the logical structure presented in the conceptual model. In some cases, quantitative methods and models are available, but often the relationship can be described only qualitatively. Because of the lack of standard methods for many of these analyses, professional judgement is an essential component of the evaluation. It is important to clearly explain the rationale for any analyses and assumptions. (p. 23)

Ambient Water Quality Criteria (AWQC) to protect aquatic life represent one of relatively few attempts to standardize the use of toxicity data in risk assessments. The guidelines for deriving these criteria (Stephan et al., 1985) focus on toxicity test endpoints that have direct applicability to population demographics – basically, survival, growth, and reproduction. Other effects are not considered unless there is strong evidence of a direct link between the measured endpoint and survival, growth, or reproduction. In general, data such as those generated by the epithelial, priming, and alarm studies would not be considered directly in the criteria derivation.

Existing criteria documents contain many types of data that were not used in the criteria derivation (the documents collate and review these data, but they are not used to actually define the criterion concentration). For example, behavioral studies with copper and other chemicals have shown avoidance behavior in the laboratory at very low concentrations (e.g., rainbow trout will avoid 1 ug Cu/L). While one could imagine this affecting populations in the field, it is also reasonable to expect that many top notch trout fisheries have ambient copper concentrations of at least 1 ug/L. Presumably, other compensatory factors keep the behavioral response measured under laboratory conditions from resulting in noticeable population-level impacts.

Histological or biochemical changes are often reported for many chemicals at concentrations below that shown to directly affect survival, growth, or reproduction in laboratory toxicity tests. These might be more similar to the epithelial studies conducted by Moore and Waring. The recent revision of the ammonia criteria document (accessible through the OW/OST website) has the following to say about the use of histological endpoints:

> Endpoint indices of abnormalities such as reduced growth, impaired reproduction, reduced survival, and gross anatomical deformities are clinical expressions of altered structure and function that originate at the cellular level. Any lesion observed in the test organism is cause for concern and such lesions often provide useful insight into the potential adverse clinical and subclinical effects of such toxicants as ammonia. For purposes of protecting human health or welfare these subclinical manifestations often serve useful in establishing 'safe' exposure conditions for certain sensitive individuals within a population.
>
> With fish and other aquatic organisms the significance of the adverse effect can be used in the derivation of criteria only after demonstration of adverse effects at the population level, such as reduced survival, growth, or reproduction. Many of the data indicate that the concentrations of ammonia that have adverse effects on cells and tissues do not correspondingly cause adverse effects on survival, growth, or reproduction. No data are available that quantitatively and systematically link the effects that ammonia is reported to have on fish tissues with effects at the population level. This is not to say that the investigators who reported both tissue effects and population effects within the same research did not correlate the observed tissue lesions and cellular changes with effects on survival, growth, or reproduction, and ammonia concentrations. Many did, but they did not attempt to relate their observations to ammonia concentrations that would be safe for populations of fish under field conditions nor did they attempt to quantify (e.g.,

> increase in respiratory diffusion distance associated with gill hyperplasia) the tissue damage and cellular changes (Lloyd 1980; Malins 1982). Additionally, for the purpose of deriving ambient water quality criteria, ammonia-induced lesions and cellular changes must be quantified and positively correlated with increasing exposures to ammonia.
>
> In summary, the following have been reported:
> 1. Fish recover from some histopathological effects when placed in water that does not contain added ammonia.
> 2. Some histopathological effects are temporary during continuous exposure of fish to ammonia.
> 3. Some histopathological effects have occurred at concentrations of ammonia that did not adversely affect survival, growth, or reproduction during the same exposures.
>
> Because of the lack of a clear connection between histopathological effects and effects on populations, histopathological endpoints are not used in the derivation of the new criterion, but the possibility of a connection should be the subject of further research.

In human health risk assessment, deviations from normal physiology are generally considered to be adverse effects. As described in the text from the ammonia document, the practice in AWQC and in other ecological risk assessments in general, is to focus on effects that cause changes at the population level; this requires the ability to make this link in a manner quantitative enough to say how strong a response in the measured parameter would adversely effect populations.

The combined evidence from the Moore and Waring and Scholz et al. studies do not clearly provide this connection. The electrophysiograph data from the epithelial studies provide strong evidence that diazinon exposure can induce measurable changes in activity of the epithelial rosettes, but there are no means to connect this directly to changes in survival, growth, or reproduction. As shown in Figures 1 and 2 of Moore and Waring, diazinon exposure produces a concentration-dependent decrease in rosette responsiveness, but responsiveness is not lost, just reduced. Thus, the question becomes, “What is the minimum level of rosette activity necessary?”

The priming studies performed by Moore and Waring provide a closer link to reproductive success; these studies link diazinon exposure to changes in reproductive hormone response to priming with female salmon urine. However, data for the endpoint most directly related to reproduction, milt production, were equivocal. The data (figure 6) show a significant increase in milt production in fish primed with urine or urine plus carrier solvent relate to unstimulated fish. However, the more relevant question would be whether diazinon treatment decreases milt production relative to the solvent control; this comparison isn’t made, but it does not appear likely that is did, based on the figure. Further, even if one concludes that there is an effect in milt release under these conditions, it isn’t clear whether this would actually affect reproductive success under field conditions.

The alarm response studies show a decrease in the so-called “alarm response” following pre-exposure to diazinon, and the nature of this response is consistent with what might be expected based on the olfactory effects shown by Moore and Waring. While a significant change was found, a substantial alarm response was still present in diazinon-exposed fish. Whether the degree of change noted is sufficient to affect survival/growth/reproduction in the field is uncertain.

The homing studies provide data that are closest to making the link to effects on populations. Clearly, relatively little supposition or extrapolation is necessary to infer that reduced migratory capability could have adverse effects on salmon populations. There is still some question about “how much is too much”, but not substantially more so than is faced in interpreting ordinary survival or growth data. Unfortunately, this study is compromised somewhat by a weak design and lack of replication. Having further data on this response using a more robust design (e.g., releasing several lots of fish over the course of several days) would be helpful.

Judging the significance of any of these findings in producing ecological risk is also dependent on determining the relationship between actual exposures that are observed in the field. Although the authors claim that they occur, pulses of diazinon to 10 ug/L are not something that occurs very often to my knowledge – this seems extreme.

Also relevant is how to interpret the likely effects of field exposures on the aquatic community in general. In a construct like AWQC, the much greater sensitivity of other organisms, such as cladocerans (toxic effects in the 0.1 ug/L range), to diazinon cause “acceptable risk” to be exceeded at diazinon concentrations below those showing significant effects on salmon olfaction. This approach doesn’t get at how to deal quantitatively with the olfaction data, it just makes it moot for diazinon. If the assessment endpoint is populations of salmon *per se*, rather than protection of aquatic communities, then the problem doesn’t go away, unless one considers cladocerans and other organisms highly sensitive to diazinon as part of the habitat essential to maintain salmon populations (after all, it takes more than just water to maintain salmon).

One of the questions you posed was in regard to a desire from the Services to include the alarm response assay as a standard screening test. Two things would generally be required: 1) that the test is shown to be sufficiently reproducible within and between laboratories; and 2) that the endpoint of the assay be more sufficiently tied to the assessment endpoint (presumably maintenance of salmon populations or aquatic communities). If one were to attempt the latter, it would seem that combining the olfaction assays with the homing studies for multiple chemicals in multiple trials would be a good first step, though I don’t know how reliable it is to assume that something that blocks the alarm response would necessarily interfere with homing (or the reverse). If no more attempt is made to relate the olfaction assays with populations response, it will be very difficult to move the olfaction issue into a part of the risk calculation rather than being simply a component of the qualitative uncertainty.

I’ve spent most of this discussion describing things that discourage the use of these data in quantitatively describing risk. I should counter this by saying that the difficulty of incorporating this information into a risk assessment should not be taken to suggest that adverse effects of diazinon on salmon populations are not possible via this mechanism (provided exposures were sufficiently high). Certainly the cluster of studies looking at the issue show a fair amount of internal consistency with regard to the existence of such an effect at concentrations below those that reduce survival or growth in salmon or other fish species. This particular case is even more troubling because it is unlikely that any traditional toxicity test could effectively measure effects on salmon reproduction directly, and, in the case of salmon, successful reproduction in the field is thought/known to be dependent on olfaction in ways that wouldn't be assessed using traditional chronic toxicity tests on this or other fish species. Describing this uncertainty qualitatively within a risk assessment would definitely be appropriate, even if olfaction data are not part of the quantitative risk calculation. The risk manager will be faced with the decision as to how this uncertainty affects management decisions; at this point, I'm not sure that our scientific understanding can do more than frame the question.

Stephan CE, Mount DI, Hansen DJ, Gentile JH, Chapman GA, Brungs WA. 1985. Guidelines for deriving numerical national water quality criteria for the protection of aquatic organisms and their uses. U.S. EPA, Environmental Research Laboratory, Duluth, MN. NTIS No. PB85-227049. 98 pp.

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Rodgers, M. H. 2005b. Diazoxon (a metabolite of the active ingredient diazinon) Dietary Toxicity (LD_{50}) to the Bobwhite Quail. Huntingdon Life Sciences Limited, Woolley Rd, Alconbury, Huntingdon, Cambridgeshire, England (Huntingdon Project ID: MAK 872). Sponsored by Makhteshim-Agan of North America Inc., 4515 Falls of Neuse Rd., Suite 300, Raleigh, NC 27609 (Makhteshim Project Number: R-18131). Study initiated: 04/05/05; study completed: 05/25/05 (**MRID 465796-02**)

The acute dietary toxicity of diazoxon, a metabolite of th active ingredient diazinon, to approximately 12-d old Bobwhite quail (*Colinus virginianus*) was assessed over 8 days (5 days of exposure plus 3-day post-exposure observation period). Diazoxon was administered to the birds in the diet at 30, 60, 120, 240, 480 and 960 mg a.i/kg diet of diet. The 5 day acute dietary LC_{50} was 72.3 mg a.i/kg of diet. The 5-day NOAEC of diazoxon based on reduced body weight was 9.4 mg a.i/kg diet of diet (based on a preliminary study). According to the US EPA classification, diazoxon would be classified as highly toxic to Bobwhite quail on a subacute dietary exposure basis.

Clinical signs were confined to unsteadiness/inability to stand and subdued behavior in the groups treated with at 60, 120, 240, 480 and 960 mg/kg diet. All birds in the groups treated at 60, 120, 240, 480 and 960 mg/kg diet displayed clinical and/or were found dead. Mortality was observed at 60 (20%), 120 (100%), 240 (100%), 480 (100%) and 960 (100%) mg/kg diet.

This toxicity study is classified as scientifically sound and is thus acceptable and does satisfy the guideline requirement for subacute dietary toxicity study for Bobwhite quail.

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Rodgers, M. H. 2005a. Diazoxon (a metabolite of the active ingredient diazinon) Acute Oral Toxicity (LD_{50}) to the Bobwhite Quail. Huntingdon Life Sciences Limited, Woolley Rd, Alconbury, Huntingdon, Cambridgeshire, England (Huntingdon Project ID: MAK 874). Sponsored by Makhteshim-Agan of North America Inc., 4515 Falls of Neuse Rd., Suite 300, Raleigh, NC 27609 (Makhteshim Project Number: R-18127) (**MRID 465796-04**).

The acute oral toxicity of diazoxon (a metabolite of the active ingredient diazinon) to 27-wk old Bobwhite quail *(Colinus virginianus*) was assessed over 14 days. Diazoxon was administered to the birds by oral intubation (gavage) at 0.79, 1.31, 2.18,.3.61 and 6.00 mg a.i./kg bw. The 14-day acute oral LD_{50} was 4.94.mg a.i/kg bw. The 14-day NOEL of diazoxon to the Bobwhite quail, based on mortality and behavioral effects was 2.18 mg a.i/kg bw. According to the US EPA classification, diazoxon would be classified as very highly toxic to Bobwhite quail on an acute oral exposure basis.

No clinical signs observed in groups dosed at 0.75, 1.30, 2.25 mg/kg bw or the control group. Clinical signs observed in the groups dosed at 3.63 and 6.16 mg a.i./kg bw were confined to subdued behavior, unsteadiness and frothy fluid around the beak on the day of dosing. No other clinical signs were observed through the remainder of the observation period.

This toxicity study is classified as scientifically sound and is acceptable; the study is consistent with guideline requirements for an acute oral toxicity study using Bobwhite quail.

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Grade, R. 1993a. Report on the acute toxicity of G27550 (Oxypyrimidine) to rainbow trout (*Oncorhynchus mykiss*). Ciba-Giegy Ltd., Product Safety, Ecotoxicology, CH-4002 Basel, Switzerland. Project Number 932504. Sponsor: Makhteshim Chemical Works, Ltd., 551 Fifth Ave. Suite 1100, New York, New York 100176. (**MRID 463643-12**).

In a 96-h acute toxicity study, rainbow trout (*Oncorhynchus mykiss*) were exposed to technical grade G 27550 (Oxypyrimidine) at measured concentrations of 0, 9.8, 18.1, 32.3, 60.8 and 101.1mg a.i./L under static conditions. The 96-h LC_{50} was greater than the highest concentration (101.1 mg a.i/L) tested. The NOEC value, based on sub-lethal effects, was 60.8 mg a.i/L. Sublethal effects (swimming behavior, loss of equilibrium, respiratory effects) were observed in the groups exposed to 101.1 mg a.i./L of G27550. Based on the results of this study, G 27550 would be classified as practically nontoxic to rainbow trout in accordance with the classification system of the U.S. EPA.

This toxicity study is scientifically sound; however, because the study was conducted under static conditions and failed to characterize water quality parameters adequately and exceeded recommended ranges for both pH and water hardness, the study is classified as supplemental.

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Grade, R. 1993b. Report on the acute toxicity of G27550 (Oxypyrimidine) on *Daphnia magna*. Ciba-Giegy Ltd., Product Safety, Ecotoxicology, CH-4002 Basel, Switzerland. Project Number 932505. Sponsor: Makhteshim Chemical Works, Ltd., 551 Fifth Ave. Suite 1100, New York, New York 100176. (**MRID 463643-13**).

The 48-hr-acute toxicity of the diazinon degradate oxypyrimidine to *Daphnia magna* was studied under static conditions. Daphnids were exposed to control and test chemical measured at 10.2, 18.4, 32.7, 59.3 and 101.6 mg a.i/L for 48 hr. Mortality and sublethal effects were observed daily. The 48- hour LC_{50} was greater than 101.6 mg a.i/L. The 48-hr NOEC based on mortality was 101.6 mg a.i/L. No sublethal effects were observed during the study period.

Based on the results of this study, oxypyrimidine would be classified as practically nontoxic to the freshwater invertebrate *Daphnia magna* in accordance with the classification system of the U. S. EPA.

This study is classified as supplemental and can be upgraded to core if the registrant can demonstrate that neither water hardness and/or pH affect the toxicity and solubility of oxypyrimidine. Additionally, the registrant should provide more information on the quality of water used in the study.

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Grade, R. 1993c. Report on the growth inhibition of G27550 (Oxypyrimidine) to Green Algae (*Scenedesmus suspicatus*). Ciba-Giegy Ltd., Product Safety, Ecotoxicology, CH-4002 Basel, Switzerland. Project Number 932507. Sponsor: Makhteshim Chemical Works, Ltd., 551 Fifth Ave. Suite 1100, New York, New York 100176. (**MRID 463643-14**).

In a 72 hour acute toxicity study, the cultures of green algae (*Scenedesmus subspicatus*) were exposed to oxypyrimidine at measured concentrations of 1.1, 3.8, 11.6, 35.2 and 109.1 mg a.i/L under static conditions. The NOAEC or EC_{05} and EC_{50}/IC_{50} values based on cell density were 109.1 mg a.i./L and >109.1 mg a.i./L, respectively. No phytotoxic effects were reported in the study; therefore, there were no compound related phytotoxic effects.

This toxicity study is classified as scientifically sound; however, because of the lack of information regarding the study water, this study is classified as supplemental.

Appendix B. Supporting Information for PRZM Scenario Development.

INTRODUCTION

EFED initiated an effort to develop a suite of new PRZM/EXAMS scenarios useful for all six chemicals in the Barton Springs endangered species lawsuit including atrazine, simazine, prometon, metolachlor, diazinon, and carbaryl. EFED initiated an evaluation of the potential use sites relevant to all six chemicals for development as possible modeling scenarios. The evaluation consisted of an investigation of geology, hydrogeology, land cover data, use information, soils information, and conversations with local experts knowledgeable in all of the above.

Initial investigation indicated that the geology and hydrogeology are the defining issues surrounding how the action area for each chemical would be defined. As noted in the atrazine assessment, the action area for the development of the Barton Springs Scenarios was comprised of three hydrologic zones (in order of importance) of the Barton Springs Segment of the Edwards Aquifer: 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 was considered in this assessment, primary emphasis was given to the recharge zone with secondary emphasis on the contributing zone.

Investigation indicated that areas to the east of the Recharge Zone might not be relevant to the assessment (groundwater flow to the Barton Spring 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). For example, agricultural uses lying east of the Recharge Zone (roughly defined by the Interstate 35 corridor) can be considered outside the area of interest and no scenario need be developed for this use. However, if any of the uses are present west of this area within either Recharge or Contributing Zones, then these scenarios should be developed as described below.

Given these facts it was quickly decided that any new scenarios developed needed to be based on the extent of the potential action area for each chemical. In general, this action area consists of three zones identified above including the Contributing Zone, the Recharge Zone, and the Transition Zone. Primary emphasis for scenario development was placed on use sites (both agricultural and non-agricultural) within the Contributing and Recharge Zones. 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/).

These new scenarios were developed under contract with specific guidelines on how to evaluate the need for a scenario and how to parameterize the scenarios that were developed. The process involved numerous interactions between the contractor and EFED and ultimately all decisions on which scenarios to develop were the responsibility of EFED. If the contractor determined that a particular use site is likely to be outside the area of interest and not likely to contribute to the exposures in Barton Springs a written description of the steps taken to determine this and rational for the exclusion was documented and is discussed in the sections that follow.

The following sections discuss the various data sources used in this assessment and ultimately provide a rational for the development of each scenario. Note that not all scenarios were used in each assessment but were selected based on specific analysis of each chemical labeled uses and an understanding of which uses are actually present in the action area for each chemical. In the case of atrazine, the scenarios ultimately used in the assessment were one agricultural site (fallow/idle land using the meadow scenario) and three non-agricultural uses including residential, turf and rights-of-way.

SOURCES OF DATA

Land use data

The contractor obtained two land use coverage's from the city of Austin (COA) and the Texas Commission on Environmental Quality (TCEQ). The land use data were important for quantifying the extent of a particular land use and for identifying representative, yet vulnerable soils. The data set from Austin includes land use by tax parcels and was particularly important for the turf (golf courses) and right-of-way scenarios. The TCEQ dataset developed by the USGS (2003) provided agricultural land cover data, including areas representative of meadows and rangelands, and residential areas. Based on a review of the data, residential areas appeared better classified in the USGS (2003) data set; the COA data set tended to include all lots zoned for residential and often included areas well outside of where pesticides would presumably be applied. Abstracts from the metadata of the two land cover data sets are included below.

<u>COA land use data set:</u> "From October 2003 until December 2004, the City of Austin Watershed Protection and Development Review Department (WPDR) and the Transportation Planning and Sustainability Department (TPSD) produced this land use and tax parcel inventory. The extent of the data includes the watersheds of Travis, Hays, Williamson, and Blanco County that drain into Austin city limits. This includes the City of Austin extra-territorial jurisdiction. The layer is used in watershed, land use, and transportation modeling. More specifically, the information will be used to estimate and forecast impervious cover, population and housing density, and land use change. Parcels were created to reflect 2003 tax maps by either updating year 2000 parcel polygons, or converting and attributing lot lines from the City base map or county appraisal district CAD files. After completing parcel polygons, appraisal district land use data was joined to the layer using the parcel identification number. In addition, historical land use data was joined through GIS overlays. We then coded land use by comparing appraisal district data to the historical data where possible. The land use coding system used in year 2000 data was expanded to reflect the needs of both the planning and watershed management disciplines and the availability of new data. Infrared and color aerial photos were used to confirm or make determinations, especially where data was unavailable or questionable. Other GIS layers such as buildings and parks were used in this verification process." (COA 2003)

<u>USGS (TCEQ) land use data set:</u> "This layer delineates the land use/land cover (LULC) polygons for the Edwards Aquifer Project in Texas from the years 1995 and 1996. Attribution of the polygons is based on a modified Anderson classification schema. LULC classification was done to Level 3 of the classification schema and a new category of Mixed Forest/Shrub was added to better represent the land cover of the area. Fieldwork was performed prior to compilation to gather local data and relate aerial photo images to corresponding ground

features. Because of the stunted or lower tree growth common in this region it was difficult at times to differentiate between Forest, Mixed Forest/Shrub, and Shrub. It should be noted that much of the Planted/cultivated land is highly managed pastureland. A detailed description of the schema can be found in the Supplemental Information Section. All the LULC data was collected from color infrared DOQQs and high-resolution (1:40,000-scale) aerial photography. The minimum mapping unit used for delineating a polygon is 5 acres and the minimum polygon width is 125 feet." (USGS 2003)

Soils data

Data for Hays and Travis counties were downloaded from Soil Data Mart (USDA 2006) and clipped to the hydrozones of the BSS AOI (ftp://www.bseacd.org/from/HCP Shape Files/). EFED indicated that scenarios should be parameterized based on representative soils that will yield high-end runoff and sediment values. Specifically, this focused on Hydrological Group C and D soils with high erodibility and slope. Quantitative descriptions of the soil selection process are provided in the metadata for each scenario with additional detail provided in later sections of this report.

Official soil series descriptions (OSD) of the selected soils were used to characterize the soils of interest for the scenarios (Soil Survey Staff 2006a, b). Soil parameters were obtained from USDA Soil Data Mart (USDA 2006).

Additional Data Sources

When exploring the extent of agricultural areas in the AOI, areas of crops grown in Hays and Travis counties were obtained from NASS (USDA 1997, 2002). This was used as a preliminary attempt to understand the types of crops grown in the AOI and their respective magnitudes.

City and County officials and extension agents were contacted to understand and verify correct parameters to represent each of the scenarios that were developed.

In cases where similar PRZM scenarios were available, parameters were reviewed for consistency. Specifically, the BS turf scenario was compared to the PA turf and FL turf scenarios.

For determination of USLEC and Manning's N values, the RUSLE EPA Pesticide project (2000) was used. Existing files were considered according to current USEPA guidance (USEPA 1998). The Barton Springs area is located in Land Resource Region (LRR) I. The San Antonio climate station is located within this LRR and is an appropriate location for which to select appropriate RUSLE data files. Available crops for this climate station include: 1) Range, 2) Pasture, warm season, 3) peanut, Spanish, 4) Sorghum, grain, and 5) Wheat, winter. For scenarios where appropriate files did not exist (i.e. impervious surfaces), appropriate values were selected to represent USLEC and Manning's N values. Curve numbers were derived based on USDA TR-55: Urban Hydrology for Small Watersheds document (USDA 1986) or from the GLEAMS (USDA 2000) manual when appropriate. Further details are provided in the metadata for each scenario.

CONCEPTUAL MODELS OF DEVELOPED SCENARIOS

Residential

This scenario intended to be used as a surrogate for all urban/suburban home and residential uses in the Barton Springs Segment (BSS) of the Edwards Aquifer. The intention is to couple the edge of field concentrations from this scenario with the edge of field concentrations from the impervious surface scenario for Barton Springs to generate weighted concentrations for areas of varying impervious cover. Crop parameters have been chosen to reflect residential turf areas, primarily lawns, within the BSS.

For this scenario estimates of typical impervious fractions in suburban watersheds were obtained from a City of Austin COA (2002) report for the COA jurisdictional section of the Barton Springs Segment (BSS) and from local runoff studies obtained from the COA. Within the city of Austin Jurisdiction of the Barton Springs Zone approximately 7.5% or 5098 acres consists of impervious surfaces. Within the recharge zone, the city of Austin restricts impervious cover for new development to 15% of the net site area and 20% of the site area in the Barton Creek contributing zone (COA, 2002). However, based on unpublished data obtained from the City of Austin some residential watersheds in the area may be as high as 40% (Rich Robinson, COA, personal communication).

The analysis of land cover information is provided in Figure 1. A conceptual model of this approach is provided in the assessment

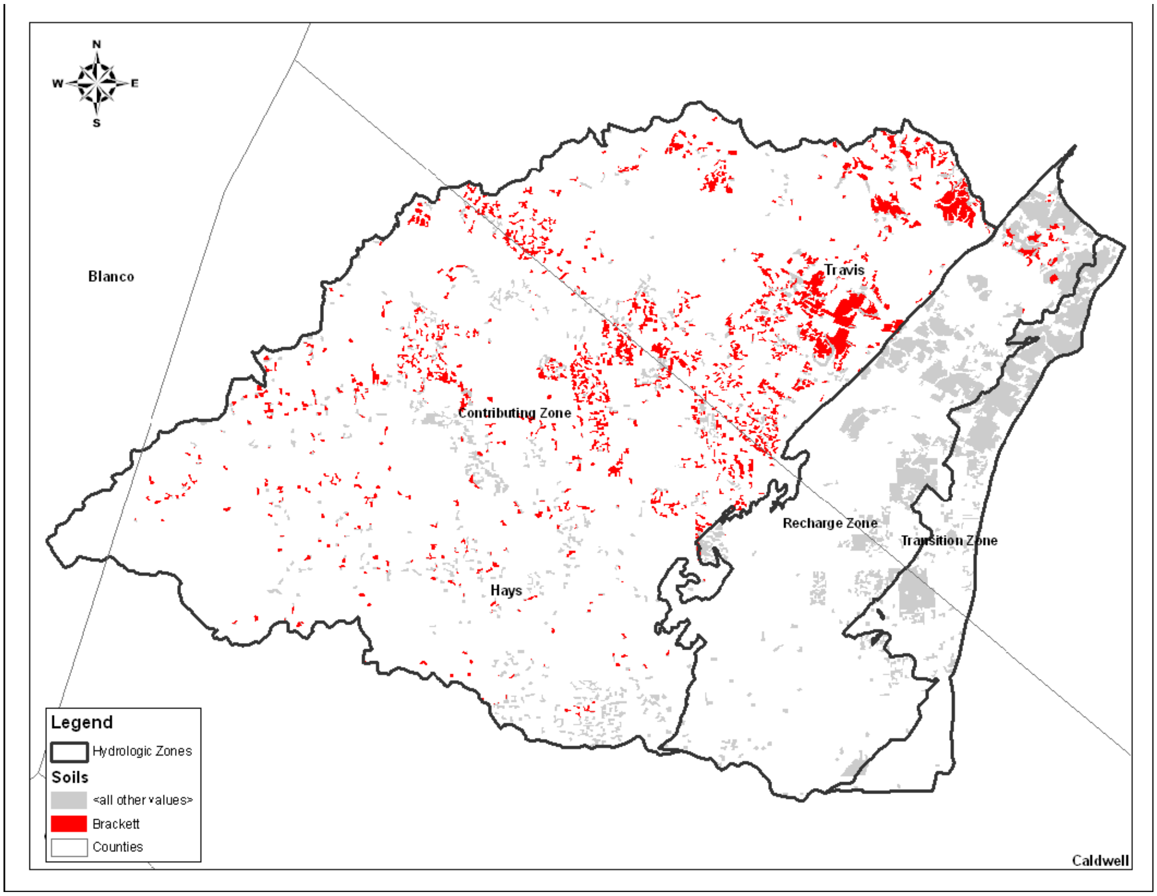

Figure 1. Location of Brackett Soils in single- and multi-family residential areas of the Barton Springs Segment of the Edwards Aquifer, Hays and Travis Counties, Texas.

Impervious

This scenario is intended to be used to mimic hydrology of untreated portions of the Barton Springs Segment (BSS) of the Edwards Aquifer. The intention is to couple the edge of field concentrations from this scenario with the edge of field concentrations from the residential scenario for Barton Springs to generate weighted concentrations for areas of varying impervious cover. Therefore, this scenario relies on a similar soil series as the residential scenario; however the upper horizon has been adjusted to a non-soil nature. As noted above, data indicate that impervious fractions of residential areas in the BSS range from less than 10% (COA 2002) to as high as approximately 40% (Rich Robinson, COA, personal communication). The analysis of land cover information is provided in Figure 2.

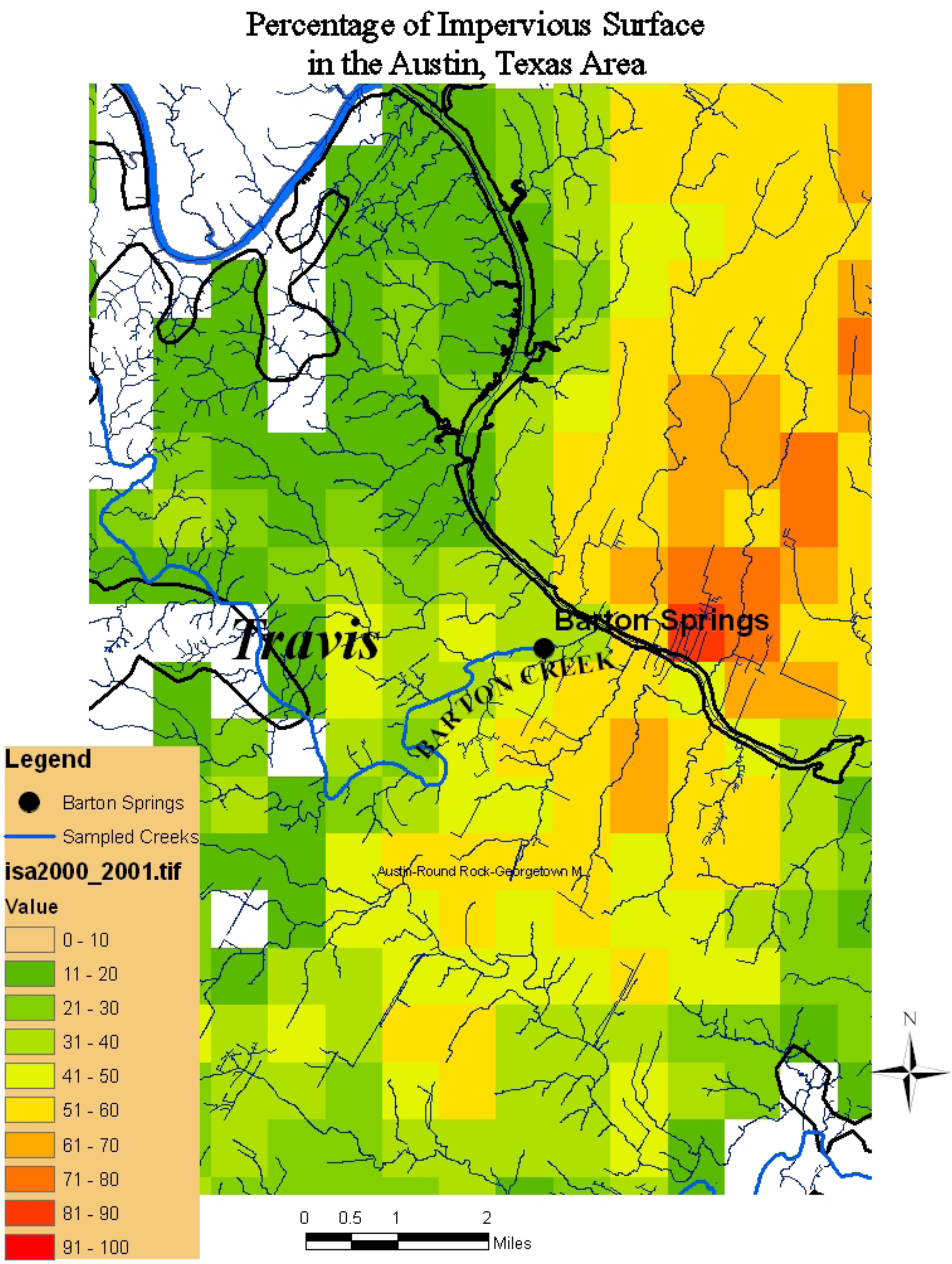

Figure 2. Percentage of Impervious Surfaces near Barton Springs.

Turf

This scenario is intended to represent turf areas (golf courses, parks, sod farms, and recreational fields) in the Barton Springs Segment (BSS) of the Edwards Aquifer. Because golf courses are expected to be the most likely turf areas where pesticides may be applied, much of this scenario has been parameterized to be reflective of golf course turf. NASS data for 1997 and 2002 (USDA 1997, 2002) contained no record of sod harvest in either Hays or Travis counties. Since there are several golf courses located within the BSS (COA 2003), this scenario was parameterized to represent turf on golf courses and may be generally representative of other potential turf areas. Crop parameters are based primarily on bermudagrass (*Cynodon* spp.) since it is a primary turf grass for golf courses and athletic fields. The analysis of land cover information is provided in Figure 3.

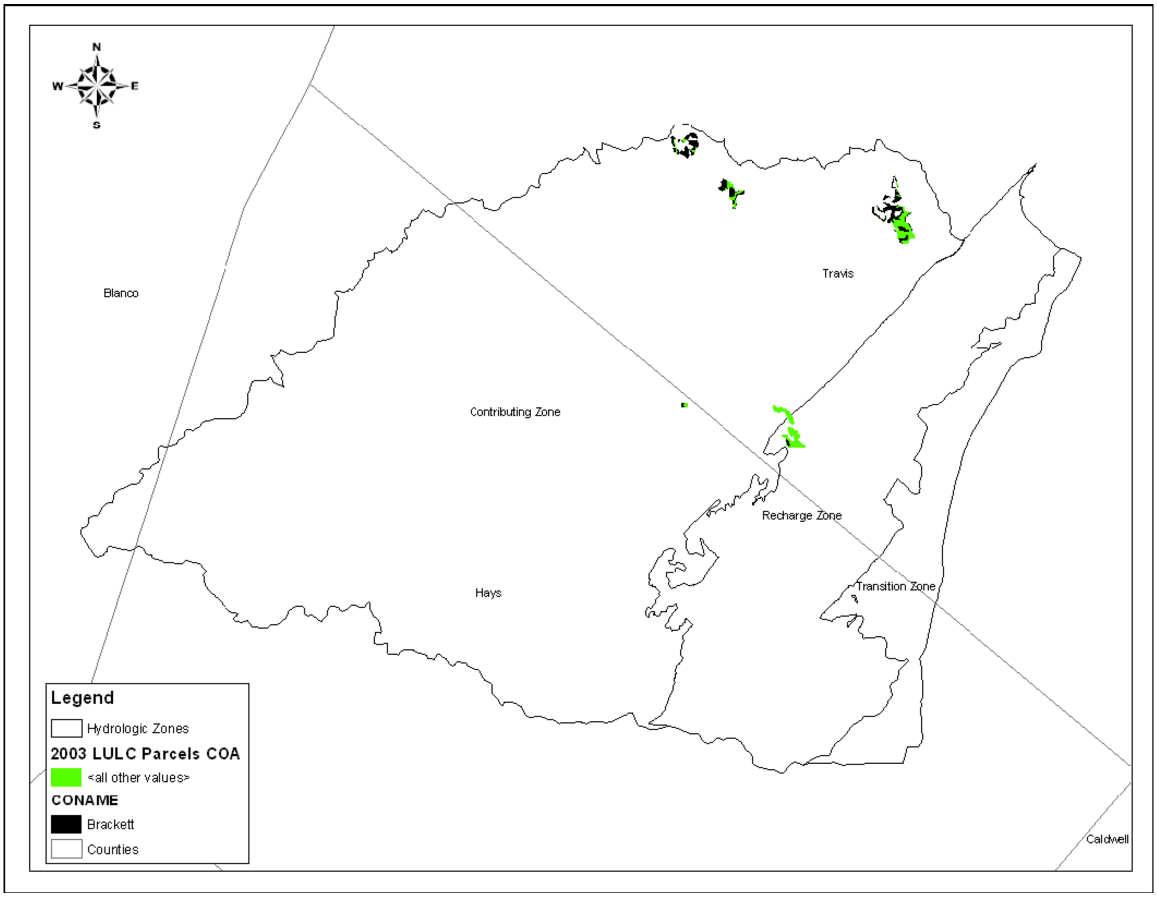

Figure 3. Location of Brackett Soils in golf course areas of the Barton Springs Segment of the Edwards Aquifer, Hays and Travis Counties, Texas.

Right-of-Way

This scenario is intended to represent right-of-way areas including roads, fence lines, power lines, and railroads in the Barton Springs Segment (BSS) of the Edwards Aquifer. Unlike most of EFED existing scenarios, the scenario is conceptually different in that it represents a linear surface that drains into an adjacent water body (drainage ditch). However, for this exercise, EFED assumes that while conceptually different, the scenario is for practicality purposes developed in a similar manner as a standard scenario that assumes a 10-hectare field draining into a 1-hectare static pond.

Crop cover parameters for this scenario were based on typical plants found adjacent to state maintained highway right-of ways. State-maintained highways include farm-to-market (FM) roads, state highways, interstates, and US highways. Bermuda grass is typically found in right-of-way areas in urban areas, while rural areas are dominated by native species such as little bluestem, side-oats grama, and hairy grama (John Mason, Vegetation Management Specialist, Texas DOT, Maintenance Div., personal communication).

The contractor attempted to determine where pesticides may or may not be applied to Right-Of-Ways (including highway/railroad/utility segments). COA was not aware of a source for this information (Nancy McClintock, personal communication). According to Texas Department of Transportation (TX DOT), Vegetation Manager Dennis Markwardt, the TX DOT applies herbicides only (no insecticides) to all of its state roadways. They only apply herbicide to a one-foot wide area along the roadway, not the entire right-of-way. They also limit the use of herbicides within the BSZ to mainly Round-Up, and to a more limited extent, Oust, OutRider and Escort. Occasionally they will need to apply spot treatment to noxious weeds.

According to Travis County Transportation and Natural Resources, Road and Bridge Division Maintenance Manager, Don Ward, Travis County applies herbicide only to their rural roads where there is no curbing gutter. They apply only Round-Up and apply it to a four foot wide area along the roadway approximately two times per year. Scott Lambert provided us with a GIS layer of the Travis County roads where herbicide may be applied. The analysis of land cover information is provided in Figure 4.

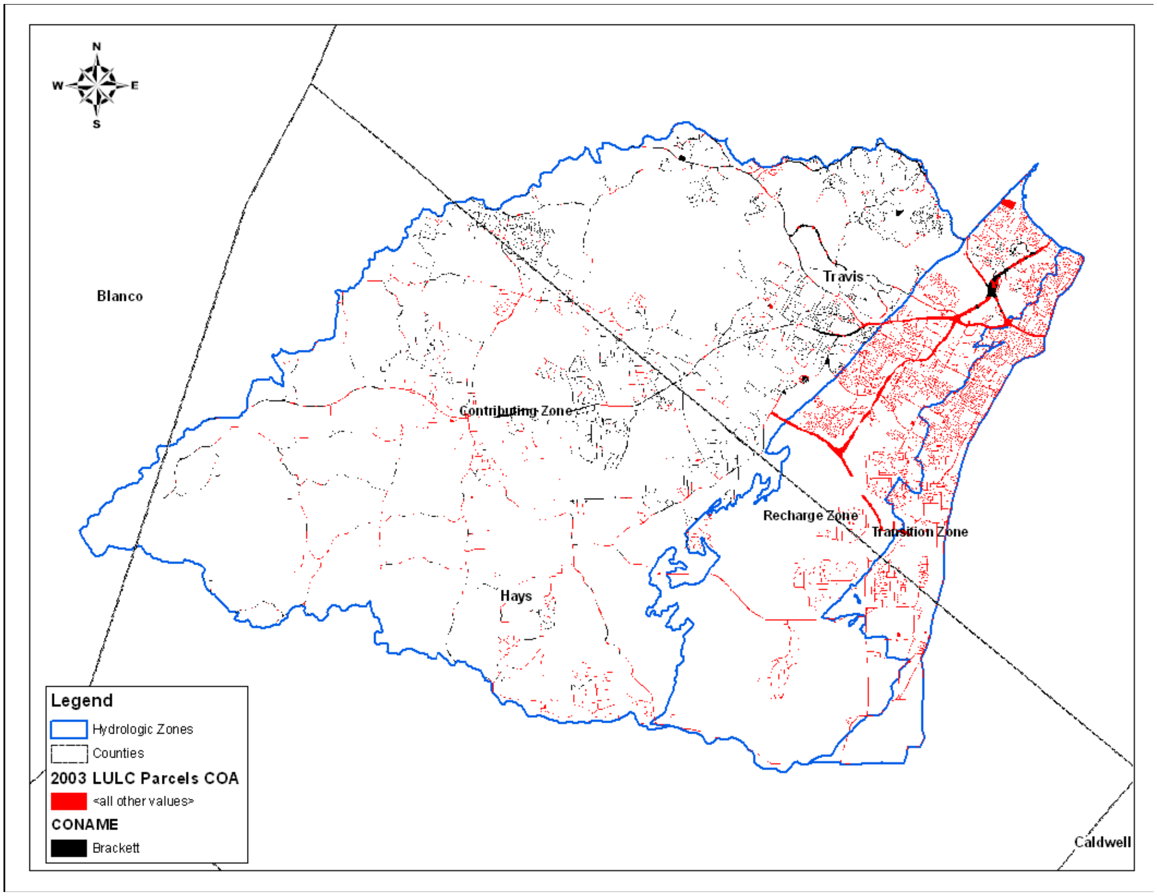

Figure 4. Location of Brackett soils in right-of-way areas (streets/roads/railroads/utilities) of the Barton Springs Segment of the Edwards Aquifer, Hays and Travis Counties, Texas.

Right-of-Way

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Crop cover parameters for this scenario were based on typical plants found adjacent to state maintained highway right-of ways. State-maintained highways include farm-to-market (FM) roads, state highways, interstates, and US highways. Bermuda grass is typically found in right-of-way areas in urban areas, while rural areas are dominated by native species such as little bluestem, side-oats grama, and hairy grama (John Mason, Vegetation Management Specialist, Texas DOT, Maintenance Div., personal communication).

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Rangeland/Pastureland

In the BSS, rangeland vegetation is a heterogeneous mixture of trees and grasses. Common tree species include: ash juniper (a nuisance species), oaks, hackberry and elms. Grass species including little blue stem, side oats gramma, Indian grass, switch grass, king ranch bluestem (introduced) and kline grass (introduced) are typical. These areas are composed of approximately 60-65% trees and 30-35% grasses (Perez 2006). Although this land cover contains a significant amount of tree cover, this “crop” was modeled as a field crop rather than an orchard in order to model a more conservative field. The analysis of land cover information is provided in Figure 5.

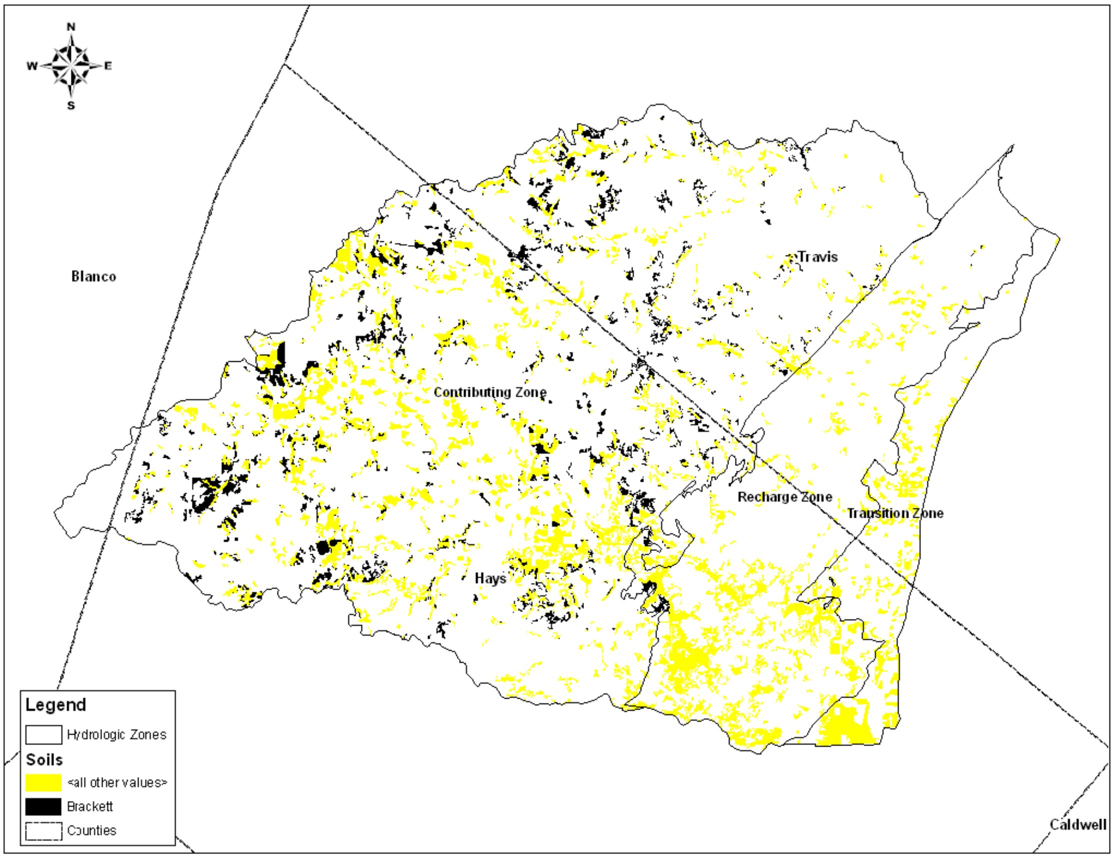

Figure 5. Location of Brackett Soils in natural herbaceous areas of the Barton Springs segment of the Edwards Aquifer, Hays and Travis Counties, Texas.

Meadow

This scenario is intended to represent a meadow that may include cultivation of herbaceous, non-grass animal feeds (forage, fodder, straw, and hay) (IR4 generalized crop group #18). The USDA census of agriculture (USDA 1997, 2002) indicates that hay of varying types is grown extensively in Travis and Hays Counties (Table 6). Discussions with extension agents in Hays and Travis counties indicated that some cultivation of sorghum hay, and hay grazer, or sweet sorghum does occur in the Barton Springs Segment. Bermuda grass is also planted but is primarily for grazing and not harvested (Perez 2006). Most of this type of crop is for livestock grazing (Davis, 2006). The analysis of land cover information is provided in Figure 6.

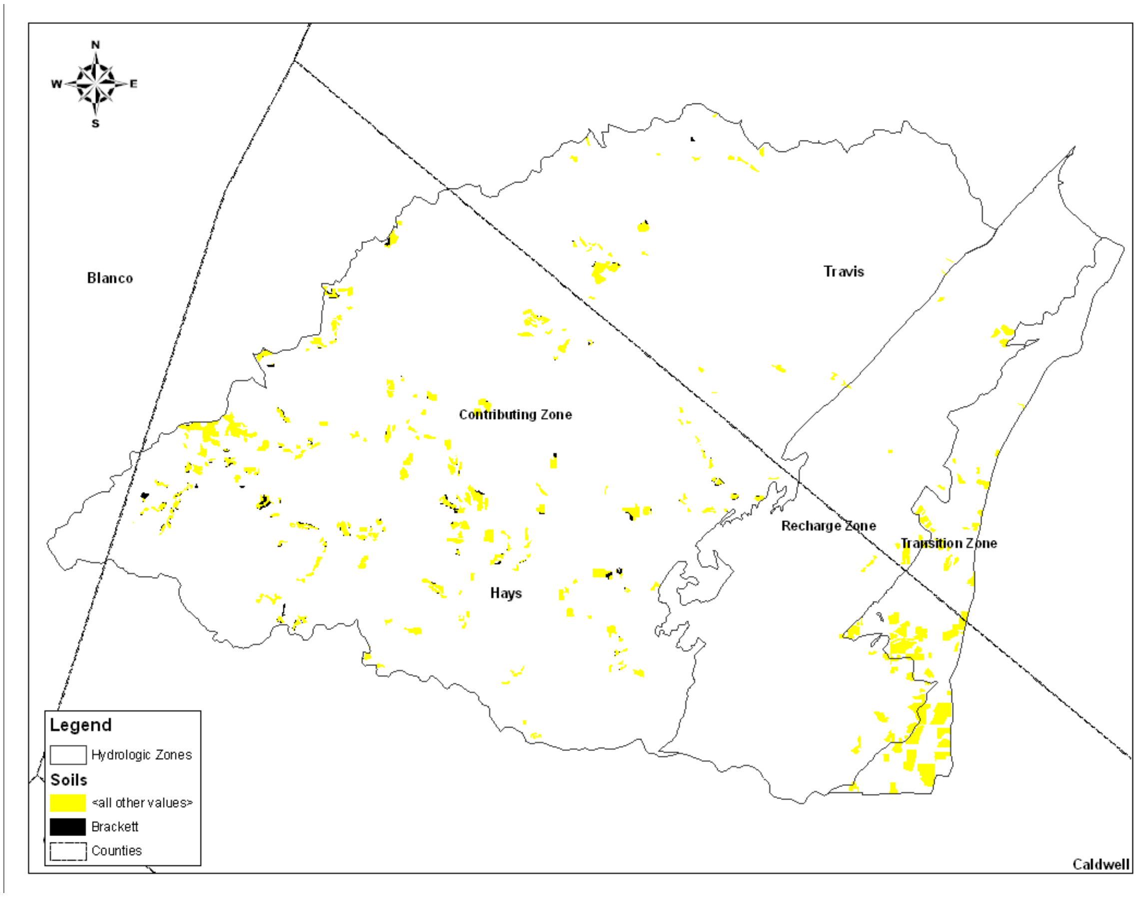

Figure 6. Location of Brackett soils in planted/cultivated areas of the Barton Springs segment of the Edwards Aquifer, Hays and Travis Counties, Texas.

Outdoor Nursery

The contractor conducted an investigation of wholesale nurseries in the BSZ using a variety of data sources to determine the extent of nurseries in the BSZ and the potential for *outside* pesticide use. NASS data for 2002 (**Table 1**) indicate that *outside* acreage for reported ornamental crops in all of Hays and Travis Counties is negligible relative to indoor acreage (< 0.1% total indoor and outdoor acreage). The majority of acreage for nursery, greenhouse, floriculture, mushrooms, sod, and vegetable seeds in both years and both counties was grown under glass or other protection. The contractor conducted a refined investigation to determine if this trend was similar in the BSZ.

Table 1. NASS 1997/2002 census of agriculture for ornamental production for open areas versus under glass in Hays and Travis Counties, Texas.

Crop	HAYS 1997 Total Acres	HAYS 2002 Total Acres	TRAVIS 1997 Total Acres	TRAVIS 2002 Total Acres
Nursery, greenhouse, floriculture, aquatic plants, mushrooms, flower seeds, vegetable seeds, sod harvested, total **In open**	x	65	x	111
Nursery, greenhouse, floriculture, aquatic plants, mushrooms, flower seeds, vegetable seeds, sod harvested, total **Under glass (not applicable for modeling)**	x	407,925	x	115,274
Nursery, floriculture, vegetable and flower seed crops, sod harvested, etc., grown in the open, irrigated	26	36	99	106
Floriculture crops – bedding/garden plants, cut flowers and cut florist greens, foliage plants, and potted flowering plants, total , in open	x	14	23	x
Bedding/garden plants, in open	4	x	6	4
Nursery stock, in open	2	27	73	90
Other nursery and greenhouse crops, in open	x	25	x	X

X = data not available, not applicable or withheld

Initially, nurseries in BSZ were identified through the Texas Nursery and Landscape Association Growers List, “Austin at a Glance Local Business Search”, and Google Local Maps. Five potential wholesale nurseries in the BSZ were identified. The contractor confirmed the existence of these nurseries and the potential for other through sources in the City of Austin Watershed Protection and Development Review Board (Kathy Shay, personal communication) and the Ladybird Johnson Wildflower Center (Andrea DeLong-Amaya, personal communication). Both sources confirmed these nurseries and neither source was aware of additional nurseries in the BSZ that would have outdoor wholesale nursery production. The contractor then contacted each of the five nurseries identified to determine the extent of outside production acreage and the potential for pesticide application. Total outside wholesale nursery production the entire Barton Spring Zone is approximately three acres. Only three of the five nurseries had outdoor wholesale production (Figure 1). Of these three, two had less than 0.5 acres outdoor production. The remaining site, Barton Springs Nursery, has approximately 2.5 acres of outdoor production. The Barton Springs Nursery has a reputation for being “environmentally conscious” (Kathy Shay, personal communication). When the nursery was contacted it indicated that it does use pesticides “when called for”.

For the purposes of modeling a nursery/ornamental operation in the BSS, one of the nurseries (Barton Springs Nursery) was used to conceptualize a facility that is representative of one located within the BSS. Communications with a staff member were used to parameterize the model. The nursery of interest has indoor and outdoor areas for growing and maintaining plants. Outdoor plants include cacti, annuals, perennials, shrubs, and trees. Outdoor plants are maintained on either weed control mat or on gravel. Plants are kept in pots of various sizes, ranging from 4” to multiple gallons, depending upon the type of plant kept within. Irrigation is carried out daily with either hose or sprinkler systems. Plants are maintained outside year-round, with some becoming dormant in the winter and some remaining green. Spring and fall represent the busiest times for plant production and sales for this nursery (personal communication with nursery employee). Several assumptions were made to parameterize the model. First, it was assumed that the area that would yield the greatest runoff potential would be from a bare surface that would be represented by the walkways between the potted plants. These areas could potentially receive direct applications of pesticides sprayed on potted plants. Therefore, the surface of the soil was conceptualized as being gravel or dirt (area under weed mats). This was an assumption that affected selection of curve numbers, USLE C and Manning’s N. Second, it was assumed that pesticide runoff of potted soil would not degrade or adsorb and would therefore, be applied directly to the soil.

The contractor also researched regulations for pesticide runoff from nurseries. Cindy Hooper of the TX Commission on Environmental Quality (TCEQ) Stormwater Team, which regulates the State TPDES for the federal NPDES, stated that the Nursery SIC code is 0181 which is an Agricultural type SIC code. Therefore nurseries are not required to have a TPDES Multi-Sector General Permit. Nancy McClintock, Assistant Director of the City of Austin Watershed Protection and Development Review Board indicated that a recent ordinance requires Integrated Pest Management (IPM) plans for new development; however the plan does not have specific pesticide runoff control requirements. It is important to note that this ordinance applies only to those areas of the BSZ under the jurisdiction of the City of Austin (approximately one-quarter of the BSZ). The analysis of land cover information is provided in Figure 7.

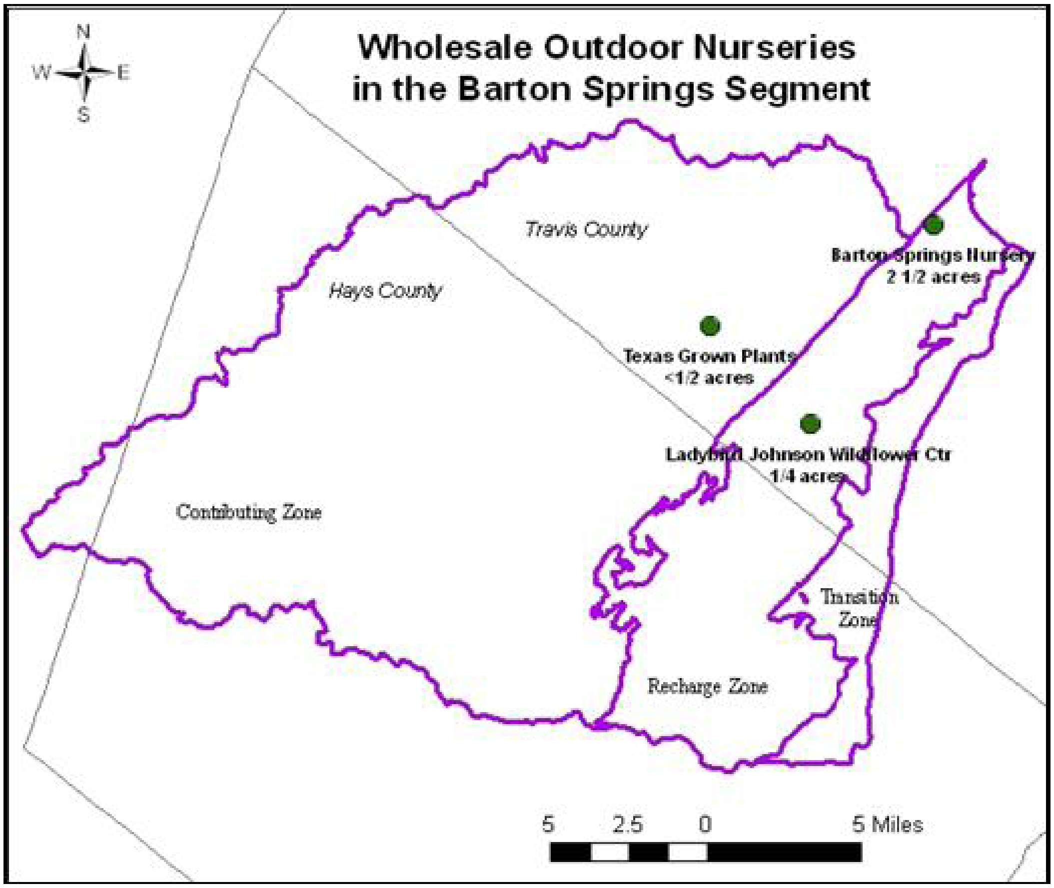

Figure 7. Location of outdoor wholesale nurseries in the Barton Springs segment of the Edwards Aquifer

LAND USE / LAND COVER ANALYSIS

Percent of each land use was computed for each of the land use / land cover datasets used in scenario development. Table 2 presents the percent of each land use as classified by USGS (2003) for the Barton Springs Segment in Hays and Travis counties, TX. Table 3 presents the percent of each land use as classified by COA (2003). Datasets were spatially "clipped" in ArcGIS to the area of interest as defined in the SOW for this assessment, specifically the Barton Springs Contributing, Recharge, and Transition zones in Hays and Travis Counties, TX.

Table 2. Percent of each land use in the Barton Springs Segment of Hays and Travis Counties, TX computed from USGS (2003) dataset. Based on the table " edw_lulc_BSS_AOI_UTM_SOIL " in the BartonSpringsAOI.mdb geodatabase

Land Use / Land Cover	Area (acres)	%	Related Scenario
Forested	138,670	54.60%	NA
Natural Herbaceous	37,700	14.84%	Rangeland
Single-Family Residential	28,352	11.16%	Residential
Mixed Forest/Shrub	26,068	10.26%	NA
Planted/Cultivated Herbaceous	8,098	3.19%	Meadow
Shrubland	5,989	2.36%	NA
Transportation	2,278	0.90%	NA
Commercial/Light Industry	1,537	0.61%	NA
Mixed Urban	1,339	0.53%	NA
Entertainment and Recreational	1,174	0.46%	NA
Institutional	854	0.34%	NA
Quarries/Strip Mines/Gravel Pits	720	0.28%	NA
Multi-Family Residential	546	0.22%	Residential
Reservoir	141	0.06%	NA
Agricultural Business	113	0.04%	NA
Communications And Utilities	90	0.04%	NA
Planted/Cultivated Woody (Orchards/Vineyards/Groves)	75	0.03%	Orchard
Transitional Bare	65	0.03%	NA
Heavy Industry	64	0.03%	NA
Stream/River	31	0.01%	NA
Bare Rock/Sand	22	0.01%	NA
Emergent Herbaceous Wetlands	20	0.01%	NA
Bare	16	0.01%	NA
Woody Wetland	12	0.00%	NA
Total*	253,974	100%	

* Note: Total area does not match exactly between the COA and USGS data sets due to differences in boundary delineations by each organization. USGS did not include Blanco

county and several fringe areas that were included in the COA dataset. Both datasets were clipped to the area of interest as defined in the SOW for this assessment, specifically the Barton Springs Contributing, Recharge, and Transition zones in Hays and Travis Counties, TX.

Table 3. Percent of each land use in the Barton Springs Segment of Hays and Travis Counties, TX computed from COA (2003) dataset. Based on the table "landuse2003_AOI_UTM_SOIL" in the BartonSpringsAOI.mdb geodatabase.

Land Use / Land Cover	Area (acres)	%	Related Scenario
Large-lot Single Family	71,669	28.2%	NA
Undeveloped	59,320	23.3%	NA
Agricultural	38,166	15.0%	NA
Single Family Residential	33,502	13.2%	NA
Preserves	20,020	7.9%	NA
Streets and Roads	10,684	4.2%	Right-of-way
Parks/Greenbelts	6,136	2.4%	NA
Mobile Homes	2,923	1.1%	NA
Commercial	2,353	0.9%	NA
Resource Extraction	1,713	0.7%	NA
Apartment/Condo	1,494	0.6%	NA
Educational	1,184	0.5%	NA
Golf Courses	1,152	0.5%	Turf
Warehousing	1,136	0.4%	NA
Office	792	0.3%	NA
Meeting and Assembly	752	0.3%	NA
Duplexes	505	0.2%	NA
Utilities	249	0.1%	Right-of-way
Three/Fourplex	157	0.1%	NA
Miscellaneous Industrial	154	0.1%	NA
Government Services	114	0.0%	NA
Aviation facilities	59	0.0%	NA
Hospitals	58	0.0%	NA
Water	52	0.0%	NA
Railroad Facilities	45	0.0%	Right-of-way
Cemeteries	39	0.0%	NA
Retirement Housing	26	0.0%	NA
Manufacturing	22	0.0%	NA
Parking	9	0.0%	NA
Marinas	3	0.0%	NA
Group Quarters	2	0.0%	NA
Semi-institutional Housing	0	0.0%	NA
Total*	254,490	100.0%	

* Note: Total area does not match exactly between the COA and USGS data sets due to differences in boundary delineations by each organization. USGS did not include Blanco county and several fringe areas that were included in the COA dataset. Both datasets were clipped to the area of interest as defined in the SOW for this assessment, specifically the Barton Springs Contributing, Recharge, and Transition zones in Hays and Travis Counties, TX.

CLIMATE AND TIME PARAMETERS

Geographic parameters located in table 1 of the metadata files were determined based on the AOI. The meteorological station selected for the scenarios was located in Austin, Texas (W13958). This station was the closest available weather station that included data required for PRZM. PFAC and ANETD values were determined for the location of the AOI as it corresponded to PRZM manual figures 5.1 and 5.2, respectively (USEPA 1998). It was assumed that snowfall could occur and persist based on meteorological data for Austin, which indicated that from 1971-2001, the average snowfall for the winter season was 0.6 inches (NOAA 2006); therefore, the SFAC value was set to correspond to the value representative of open areas (Table 5.1, USEPA 1998).

SOIL SELECTION/PARAMETERIZATION

Soil series were selected for the Barton Springs scenarios based on geospatial analysis and discussions with local experts. Percent of each soil type within a particular LULC of interest in the Barton Springs Segment (BSS) was determined by intersecting the LULC data sets (USGS 2003, COA 2003) with soils data (USDA 2006). Soils were then selected based on various factors, including: extent, representativeness, benchmark soil, and/or high vulnerability of soil to erosion.

The Brackett soil series was selected for six of the seven scenarios, including: residential, impervious, right-of-way, turf, meadow and rangeland/pastureland. The Tarrant soil series was selected for the nursery scenario. Data for these soils was obtained from Soil Data Mart (USDA 2006) for the county with the most extensive amount of the relevant LULC (Table 4). Values for thickness, bulk density, initial water content, field capacity, and wilting point were taken from soil data mart for the horizons of interest. Organic carbon was determined for each horizon with organic matter data that were adjusted using the relationship % OC = % Organic Matter/1.724 (Doucette 2000). In all scenarios, Soil Data Mart included information for an additional soil horizon. Since this horizon was bedrock, the horizon was not added to the soil profiles.

Table 4. Soil types and county locations of soil data for each of the Barton Springs scenarios.

Scenario	Soil	Soil Confirmed?	County
Meadow	Brackett-Rock Outcrop-Comfort Complex	yes	Hays
Rangeland/Pastureland	Brackett-Rock Outcrop-Comfort Complex	yes	Hays
Residential	Brackett-Rock Outcrop-Complex	yes	Travis
Impervious	Brackett-Rock Outcrop-Complex	yes	Travis
Turf	Brackett-Rock Outcrop-Complex	yes	Travis
Right-of-Way	Brackett-Rock Outcrop-Complex	yes	Travis
Nursery	Tarrant soils and urban land	No*	Travis

* See nursery soil selection information below.

The Brackett series approximates the 90th percentile of vulnerability, drainage, erodibility, and slope. The relatively low organic matter content is also expected to result in lower microbial activity and thus reduced potential for pesticide degradation. Brackett soils have a USLE K factor of 0.37 which includes the 90th percentile of these soils in erodibility. Brackett is a benchmark soil as well as a Hydrologic Group C. Slopes can range from 1 to 60 percent (Soil Survey Staff, 2006a); however the most typical range for the Brackett series in residential areas is either 1-8 percent (Hays County) or 1-12 percent (Travis County) (USDA 2006).

Tarrant is a Hydrologic Group D soil, with a USLE K factor of 0.32 (USDA 2006). Slopes range from 1 to 8 percent for this series (USDA 1997), but for the portion that overlaps with the nursery, the slope range is 0 to 2 percent. Since all three outdoor nursery operations in the BSS are located within Travis County, soil parameters were obtained soil data mart information pertaining to Travis County (USDA 2006).

Residential and Impervious

Soils were selected based on vulnerability and the extent within single- and multi-family residential areas in BSS. Based on a geospatial analysis of soils (USDA 2006) and land use data (USGS 2003) for residential areas as well as conversations with local soil experts, Brackett soils were chosen to represent residential areas in the BSS. Brackett soils are in Hydrologic Group C, are found in both the contributing and recharge zones of the Edwards Aquifer (Figure 1), and are the most common soil on which residential dwellings are located, accounting for 35% of all soils in residential areas (**Table** 5). Brackett soils are often undulating (Soil Survey Staff 2006a) making them desirable for development due to their scenic nature (Volente 2004). The location of Brackett soils was also cross-checked with aerial photography (TWDB 2004) to ensure that the soil chosen coincided with residential areas where pesticides would reasonably be applied. A local soil expert also confirmed that Brackett soil is a common soil type in residential areas of the BSS (Perez, 2006). A thatch layer was added to the top of the soil layer according to USEPA guidance on modeling turf, as provided with the SOW.

The impervious scenario is intended to be coupled to the residential scenario to mimic hydrology of untreated portions of the Barton Springs Segment (BSS) of the Edwards Aquifer. The intention is to couple the edge of field concentrations from this scenario with the edge of field concentrations from the residential scenario for Barton Springs to generate weighted concentrations for areas of varying impervious cover. Therefore, this scenario relies on a similar soil series as the residential scenario (Brackett); however the upper horizon has been adjusted to a non-soil nature. This included setting a high curve number, high bulk density, low curve number, and setting organic carbon to zero.

Percent area of soils in each Hydrologic Group within single/multi-family residential land use type (USGS 2003) in Barton Springs Segment of the Edwards Aquifer.	
Hydrologic Group	**Percent**
water/cut & fill /etc.	0.06%
A	0.37%
B	1.35%
C	47.14%
D	51.09%
	100.00%

Table 5. Analysis of Residential Soils Types.
Types of D soils in single- and multi-family residential land use type in the Barton Springs Segment of The Edwards Aquifer (percent of LULC in parenthesis).
Speck stony clay loam 16.9% (8.64%)
Comfort-Rock outcrop complex 12.6% (6.47%)
Real-Comfort-Doss complex 12.0% (6.13%)
Tarrant and Speck soils 8.55% (4.37%)
Tarrant soils and Urban land 7.11% (3.63%)
Tarrant soils 6.09% (3.11%)
Doss silty clay 5.55% (2.83%)
Denton silty clay 3.68% (1.88%)
Urban land and Brackett soils 2.61% (1.33%)
Urban land and Austin soils 2.57% (1.31%)
Crawford clay 2.42% (1.23%)
Urban land, Austin, and Whitewright soils 2.40% (1.23%)
Purves silty clay 2.13% (1.09%)
Krum clay 2.13% (1.09%)
Houston Black soils and Urban land 1.97% (1.01%)
Heiden clay 1.27% (0.65%)
San Saba soils and Urban land 1.12% (0.57%)
Medlin-Eckrant association 1.07% (0.54%)
Tarpley clay 1.01% (0.51%)
San Saba clay 0.95% (0.49%)
Purves clay 0.90% (0.46%)
Real gravelly loam 0.80% (0.41%)
Tarrant-Rock outcrop complex 0.75% (0.38%)
Speck clay loam 0.65% (0.33%)
Anhalt clay 0.63% (0.32%)
Urban land and Ferris soils 0.58% (0.29%)
Urban land 0.41% (0.21%)
Gruene clay 0.39% (0.20%)

Eckrant-Rock outcrop complex 0.19% (0.09%)
Ferris-Heiden complex 0.17% (0.09%)
Houston Black clay 0.10% (0.05%)
Tinn clay 0.03% (0.01%)
Types of C soils in single- and multi-family residential land use type in the Barton Springs Segment of The Edwards Aquifer (percent of LULC in parenthesis).
Brackett-Rock outcrop (Comfort or Real) complex 73.6% (34.7%)
Rumple-Comfort association 8.22% (3.88%)
Eddy soils and Urban land 4.88% (2.30%)
Volente silty clay loam 4.87% (2.29%)
Eddy gravelly loam 2.15% (1.01%)
Austin silty clay 2.09% (0.98%)
Bolar clay loam 1.26% (0.59%)
Volente soils and Urban land 1.23% (0.58%)
Castephen silty clay loam 0.94% (0.44%)
Austin-Castephen complex 0.42% (0.19%)
Altoga soils and Urban land 0.07% (0.03%)
Altoga silty clay 0.04% (0.02%)
Travis soils and urban land 0.02% (0.01%)
Whitewright clay loam 0.01% (0.00%)
Castephen clay loam 0.00% (0.00%)
Types of B soils in single- and multi-family residential land use type in the Barton Springs Segment of the Edwards Aquifer (percent of LULC in parenthesis).
Sunev clay loam 39.0% (0.52%)
Lewisville silty clay 19.7% (0.26%)
Patrick soils 14.9% (0.20%)
Lewisville soils and Urban land 10.4% (0.14%)
Patrick soils and urban land 6.90% (0.09%)
Sunev silty clay loam 2.82% (0.03%)
Seawillow clay loam 2.36% (0.03%)
Oakalla soils 2.08% (0.02%)
Hardeman soils and Urban land 0.80% (0.01%)
Oakalla silty clay loam 0.41% (0.00%)
Bergstrom soils and Urban land 0.33% (0.00%)
Boerne fine sandy loam 0.12% (0.00%)
Types of A soils in single- and multi-family residential land use type in the Barton Springs Segment of the Edwards Aquifer (percent of LULC in parenthesis).
Mixed alluvial land 82.4% (0.30%)
Orif soils 15.7% (0.05%)
Gaddy soils and Urban land 1.76% (0.00%)

Turf

Soil parameters were determined using data from Soil Data Mart (USDA 2006) for Travis County and land use data from the City of Austin (COA, 2003). This county data set was used since the majority of golf courses in the AOI reside within Travis County. The specific soil chosen was Brackett-Rock Outcrop-Complex, with 1-12% slopes, which is the most common soil located within golf course areas of BSS (Figure 3). A thatch layer was added to the top of the soil layer according to USEPA guidance on modeling turf, as provided with the SOW. The properties of the thatch layer are consistent with existing turf scenarios: PA turf and FL turf.

The Brackett series was chosen to represent turf areas in the BSS (Table 5) because it is a benchmark soil, is highly representative of golf course areas in the BSS, and it approximates the 90^{th} percentile of vulnerability in drainage, erodibility, and slope. Brackett soils are in Hydrologic Group C soils and are found in both the contributing and recharge zones of the Edwards Aquifer. Bracket soils are the most common soil type found in golf course areas of the BSS (Table 6).

Table 6. Analysis of Golf Course Soil Types.
Types of D soils in golf course land use type in the Barton Springs Segment of Edwards Aquifer (percent of LULC in parenthesis).
Tarrant 38.0% (12.5%)
Speck 28.6% (9.45%)
San Saba 19.3% (6.39%)
Crawford 11.4% (3.76%)
Doss 2.52% (0.83%)
Types of C soils in golf course land use type in the Barton Springs Segment of Edwards Aquifer (percent of LULC in parenthesis).
Brackett 77.6% (50.5%)
Volente 22.3% (14.5%)
Types of A soils in golf course land use type in the Barton Springs Segment of Edwards Aquifer (percent of LULC in parenthesis).
Alluvial land 100% (1.91%)

Right-of-way

Soils were chosen based on co-location with right-of-way areas based on land use coverage developed by the City of Austin (City of Austin 2003). The land use data set include streets, roads, utilities, and railroads, but does not include fence lines. Based on a geospatial analysis of right-of-way land uses (City of Austin 2003) and USDA soils data (USDA 2006), Brackett soils were chosen to represent right-of-way areas in the BSS. Brackett soils are found in both the contributing and recharge zones of the Edwards Aquifer and are the most common soil on which right-of-way areas are located (Figure 4), accounting for 32% of soils in right-of-way areas

(Table 7). The soil data for Travis County, Brackett-Rock Outcrop-Complex soil with slopes 112% was used to parameterize the soil component of this scenario (USDA 2006).

Table 7. Analysis of Right-of-way Soil Types.
Types of D soils in right-of-way (streets/roads/utilities/railroads) land use type in the Barton Springs Segment of Edwards Aquifer (percent of AOI in parenthesis).
Speck stony clay loam 23.5% (12.8%)
Tarrant and Speck soils 10.2% (5.54%)
Tarrant soils 7.05% (3.83%)
Real-Comfort-Doss complex 6.85% (3.72%)
Crawford clay 6.85% (3.72%)
Comfort-Rock outcrop complex 6.50% (3.53%)
Tarrant soils and Urban land 5.75% (3.12%)
Doss silty clay 4.07% (2.21%)
Denton silty clay 3.55% (1.93%)
Urban land and Austin soils 2.28% (1.23%)
San Saba clay 2.24% (1.21%)
Krum clay 2.22% (1.20%)
Heiden clay 2.08% (1.13%)
Purves silty clay 1.83% (0.99%)
Urban land Austin and Whitewright soils 1.59% (0.86%)
Houston Black soils and Urban land 1.54% (0.83%)
San Saba soils and Urban land 1.53% (0.83%)
Urban land and Brackett soils 1.38% (0.75%)
Urban land 1.18% (0.64%)
Tarpley clay 1.01% (0.55%)
Gruene clay 0.96% (0.52%)
Purves clay 0.84% (0.45%)
Medlin-Eckrant association 0.80% (0.43%)
Tarrant-Rock outcrop complex 0.77% (0.41%)
Speck clay loam 0.66% (0.36%)
Ferris-Heiden complex 0.59% (0.32%)
Anhalt clay 0.42% (0.23%)
Branyon clay 0.41% (0.22%)
Real gravelly loam 0.36% (0.19%)
Houston Black clay 0.32% (0.17%)
Urban land and Ferris soils 0.23% (0.12%)
Eckrant-Rock outcrop complex 0.15% (0.08%)
Tinn clay 0.07% (0.03%)
Types of C soils in right-of-way (streets/roads/utilities/railroads) land use type in the Barton Springs Segment of Edwards Aquifer (percent of AOI in parenthesis).

Brackett-Rock outcrop (Comfort or Real) complex 73.8% (32.2%)
Rumple-Comfort association 7.41% (3.23%)
Volente silty clay loam 6.52% (2.84%)
Eddy soils and Urban land 3.14% (1.37%)
Austin silty clay 2.56% (1.11%)
Bolar clay loam 1.95% (0.85%)
Eddy gravelly loam 1.68% (0.73%)
Castephen silty clay loam 1.06% (0.46%)
Volente soils and Urban land 0.89% (0.39%)
Austin-Castephen complex 0.60% (0.26%)
Castephen clay loam 0.18% (0.07%)
Travis soils and urban land 0.05% (0.02%)
Altoga soils and Urban land 0.03% (0.01%)
Whitewright clay loam 0.03% (0.01%)
Altoga silty clay 0.01% (0.00%)
Types of B soils in right-of-way (streets/roads/utilities/railroads) land use type in the Barton Springs Segment of Edwards Aquifer (percent of AOI in parenthesis).
Sunev clay loam 40.7% (0.60%)
Lewisville silty clay 21.5% (0.32%)
Patrick soils 10.9% (0.16%)
Lewisville soils and Urban land 5.63% (0.08%)
Hardeman soils and Urban land 5.36% (0.07%)
Patrick soils and urban land 4.93% (0.07%)
Oakalla silty clay loam 3.01% (0.04%)
Oakalla soils 2.92% (0.04%)
Bergstrom soils and Urban land 2.64% (0.03%)
Sunev silty clay loam 1.43% (0.02%)
Seawillow clay loam 0.77% (0.01%)
Types of A soils in right-of-way (streets/roads/utilities/railroads) land use type in the Barton Springs Segment of Edwards Aquifer (percent of AOI in parenthesis).
Mixed alluvial land 80.3% (0.46%)
Orif soils 19.2% (0.11%)
Gaddy soils and Urban land 0.30% (0.00%)

Rangeland/pastureland

Rangeland and pastureland were identified based on the natural herbaceous land cover classification in the BSS (USGS 2003). Based on the analysis of land use and soils data, Brackett soils were chosen to represent rangelands and pasturelands in the BSS (Table 5). Brackett soils are found in both the contributing and recharge zones of the Edwards Aquifer and are the most common soil on which rangeland is located (**Table** 8). This soil type was confirmed by an extension agent (Perez, 2006).

Percent area of soils in each Hydrologic Group within the natural herbaceous land use type (USGS 2003) in Barton Springs Segment of Edwards Aquifer.	
Hydrologic Group	**Percent**
water/cut & fill /etc.	0.25%
A	0.68%
B	6.67%
C	49.95%
D	42.45%
	100.00%

Table 8. Analysis of Rangeland Soil Types.
Types of D soils in natural herbaceous land use type in the Barton Springs Segment of Edwards Aquifer (percent of LULC in parenthesis).
Doss silty clay 25.1% (10.6%)
Real-Comfort-Doss complex 15.4% (6.54%)
Comfort-Rock outcrop complex 10.3% (4.40%)
Krum clay 6.58% (2.79%)
Tarpley clay 4.83% (2.04%)
Denton silty clay 4.74% (2.01%)
Purves clay 4.44% (1.88%)
Speck stony clay loam 3.14% (1.33%)
Crawford clay 2.86% (1.21%)
Houston Black clay 2.43% (1.03%)
Anhalt clay 2.22% (0.94%)
Gruene clay 2.14% (0.90%)
Tarrant soils 2.12% (0.89%)
Krum clay 1.99% (0.84%)
Purves silty clay 1.59% (0.67%)
Tarrant and Speck soils 1.51% (0.64%)
San Saba clay 1.10% (0.46%)
Branyon clay 0.98% (0.41%)

Heiden clay 0.87% (0.37%)
Denton silty clay 0.68% (0.28%)
Tinn clay 0.62% (0.26%)
Heiden clay 0.54% (0.22%)
Speck clay loam 0.43% (0.18%)
Real gravelly loam 0.39% (0.16%)
Eckrant-Rock outcrop complex 0.35% (0.15%)
Heiden clay 0.33% (0.14%)
Medlin-Eckrant association 0.32% (0.13%)
Denton silty clay 0.27% (0.11%)
Medlin-Eckrant association 0.27% (0.11%)
Krum clay 0.24% (0.10%)
Urban land and Austin soils 0.21% (0.09%)
Crawford clay 0.18% (0.07%)
Heiden clay 0.10% (0.04%)
Houston Black clay 0.10% (0.04%)
Tarrant soils and Urban land 0.08% (0.03%)
San Saba soils and Urban land 0.07% (0.03%)
Urban land, Austin and Whitewright soils 0.06% (0.02%)
Urban land 0.03% (0.01%)
Tarrant-Rock outcrop complex 0.02% (0.01%)
Branyon clay 0.02% (0.00%)
Houston Black clay 0.00% (0.00%)
Houston Black soils and Urban land 0.00% (0.00%)
Ferris-Heiden complex 0.00% (0.00%)
Tarrant soils and Urban land 0.00% (0.00%)
Tarrant soils and Urban land 1.48% (6.31%)
Types of C soils in natural herbaceous land use type in the Barton Springs
Segment of Edwards Aquifer (percent of LULC in parenthesis).
Brackett-Rock outcrop (Comfort or Real) complex 82.9% (22.7%)
Rumple-Comfort association 57.7% (15.8%)
Bolar clay loam 15.4% (4.24%)
Volente silty clay loam 14.3% (3.93%)
Austin-Castephen complex 4.78% (1.31%)
Austin silty clay 1.73% (0.47%)
Austin-Castephen complex 1.63% (0.44%)
Volente silty clay loam 1.44% (0.39%)
Castephen silty clay loam 1.27% (0.34%)
Castephen silty clay loam 0.40% (0.11%)
Altoga silty clay 0.33% (0.09%)

Castephen clay loam 0.33% (0.09%)
Austin silty clay 0.26% (0.07%)
Altoga silty clay 0.11% (0.03%)
Eddy gravelly loam 0.08% (0.02%)
Eddy gravelly loam 0.03% (0.00%)
Eddy soils and Urban land 0.02% (0.00%)
Travis soils and urban land 0.00% (0.00%)
Types of B soils in natural herbaceous land use type in the Barton Springs Segment of Edwards Aquifer (percent of LULC in parenthesis).
Sunev clay loam 54.1% (3.62%)
Lewisville silty clay 25.0% (1.67%)
Seawillow clay loam 3.10% (0.20%)
Boerne fine sandy loam 2.89% (0.19%)
Seawillow clay loam 2.49% (0.16%)
Lewisville silty clay 2.26% (0.15%)
Oakalla silty clay loam 2.05% (0.13%)
Sunev silty clay loam 2.05% (0.13%)
Lewisville silty clay 1.49% (0.09%)
Oakalla soils 1.27% (0.08%)
Patrick soils 1.21% (0.08%)
Lewisville silty clay 1.16% (0.07%)
Patrick soils 0.43% (0.02%)
Oakalla soils 0.17% (0.01%)
Patrick soils and urban land 0.12% (0.00%)
Hardeman soils and Urban land 0.06% (0.00%)
Lewisville soils and Urban land 0.04% (0.00%)
Types of A soils in natural herbaceous land use type in the Barton Springs Segment of Edwards Aquifer (percent of LULC in parenthesis).
Mixed alluvial land 76.3% (0.52%)
Orif soils 23.6% (0.16%)
Gaddy soils and Urban land 0.02% (0.00%)

Meadow

Soils were selected based on the extent within herbaceous planted areas in BSS and the potential to yield high-end runoff and erosion. Based on a geospatial analysis of soils (USDA 2006) and land use data (USGS 2003) for herbaceous planted areas as well as conversations with local soil experts, Brackett soils were chosen to represent meadow areas in the BSS (Table 5). Location of the Brackett soils was also cross-checked with aerial photography (TWDB 2004) to ensure that the soil chosen coincided with herbaceous planted areas where pesticides would reasonably be applied. A local soil expert also confirmed that Brackett soils are extensive soil types of meadows in the BSS (Perez 2006). Brackett soils while not the most extensive soil in this land use; it is the second most extensive *benchmark soil* in the herbaceous planted land use. One benchmark soil is more extensive (Denton), however Brackett was chosen over this soil since Brackett soils have a higher erodibility potential. Data from Hays County were selected since the majority of this LULC is located in this county.

Planted/Cultivated herbaceous land use type in USGS (2003) data set	
Hydrologic Group	**Percent**
water	0.03%
A	0.15%
B	16.27%
C	17.76%
D	65.79%
	100.00%

Table 9. Analysis of Meadow Soil Types.
Types of D soils in herbaceous planted land use type in the Barton Springs Segment of Edwards Aquifer (percent in LULC in parenthesis).
Doss silty clay 28.2% (18.5%)
Krum clay 21.4% (14.0%)
Denton silty clay 7.91% (5.20%)
Heiden clay 6.61% (4.35%)
Houston Black clay 5.84% (3.84%)
Tarpley clay 4.05% (2.66%)
Anhalt clay 3.73% (2.45%)
Purves clay 3.64% (2.39%)
Crawford clay 3.48% (2.29%)
Gruene clay 3.10% (2.04%)
Branyon clay 2.24% (1.47%)
Purves silty clay 2.19% (1.44%)
Speck clay loam 1.95% (1.28%)
Real-Comfort-Doss complex 1.94% (1.28%)
San Saba clay 1.28% (0.84%)
Comfort-Rock outcrop complex 0.84% (0.55%)
Medlin-Eckrant association 0.59% (0.39%)
Real gravelly loam 0.22% (0.14%)
Speck stony clay loam 0.20% (0.13%)
Tarrant and Speck soils 0.13% (0.09%)
Tinn clay 0.12% (0.08%)
Tarrant soils 0.10% (0.07%)
Urban land and Austin soils 0.07% (0.04%)
Urban land, Austin, and Whitewright soils 0.02% (0.01%)
Eckrant-Rock outcrop complex 0.00% (0.00%)

Types of C soils in herbaceous planted land use type in the Barton Springs Segment of Edwards Aquifer (percent in LULC in parenthesis).
Brackett-Rock outcrop (Comfort or Real) complex 25.5% (4.54%)
Bolar clay loam 23.8% (4.24%)
Austin-Castephen complex 23.6% (4.20%)
Volente silty clay loam 13.4% (2.38%)
Rumple-Comfort association 6.66% (1.18%)
Castephen clay loam 3.84% (0.68%)
Austin silty clay 1.91% (0.33%)
Castephen silty clay loam 0.93% (0.16%)
Eddy soils and Urban land 0.12% (0.02%)
Volente soils and Urban land 0.03% (0.00%)
Eddy gravelly loam 0.03% (0.00%)
Types of B soils in herbaceous planted land use type in the Barton Springs
Segment of Edwards Aquifer (percent in LULC in parenthesis).
Sunev clay loam 55.6% (9.06%)
Lewisville silty clay 30.1% (3.98%)
Seawillow clay loam 16.7% (2.22%)
Sunev silty clay loam 3.89% (0.51%)
Oakalla silty clay loam 1.97% (0.26%)
Boerne fine sandy loam 0.66% (0.08%)
Patrick soils 0.66% (0.08%)
Oakalla soils 0.51% (0.06%)
Types of A soils in herbaceous planted land use type in the Barton Springs
Segment of Edwards Aquifer (percent in LULC in parenthesis).
Orif soils 81.1% (0.12%)
Mixed alluvial land 18.8% (0.02%)

Outdoor nursery

The soil selected for the nursery scenario was selected based on the overlap between the nursery of interest (Barton Springs Nursery) and soil extents (USDA 2006). Aerial photography (TWDB 2004) was used to identify the location of the nursery operation and the locations of the outdoor areas of production. Only one soil type overlapped with the nursery operation: Tarrant soils and urban land. Therefore, it was determined that this soil type was a representative soil that an outdoor nursery operation in the BSS would reside upon. Since all three outdoor nursery operations in the BSS are located within Travis County, soil parameters were obtained soil data mart information pertaining to Travis County (USDA 2006).

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RESEARCH AND DOCUMENTATION FOR AGRICULTURAL SCENARIOS EVALUATED FOR THE BARTON SPRINGS SALAMANDER ASSESSMENT

Overview

This appendix is intended to supplement the summary report submitted by the contractor under technical direction (TD) No. 3 (GSA Contract No. GS-00F-0019L, Order Number. EP06H000149). The SOW for TD3 indicated that seven optional scenarios may be required, depending on the existence of potential uses in the Barton Springs Segment. The scenarios included:

1 Forestry;
2 Row crops (Table 2-2b of USDA TR55);
3 Small grains (Table 2-2b of USDA TR55);
4 Close seeded legumes (Table 2-2b of USDA TR55);
5 Woods-grass combination (orchard or tree farm) (Table 2-2c of USDA TR55);
6 Meadow (Table 2-2c of USDA TR55); and
7 Cotton

For the seven optional generic scenarios, the contractor conducted preliminary background research on each of the suggested uses to determine the presence of the use site in the area of interest the level of significance of the use. The contractor provided an interim deliverable report documenting the preliminary research on 6 March 2006. The Agency directed the contractor to proceed based on the recommendations, but to also further investigate the need for the orchard scenario. The Agency indicated if the contractor can confirm these are in the contributing zone but not the recharge zone then document as such and do not develop these scenarios. If the crop is possibly in the recharge zone then the scenario may need to developed, even with a limited acreage. The contractor determined that the one (1) orchard located in the recharge zone based on land use (USGS 2003) is no longer active; the land has been converted to a Lowes home center.

According to GIS land use coverage from the Texas Commission on Environmental Quality and the City of Austin, agricultural land uses do exist extensively throughout the in the Barton Springs Recharge and Contributing Zones (hereafter referred to as the AOI or “Area of Interest”), However, most of this agricultural land is used for range land, livestock grazing, and pasture, according to the extension agents from Hays and Travis Counties. All extension agents indicated the prevailing trend of agricultural and range land being broken up and converted to residential and commercial development.

Eddie Garcia from Travis County indicated that there are no crops commercially grown and harvested in the AOI of Travis County. There may be some grazing but usually it’s not even enough pasture so that supplemental food must be purchased for the livestock. There is forested/wooded land but no forestry operations for planting and harvesting. The Nature Conservancy owns 4600 acres in the AOI and is managing it as a natural area. There are no agricultural producers registered with the Farm Service Agency (FSA) in the Barton Springs AOI.

Scenario Background Research

1. Forestry

NASS data indicates that a small amount of Christmas trees are grown in Travis County (Table 10), however the extension agents from Travis and Hays Counties indicated that these crops are not grown the AOI. There is some cedar and juniper removal. These are considered pests and are removed and not sold (Perez 2006). There is a chemical that can be used for removing cedar, but no one uses it in the BSS; most people cut nuisance trees down (Davis 2006). Based on the information from local extension agents, this use was deemed outside the area of interest and was not developed

Table 10. NASS 1997/2002 census of agriculture for Christmas trees in Hays and Travis Counties, Texas (USDA 1997, 2002).

	HAYS		TRAVIS	
Crop	**1997 Acres in Production**	**2002 Acres in Production**	**1997 Acres in Production**	**2002 Acres in Production**
Cut Christmas trees	X	X	X	9

X = data not available, not applicable or withheld

2. Row Crops

NASS data indicates that a small amount of vegetable crops are the only row crops that are grown in Travis and Hays Counties (Table 11), however the extension agents from Travis and Hays Counties indicated that these crops are not grown the AOI commercially, only in residential gardens. There is one certified organic farm near Wimberly but not within the AOI (Perez 2006). The only vegetables are in home gardens (Davis 2006). Based on the information from local extension agents, this use was deemed outside the area of interest and was not developed

Table 11. NASS 1997/2002 census of agriculture for vegetable crops in Hays and Travis Counties, Texas (USDA 1997, 2002).				
	HAYS		TRAVIS	
Crop	1997 Harvested Acres	2002 Harvested Acres	1997 Harvested Acres	2002 Harvested Acres
Land Used For Vegetables	13	11	19	17
Vegetables Harvested For Sale	24	39	52	37
Turnips	X	1	X	X
Herbs, Fresh Cut	10	4	X	X
Carrots	1	X	X	X
Dry Onions	X	1	X	2
Peppers, Bell	X	X	X	1
Peppers, Chile (All Peppers - Excluding Bell)	X	X	X	3
Tomatoes	2	4	2	9
Okra	X	3	1	3
Cantaloups	1	3	X	2
Watermelons	1	X	X	1
Cucumbers And Pickles	1	X	X	X
Squash	1	3	X	X
Beets	X	X	X	2

X = data not available, not applicable or withheld

3. Small Grains

NASS data indicate that corn, oats, sorghum, and wheat are grown extensively in Travis and Hays Counties (Table 12). According to Soil Data Mart, there are numerous soils in the BSS that are suitable for growing corn, grain sorghum, and wheat; however, Hays and Travis County extension agents from Travis and Hays Counties indicated that small grain crops are not cultivated in the BSS. In cases where small grains are planted such as winter wheat or oats they are used exclusively for harvesting from small plots from 5 to 15 acres (Davis 2006). All other grain crops like corn, sorghum, wheat, oats and milo are grown East of I-35 in the Blackland Prairie region (Perez 2006). Based on the information from local extension agents, this use was deemed outside the area of interest and was not developed

Table 12. NASS 1997/2002 census of agriculture for grain crops in Hays and Travis Counties, Texas (USDA 1997, 2002).				
	HAYS		**TRAVIS**	
Crop	**1997 Harvested Acres**	**2002 Harvested Acres**	**1997 Harvested Acres**	**2002 Harvested Acres**
Corn For Grain	5915	3084	12139	12378
Oats For Grain	836	X	215	206
Sorghum For Grain	5406	1435	21298	14684
Wheat For Grain, All	4674	3527	4849	3320
Winter Wheat For Grain	X	3527	X	3320
Sweet Corn	1	1	X	3

X = data not available, not applicable or withheld

4. Close-seeded legumes

NASS data indicates that a small amount of close-seeded legumes are grown in Travis and Hays Counties (Table 13), however the extension agents from Travis and Hays Counties indicated that these crops are not grown in the AOI (Perez 2006; Davis 2006). Based on the limited extent of legumes in Hays and Travis counties and information from local extension agents, this use was deemed outside the area of interest and was not developed

Table 13. NASS 1997/2002 census of agriculture for legumes in Hays and Travis Counties, Texas (USDA 1997, 2002).				
	HAYS		**TRAVIS**	
Crop	**1997 Harvested Acres**	**2002 Harvested Acres**	**1997 Harvested Acres**	**2002 Harvested Acres**
Peas, Green Southern (Cowpeas) - Blackeyed, Crowder, Etc.	X	1	X	X
Snap Beans	X	4	X	1

X = data not available, not applicable or withheld

5. Orchard or Tree Farms

NASS data indicates that orchard crops are grown in Travis and Hays Counties (Table 14); however the extension agent from Travis County indicated that there are no orchards in the BSS. The extension agent from Hays County indicated that there is one location in the BSS where orchard crops are grown: the orchard at the Barsana Dham-Isdl Temple (on FM1826) where they grow persimmons, peaches, pecans, etc. These are grown for Pick-Your-Own and they use low toxicity IPM (Integrated Pest Management) practices there (Davis 2006). All orchard crops like peaches and pecans are not in the AOI but near the San Marcos and Blanco Rivers (Perez 2006). EFED reviewed the initial recommendation and directed the contractor to further investigate the need for the orchard scenario. The Agency indicated that if there is minimal acreage in the recharge zone (e.g., nurseries) that could contribute to exposures, then the scenario may be developed. Based on USGS (2003) land use data, the contractor identified one (1) orchard located in the recharge zone (Figure 15). Conversations with personnel in the city of Austin GIS department indicated the orchard is no longer active and has been rezoned for a Lowes® home center (COA, personal communication). Based on this information it was deemed that this orchard will not contribute to potential exposures in the BSS and therefore has not been developed.

Table 14. NASS 1997/2002 census of agriculture for orchard crops in Hays and Travis Counties, Texas (USDA 1997, 2002).

	HAYS		TRAVIS	
Crop	**1997 Total Acres**	**2002 Total Acres**	**1997 Total Acres**	**2002 Total Acres**
Land In Orchards	260	290	1394	1793
Apples	X	10	X	X
Pears, All	X	9	X	7
Apricots	X	16	X	X
Peaches, All	X	76	X	22
Plums And Prunes	X	6	X	X
Pecans	X	143	X	1720
Grapes	X	31	X	38

X = data not available, not applicable or withheld

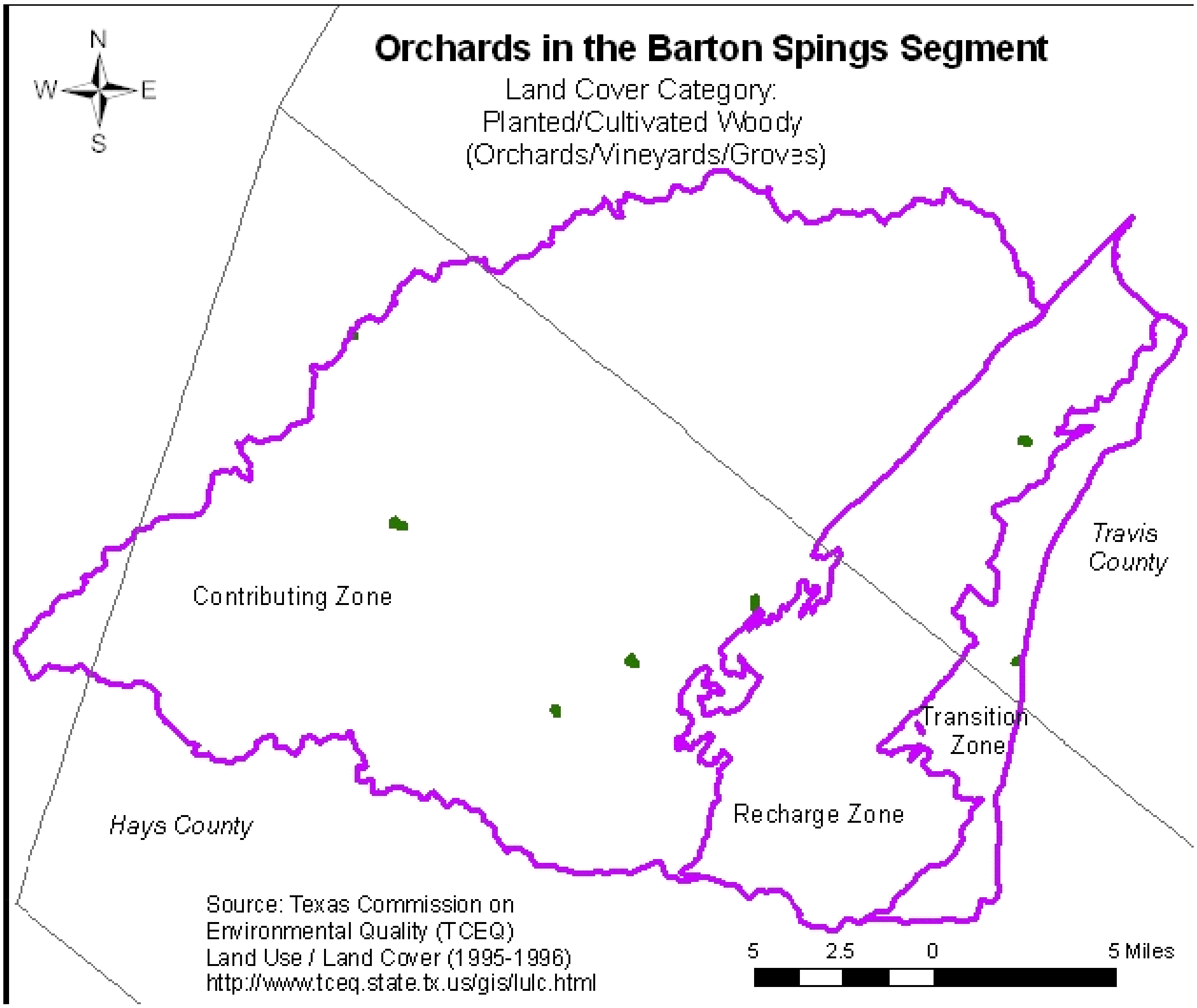

Figure 15. Location of woody planted areas in the BSS segment based on land use data. Local contacts indicated orchards are not present or not active in the BSS. See description for more information.

6. Meadow

NASS Data indicates that hay of varying types is grown extensively in Travis and Hays Counties (Table 15). According to Soil Data Mart, there are a number of soils in the BSS that are suitable for growing improved bermudagrass. In addition, extension agents indicated that some hay crops are cultivated in the BSS. There is some cultivation of sorghum hay, and hay grazer, or sweet sorghum in the BSS. There is also some bermuda grass planted but this is permanent for grazing and not harvested (Perez 2006). Most of this type of crop is for livestock grazing (Davis 2006). Based on this information, this scenario was developed.

Table 15. NASS 1997/2002 census of agriculture for hay crops in Hays and Travis Counties, Texas (USDA 1997, 2002).

	HAYS		TRAVIS	
Crop	**1997 Harvested Acres**	**2002 Harvested Acres**	**1997 Harvested Acres**	**2002 Harvested Acres**
Hay - All Hay Including Alfalfa, Other Tame, Small Grain, And Wild	X	7657	X	20471
All Haylage, Grass Silage, And Greenchop	140	229	769	357
Forage - Land Used For All Hay And All Haylage, Grass Silage, And Greenchop	X	7855	X	20367
Other Haylage, Grass Silage, And Greenchop	X	229	X	357
Other Tame Hay	8287	5358	14020	16737
Small Grain Hay	600	X	943	2219
Wild Hay	840	1228	X	1411
Alfalfa Hay	65	X	X	104

X = data not available, not applicable or withheld

7. Cotton

NASS data indicates that cotton is grown in Travis County (Table 16). According to Soil Data Mart, there are many soils in the AOI that are suitable for growing cotton. However, the extension agents from Travis and Hays Counties indicated that this crop is not grown in the AOI. All cotton is grown East of I-35 (Perez 2006 and Davis 2006). Based on the information from local extension agents, this use was deemed outside the area of interest and was not developed.

Table 16. NASS 1997/2002 census of agriculture for cotton in Hays and Travis Counties, Texas (USDA 1997, 2002).				
	HAYS		TRAVIS	
Crop	1997 Harvested Acres	2002 Harvested Acres	1997 Harvested Acres	2002 Harvested Acres
Cotton, All	X	X	5661	2151
Upland Cotton	X	X	X	2151

X = data not available, not applicable or withheld

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Appendix C. USGS Monitoring Data for Barton Springs Area.

Samples were collected by USGS from the 4 springs, from surface waters in the action area (creeks) and from ground water wells in and around the action area. Samples were later measured for diazinon. Tables C.1, C.2, and C.3 contain detailed information of all samples collected and their measured concentrations of diazinon in the springs, creeks and ground water wells. Figures 3.9 and 3.10 (in the risk assessment) contain locations of surface water sites and ground water wells which correspond to the site nicknames cited in tables C.2 and C.3, respectively.

Samples were collected from the four springs between 2000 and 2005. During August and September of 2003, samples were collected every two weeks. From Mid June to December, 2004, samples were collected every three weeks. Stormflow sampling was also conducted in 2000, 2001, 2004 and 2005.

Table C.1. USGS targeted monitoring data for Barton Springs.

pk_siteID	siteNickname	sampleDate (year, month, date)	sampleTime	Diazinon* Conc. (ppb)	U/F**
08155500	Main Barton Spring	19780718	0850	.03	U
08155500	Main Barton Spring	19780927	1300	0	U
08155500	Main Barton Spring	19781205	1245	0	U
08155500	Main Barton Spring	19790228	0950	0	U
08155500	Main Barton Spring	19800116	0830	0	U
08155500	Main Barton Spring	19800604	0920	0	U
08155500	Main Barton Spring	19801017	0850	0	U
08155500	Main Barton Spring	19810408	1315	0	U
08155500	Main Barton Spring	19810527	1000	0	U
08155500	Main Barton Spring	19810824	0845	0	U
08155500	Main Barton Spring	19910826	2040	< .01	U
08155500	Main Barton Spring	19920324	0930	< .01	U
08155500	Main Barton Spring	19920330	0945	< .01	U
08155500	Main Barton Spring	19920521	1315	< .01	U
08155500	Main Barton Spring	19930114	1330	< .01	U
08155500	Main Barton Spring	19930211	1112	< .01	U
08155500	Main Barton Spring	19931129	1429	< .01	U
08155500	Main Barton Spring	20000501	1055	< .002	F
08155500	Main Barton Spring	20000501	1820	< .002	F
08155500	Main Barton Spring	20000501	2305	.0089	F
08155500	Main Barton Spring	20000502	1145	.0208	F
08155500	Main Barton Spring	20000502	1420	.0235	F
08155500	Main Barton Spring	20000502	1812	.0281	F
08155500	Main Barton Spring	20000503	1240	.0192	F
08155500	Main Barton Spring	20000504	1015	.0075	F
08155500	Main Barton Spring	20000508	1300	< .002	F
08155500	Main Barton Spring	20000609	1940	< .002	F

08155500	Main Barton Spring	20000609	2035	< .002	F
08155500	Main Barton Spring	20000610	1030	.00904	F
08155500	Main Barton Spring	20000705	0930	< .002	F
08155500	Main Barton Spring	20010503	2320	< .005	F
08155500	Main Barton Spring	20010508	1950	E .00459	F
08155500	Main Barton Spring	20010510	1440	< .005	F
08155500	Main Barton Spring	20010510	1442	< .005	F
08155500	Main Barton Spring	20010513	1955	< .005	F
08155500	Main Barton Spring	20010518	2100	< .005	F
08155500	Main Barton Spring	20011116	1200	.0069	F
08155500	Main Barton Spring	20021106	1243	< .005	F
08155500	Main Barton Spring	20030220	1845	< .005	F
08155500	Main Barton Spring	20030806	1145	< .005	F
08155500	Main Barton Spring	20030820	0830	< .005	F
08155500	Main Barton Spring	20030903	0800	< .005	F
08155500	Main Barton Spring	20030916	0730	< .005	F
08155500	Main Barton Spring	20030930	0700	< .005	F
08155500	Main Barton Spring	20040117	0830	< .005	F
08155500	Main Barton Spring	20040609	0900	< .005	F
08155500	Main Barton Spring	20040621	1430	< .005	F
08155500	Main Barton Spring	20040707	1300	< .005	F
08155500	Main Barton Spring	20040721	0730	< .005	F
08155500	Main Barton Spring	20040804	0800	< .005	F
08155500	Main Barton Spring	20040825	1000	< .005	F
08155500	Main Barton Spring	20040915	0900	< .005	F
08155500	Main Barton Spring	20041004	1200	< .005	F
08155500	Main Barton Spring	20041023	1400	< .005	F
08155500	Main Barton Spring	20041023	1402	< .005	F
08155500	Main Barton Spring	20041024	1000	< .005	F
08155500	Main Barton Spring	20041024	2100	< .005	F
08155500	Main Barton Spring	20041024	2102	< .005	F
08155500	Main Barton Spring	20041025	1030	< .005	F
08155500	Main Barton Spring	20041026	0900	< .005	F
08155500	Main Barton Spring	20041027	1100	< .005	F
08155500	Main Barton Spring	20041028	0900	< .005	F
08155500	Main Barton Spring	20041030	1000	< .005	F
08155500	Main Barton Spring	20041105	1030	< .005	F
08155500	Main Barton Spring	20041124	1100	< .005	F
08155500	Main Barton Spring	20041214	1500	< .005	F
08155500	Main Barton Spring	20050103	0930	< .005	F
08155500	Main Barton Spring	20050126	0930	< .005	F
08155500	Main Barton Spring	20050216	0800	< .005	F
08155500	Main Barton Spring	20050309	0730	< .005	F
08155500	Main Barton Spring	20050330	0800	< .005	F
08155500	Main Barton Spring	20050420	0730	< .005	F
08155500	Main Barton Spring	20050511	0800	< .005	F
08155500	Main Barton Spring	20050530	0730	< .005	F
08155500	Main Barton Spring	20050530	1400	< .005	F

08155500	Main Barton Spring	20050530	2100	< .005	F
08155500	Main Barton Spring	20050531	1030	< .005	F
08155500	Main Barton Spring	20050601	0730	< .005	F
08155500	Main Barton Spring	20050602	0730	< .005	F
08155500	Main Barton Spring	20050604	0930	< .005	F
08155500	Main Barton Spring	20050606	0730	< .005	F
08155500	Main Barton Spring	20050609	0800	< .005	F
08155395	Upper Barton Spring	20010508	1000	.143	F
08155395	Upper Barton Spring	20010510	1510	E .00478	F
08155395	Upper Barton Spring	20010513	1935	< .005	F
08155395	Upper Barton Spring	20020503	1630	< .005	F
08155395	Upper Barton Spring	20030806	1230	< .005	F
08155395	Upper Barton Spring	20030820	0900	< .005	F
08155395	Upper Barton Spring	20030903	0730	< .005	F
08155395	Upper Barton Spring	20030916	0630	< .005	F
08155395	Upper Barton Spring	20030930	0730	< .005	F
08155395	Upper Barton Spring	20040621	1500	< .005	F
08155395	Upper Barton Spring	20040707	1230	< .005	F
08155395	Upper Barton Spring	20040721	0930	< .005	F
08155395	Upper Barton Spring	20040804	0900	< .005	F
08155395	Upper Barton Spring	20040825	0830	< .005	F
08155395	Upper Barton Spring	20040915	0800	< .005	F
08155395	Upper Barton Spring	20041004	1030	< .005	F
08155395	Upper Barton Spring	20041023	1500	< .005	F
08155395	Upper Barton Spring	20041024	0930	.0165	F
08155395	Upper Barton Spring	20041024	2030	E .0037	F
08155395	Upper Barton Spring	20041025	1000	E .0038	F
08155395	Upper Barton Spring	20041026	0830	< .005	F
08155395	Upper Barton Spring	20041027	1030	< .005	F
08155395	Upper Barton Spring	20041028	0830	< .005	F
08155395	Upper Barton Spring	20041030	0900	< .005	F
08155395	Upper Barton Spring	20041105	0930	< .005	F
08155395	Upper Barton Spring	20041124	1000	< .005	F
08155395	Upper Barton Spring	20041214	1430	< .005	F
08155395	Upper Barton Spring	20050103	0830	< .005	F
08155395	Upper Barton Spring	20050126	0800	< .005	F
08155395	Upper Barton Spring	20050216	0730	< .005	F
08155395	Upper Barton Spring	20050309	0700	< .005	F
08155395	Upper Barton Spring	20050330	0730	< .005	F
08155395	Upper Barton Spring	20050420	0700	< .005	F
08155395	Upper Barton Spring	20050511	0730	< .005	F
08155395	Upper Barton Spring	20050530	0700	< .005	F
08155395	Upper Barton Spring	20050530	1430	< .005	F
08155395	Upper Barton Spring	20050530	2000	< .005	F
08155395	Upper Barton Spring	20050531	1130	< .005	F
08155395	Upper Barton Spring	20050601	0630	< .005	F
08155395	Upper Barton Spring	20050602	0700	< .005	F
08155395	Upper Barton Spring	20050604	0800	< .005	F

08155395	Upper Barton Spring	20050606	0700	< .005	F
08155395	Upper Barton Spring	20050609	0730	< .005	F
08155503	Old Mill Spring	20010503	2240	< .005	F
08155503	Old Mill Spring	20010507	1715	< .005	F
08155503	Old Mill Spring	20010508	2005	< .005	F
08155503	Old Mill Spring	20010513	2010	< .005	F
08155503	Old Mill Spring	20030806	1100	< .005	F
08155503	Old Mill Spring	20030820	1030	< .005	F
08155503	Old Mill Spring	20030903	0900	< .005	F
08155503	Old Mill Spring	20030916	0800	< .005	F
08155503	Old Mill Spring	20030930	0830	< .005	F
08155503	Old Mill Spring	20040825	0900	< .005	F
08155503	Old Mill Spring	20041214	1530	< .005	F
08155503	Old Mill Spring	20050309	0830	< .005	F
08155501	Eliza Spring	20000502	1855	.00509	F
08155501	Eliza Spring	20010504	0005	< .005	F
08155501	Eliza Spring	20010507	1720	< .005	F
08155501	Eliza Spring	20010508	1930	< .005	F
08155501	Eliza Spring	20010508	1935	< .005	F
08155501	Eliza Spring	20010510	1450	< .005	F
08155501	Eliza Spring	20010513	1900	E .00239	F
08155501	Eliza Spring	20030806	1315	< .005	F
08155501	Eliza Spring	20030820	1100	< .005	F
08155501	Eliza Spring	20030903	1000	< .005	F
08155501	Eliza Spring	20030916	0830	< .005	F
08155501	Eliza Spring	20030930	0900	< .005	F
08155501	Eliza Spring	20040825	1030	< .005	F
08155501	Eliza Spring	20041214	1630	< .005	F
08155501	Eliza Spring	20050309	0900	< .005	F
* E=estimated **U=unfiltered, F=filtered					

Table C.2. USGS monitoring data for creeks in and near action area.

pk_siteID	siteNickname	sample Date	sampleTime	Diazinon Conc (ppb)*	U/F**
08155200	Barton 71	19780607	1100	0	U
08155200	Barton 71	19780905	1230	0	U
08155200	Barton 71	19780927	0850	0	U
08155200	Barton 71	19781106	1235	0	U
08155200	Barton 71	19790227	1325	0	U
08155200	Barton 71	19790425	1252	0	U
08155200	Barton 71	19790911	1145	0	U
08155200	Barton 71	19800116	1315	0	U
08155200	Barton 71	19810408	0852	0	U
08155200	Barton 71	19810819	1000	0	U
08155200	Barton 71	19930125	1001	< .01	U

08155200	Barton 71	19931130	0940	< .01	U
08155200	Barton 71	19940222	1205	< .01	U
08155200	Barton 71	19941216	0200	< .02	U
08155200	Barton 71	19950529	0718	.04	U
08155200	Barton 71	19950607	1237	< .01	U
08155200	**Barton 71 *** **	20020630	0505	.0099	F
08155200	**Barton 71 *** **	20020716	0855	E .0037	F
08155200	**Barton 71 *** **	20021019	1019	< .005	F
08155200	**Barton 71 *** **	20021209	0510	< .005	F
08155200	Barton 71	20030909	0900	< .005	F
08155200	Barton 71	20040229	1105	< .01	F
08155200	**Barton 71 *** **	20040406	1525	.0059	F
08155200	**Barton 71 *** **	20041023	0225	< .005	F
08155240	Barton, Lost Ck.	19930125	1150	< .01	U
08155240	Barton, Lost Ck.	19931130	1149	< .01	U
08155240	Barton, Lost Ck.	19940222	1357	< .01	U
08155240	Barton, Lost Ck.	19941228	1434	.04	U
08155240	Barton, Lost Ck.	19950529	0600	.03	U
08155300	Barton 360	19790110	2330	0	U
08155300	Barton 360	19790110	2340	0	U
08155300	Barton 360	19790111	1220	0	U
08155300	Barton 360	19790321	0800	.1	U
08155300	Barton 360	19790321	0930	0	U
08155300	Barton 360	19790322	1325	.01	U
08155300	Barton 360	19790521	2030	.26	U
08155300	Barton 360	19790612	0930	0	U
08155300	Barton 360	19800415	1050	0	U
08155300	Barton 360	19801016	1240	.01	U
08155300	Barton 360	19801017	0810	0	U
08155300	Barton 360	19810408	0942	0	U
08155400	Barton Above	20000502	0135	.179	F
08155400	Barton Above	20010503	2315	< .005	F
08155400	Barton Above	20010506	2245	.104	F
08155400	Barton Above	20010507	1700	.0546	F
08155400	Barton Above	20010507	1702	< .005	F
08155400	Barton Above	20010508	1940	.0126	F
08155400	Barton Above	20010510	1505	E .00215	F
08155400	**Barton Above *** **	20020630	0455	.0106	F
08155400	**Barton Above *** **	20021019	1210	.0342	F
08155400	Barton Above	20021209	0315	E .0075	F
08155400	**Barton Above *** **	20040117	0615	< .005	F
08155400	**Barton Above *** **	20040407	0115	< .005	F
08155400	**Barton Above *** **	20041023	0520	.0225	F

08155505	Barton Below	19750115	0945	0	U
08155505	Barton Below	19750421	1300	0	U
08155505	Barton Below	19750523	2200	.02	U
08155505	Barton Below	19750922	1250	0	U
08155505	Barton Below	19760106	1005	0	U
08155505	Barton Below	19760419	1015	0	U
08155505	Barton Below	19760622	0940	0	U
08155505	Barton Below	19761103	1403	0	U
08155505	Barton Below	19770415	1330	.01	U
08155505	Barton Below	19770518	1250	0	U
08155505	Barton Below	19770922	1045	0	U
08155505	Barton Below	19780608	1600	.05	U
08155505	Barton Below	19780808	1600	0	U
08155505	Barton Below	19780927	1200	0	U
08155505	Barton Below	19790425	1045	0	U
08155505	Barton Below	19790919	1130	0	U
08155505	Barton Below	19800116	1045	0	U
08155505	Barton Below	19810408	1240	0	U
08155505	Barton Below	19810824	1300	0	U
08158920	Williamson at Oak Hill	19780607	1230	.05	U
08158920	Williamson at Oak Hill	19781106	1130	0	U
08158920	Williamson at Oak Hill	19790424	1155	0	U
08158920	Williamson at Oak Hill	19790522	1045	.09	U
08158920	Williamson at Oak Hill	19790612	0855	0	U
08158920	Williamson at Oak Hill	19790911	1240	0	U
08158920	Williamson at Oak Hill	19800425	1045	.19	U
08158930	Williamson Manchaca	20000501	0400	.26	F
08158930	**Williamson Manchaca *****	20020319	2115	.158	F
08158930	**Williamson Manchaca *****	20020616	0455	.0469	F
08158930	**Williamson Manchaca *****	20021008	1250	.0285	F
08158930	**Williamson Manchaca *****	20030220	0415	.0542	F
08158930	Williamson Manchaca	20031117	1600	.0151	F
08158930	Williamson Manchaca	20040429	0240	.0321	F
08158930	**Williamson Manchaca *****	20041023	0750	.0121	F
08158930	**Williamson Manchaca *****	20050529	2105	.0316	F
08158970	Williamson Jimmy Clay	19750116	1400	.01	U
08158970	Williamson Jimmy Clay	19750422	0900	0	U
08158970	Williamson Jimmy Clay	19750523	2030	.14	U
08158970	Williamson Jimmy Clay	19750923	1530	.03	U
08158970	Williamson Jimmy Clay	19760106	1220	0	U
08158970	Williamson Jimmy Clay	19760614	1000	0	U
08158970	Williamson Jimmy Clay	19760903	1345	.11	U
08158970	Williamson Jimmy Clay	19761102	1325	0	U
08158970	Williamson Jimmy Clay	19770415	1530	.13	U
08158970	Williamson Jimmy Clay	19770516	1300	0	U

08158970	Williamson Jimmy Clay	19770920	1005	.47	U
08158970	Williamson Jimmy Clay	19780111	1120	.01	U
08158970	Williamson Jimmy Clay	19780607	1400	.41	U
08158970	Williamson Jimmy Clay	19780725	1325	.01	U
08158970	Williamson Jimmy Clay	19780925	1342	.01	U
08158970	Williamson Jimmy Clay	19781106	0815	.2	U
08158970	Williamson Jimmy Clay	19790424	1120	.15	U
08158970	Williamson Jimmy Clay	19790911	0720	0	U
08158970	Williamson Jimmy Clay	19800114	1335	0	U
08158970	Williamson Jimmy Clay	19810819	1245	.55	U
08158860	Slaughter at 2304	19790111	1445	0	U
08158860	Slaughter at 2304	19790112	1330	0	U
08158860	Slaughter at 2304	19800513	1030	.12	U
08158860	Slaughter at 2304	19810304	0743	.01	U
08158860	**Slaughter at 2304 *****	20041023	0200	< .01	F
08158860	**Slaughter at 2304 *****	20050529	2055	.0139	F
08158860	**Slaughter at 2304 *****	20050530	0255	.0333	F
08158810	Bear at 1826	19780607	1630	0	U
08158810	Bear at 1826	19780927	1000	0	U
08158810	Bear at 1826	19781106	1310	.03	U
08158810	Bear at 1826	19790112	1415	0	U
08158810	Bear at 1826	19790223	1215	0	U
08158810	Bear at 1826	19790425	1145	0	U
08158810	Bear at 1826	19800116	1220	0	U
08158810	Bear at 1826	19810819	0920	0	U
08158810	Bear at 1826	19930113	1044	< .01	U
08158819	**Bear nr Brodie *****	20041023	0200	< .005	F
08158825	Little Bear 1626	19781106	0900	.16	U
08158825	Little Bear 1626	19790111	1645	0	U
08158825	Little Bear 1626	19800425	0940	.27	U
08158700	Onion at Driftwood	19780112	1005	0	U
08158700	Onion at Driftwood	19780607	1520	0	U
08158700	Onion at Driftwood	19780926	1230	0	U
08158700	Onion at Driftwood	19781106	1350	0	U
08158700	Onion at Driftwood	19790227	1250	0	U
08158700	Onion at Driftwood	19790613	1350	0	U
08158700	Onion at Driftwood	19790911	1030	0	U
08158700	Onion at Driftwood	19800115	1410	0	U
08158700	Onion at Driftwood	19800930	1210	0	U
08158700	Onion at Driftwood	19810818	1215	0	U
08158700	Onion at Driftwood	20030909	1200	< .005	F
08158700	Onion at Driftwood	20040721	1200	< .005	F

08158700	**Onion at Driftwood *****	20041023	1130	< .005	F
08158700	Onion at Driftwood	20041110	0800	< .005	F
08158700	Onion at Driftwood	20050311	1230	< .005	F
08158800	Onion at Buda	19780607	1400	.1	U
08158800	Onion at Buda	19780926	1121	0	U
08158800	Onion at Buda	19781106	0945	0	U
08158800	Onion at Buda	19790227	1100	0	U
08158800	Onion at Buda	19790320	0930	0	U
08158800	Onion at Buda	19790613	1205	0	U
08158800	Onion at Buda	19800117	1315	0	U
08158827	**Onion at Twin Cks *****	20041023	1000	< .005	F
08158827	Onion at Twin Cks	20041026	1100	< .005	F
08158827	**Onion at Twin Cks *****	20050529	2140	< .005	F

* E=estimated
**U=unfiltered, F=filtered
*** Flow weighted storm composite samples

Table C.3. USGS monitoring data for groundwater wells in and near action area.

pk_siteID	siteNickname	sampleDate (year, month, date)	sampleTime	Diazinon Conc. (ppb)	U/F
300453097503301	LR-58-58-403 (BPS)	19770504	1100	0	U
300453097503301	LR-58-58-403 (BPS)	19780724	1010	0	U
300453097503301	LR-58-58-403 (BPS)	19810812	0810	0	U
300453097503301	LR-58-58-403 (BPS)	19930819	1220	< .01	U
300453097503301	LR-58-58-403 (BPS)	20010612	1100	< .005	F
300453097503301	LR-58-58-403 (BPS)	20020606	1100	< .005	F
300453097503301	LR-58-58-403 (BPS)	20030522	1100	< .005	F
300453097503301	LR-58-58-403 (BPS)	20040716	1330	< .005	F
300453097503301	LR-58-58-403 (BPS)	20050524	1330	< .005	F
300646097533202	LR-58-57-311 (BDW)	20010605	1300	< .005	F
300646097533202	LR-58-57-311 (BDW)	20020605	1300	< .005	F
300646097533202	LR-58-57-311 (BDW)	20030520	1300	< .005	F
300646097533202	LR-58-57-311 (BDW)	20040713	1100	< .005	F
300646097533202	LR-58-57-311 (BDW)	20050524	1020	< .005	F
300813097512101	YD-58-50-704 (MCH)	20010620	1100	< .005	F
300813097512101	YD-58-50-704 (MCH)	20020604	1100	< .005	F
300813097512101	YD-58-50-704 (MCH)	20030520	1200	< .005	F
300813097512101	YD-58-50-704 (MCH)	20040712	1140	< .005	F
300813097512101	YD-58-50-704 (MCH)	20050525	1353	< .005	F
301031097515801	YD-58-50-408 (FOW)	20010619	1000	E .0017	F
301031097515801	YD-58-50-408 (FOW)	20020605	1000	< .005	F
301031097515801	YD-58-50-408 (FOW)	20030521	1000	< .005	F
301031097515801	YD-58-50-408 (FOW)	20040709	1145	< .005	F
301031097515801	YD-58-50-408 (FOW)	20050526	1122	< .005	F

301142097504701	YD-58-50-417 (FON)	20010622	1100	< .005	F
301142097504701	YD-58-50-417 (FON)	20020604	1400	< .005	F
301142097504701	YD-58-50-417 (FON)	20030728	1000	< .005	F
301142097504701	YD-58-50-417 (FON)	20040708	1430	< .005	F
301142097504701	YD-58-50-417 (FON)	20050526	1249	< .005	F
301226097480701	YD-58-50-520 (PLS)	20010608	1100	< .005	F
301226097480701	YD-58-50-520 (PLS)	20020523	1100	< .005	F
301226097480701	YD-58-50-520 (PLS)	20030521	1200	< .005	F
301226097480701	YD-58-50-520 (PLS)	20040721	1105	< .005	F
301226097480701	YD-58-50-520 (PLS)	20050527	1221	< .005	F
301339097483701	YD-58-50-215 (SVS)	19780808	0750	.04	U
301339097483701	YD-58-50-215 (SVS)	19810810	1407	0	U
301339097483701	YD-58-50-215 (SVS)	20010618	1200	E .0017	F
301339097483701	YD-58-50-215 (SVS)	20020606	1300	< .005	F
301339097483701	YD-58-50-215 (SVS)	20030519	1300	< .005	F
301339097483701	YD-58-50-215 (SVS)	20040716	1050	< .005	F
301339097483701	YD-58-50-215 (SVS)	20050525	1042	< .005	F
301356097473301	YD-58-50-216 (SVE)	20010614	1200	< .005	F
301356097473301	YD-58-50-216 (SVE)	20020807	1200	< .005	F
301356097473301	YD-58-50-216 (SVE)	20030528	1200	< .005	F
301356097473301	YD-58-50-216 (SVE)	20040715	1525	< .005	F
301356097473301	YD-58-50-216 (SVE)	20050615	1145	< .005	F
301423097495901	YD-58-50-211 (SVW)	19780627	1220	0	U
301423097495901	YD-58-50-211 (SVW)	19810810	1340	0	U
301423097495901	YD-58-50-211 (SVW)	20010606	1200	< .005	F
301423097495901	YD-58-50-211 (SVW)	20020603	1400	< .005	F
301423097495901	YD-58-50-211 (SVW)	20030519	1000	< .005	F
301423097495901	YD-58-50-211 (SVW)	20040708	1113	< .005	F
301423097495901	YD-58-50-211 (SVW)	20050523	1207	< .01	F
301432097480001	YD-58-50-217 (SVN)	20010615	1100	< .005	F
301432097480001	YD-58-50-217 (SVN)	20020807	1000	< .005	F
301432097480001	YD-58-50-217 (SVN)	20030528	1000	< .005	F
301432097480001	YD-58-50-217 (SVN)	20040715	1120	< .005	F
301432097480001	YD-58-50-217 (SVN)	20050614	0950	< .005	F
301526097463201	YD-58-42-915 (RAB)	20010607	1600	< .005	F
301526097463201	YD-58-42-915 (RAB)	20020603	1100	< .005	F
301526097463201	YD-58-42-915 (RAB)	20030530	1000	< .005	F
301526097463201	YD-58-42-915 (RAB)	20040707	1355	< .005	F
301526097463201	YD-58-42-915 (RAB)	20050523	1424	< .005	F
302146097445101	YD-58-43-103	20010619	1300	< .005	F
302218097454901	YD-58-42-311	20020522	1100	< .005	F
302218097454901	YD-58-42-311	20030516	1000	< .005	F
302218097454901	YD-58-42-311	20040707	1020	< .005	F
302218097454901	YD-58-42-311	20050613	1115	< .005	F
302316097430401	YD-58-35-701	20010604	1000	< .005	F
302551097465501	YD-58-34-617	20010621	1200	< .005	F
302551097465501	YD-58-34-617	20020516	1130	< .005	F
302551097465501	YD-58-34-617	20030515	1100	< .005	F

302551097465501	YD-58-34-617	20040706	1030	< .005	F
302551097465501	YD-58-34-617	20050616	1000	< .005	F
302554097494701	YD-58-34-414	20010621	1000	< .005	F
302554097494701	YD-58-34-414	20020520	1015	< .005	F
302554097494701	YD-58-34-414	20030513	1100	< .005	F
302554097494701	YD-58-34-414	20040706	1445	< .005	F
302554097494701	YD-58-34-414	20050527	1000	< .005	F
302652097430501	YD-58-35-415	19780621	1130	.02	U
302652097430501	YD-58-35-415	19810804	1330	0	U
300356097563801	LR-58-57-502	19780712	1300	.01	U
300356097563801	LR-58-57-502	19810818	1110	0	U
302148097422801	YD-58-43-206	19780719	0940	.02	U
302148097422801	YD-58-43-206	19810810	1050	0	U
300639097571001	LR-58-57-202	19810812	0905	0	U
300803097483801	YD-58-50-810	19780710	0940	0	U
300803097483801	YD-58-50-810	19810811	1305	0	U
300934097552201	LR-58-49-801	19780711	0810	0	U
300934097552201	LR-58-49-801	19810819	0850	0	U
301604097465601	YD-58-42-913	19780626	1310	0	U
301811097470401	YD-58-42-608	19780719	1150	0	U
301811097470401	YD-58-42-608	19810805	1415	0	U

* E=estimated
**U=unfiltered, F=filtered

Appendix D. Status and Life History of the Barton Springs Salamander.

D.1 Species Listing Status

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) based on the following threats:

(1) degradation of the water quality in Barton Springs as a result of urban expansion,
(2) decreased quantity of water that feeds Barton Springs as a result of urban expansion,
(3) modification of the salamander's structural habitat,
(4) inadequacy of existing regulatory mechanisms to protect the salamander and lack of a comprehensive plan to protect the Barton Springs watershed from increasing threats to water quality and quantity, and
(5) the salamander's extreme vulnerability to environmental degradation because of its restricted range in an entirely aquatic environment.

USFWS is the branch of the Department of Interior responsible for listing endangered amphibians, such as the Barton Springs salamander. The extent to which any these threats is considered to predominate is unknown and presumably their cumulative effect may be of primary concern.

D.2 Description and Taxonomy

The Barton Springs salamander (Figure D.1) is a member of the Family Plethodontidae (lungless salamanders). Texas species within the genus *Eurycea* inhabit springs, spring-runs, and water-bearing karst formations of the Edwards Aquifer (Chippindale, 1993). These salamanders are aquatic and neotenic, meaning they retain a larval, gill-breathing morphology throughout their lives. Neotenic salamanders, including the Barton Springs salamander, do not metamorphose into a terrestrial form. Rather, they live their entire life cycle in water, where they become sexually mature and eventually reproduce.

Figure D.1. Barton Springs Salamander
(courtesy of Lisa O'Donnell; City of Austin Watershed Protection and Development Review Department)

The Barton Springs salamander was first collected from Barton Springs in 1946 (Brown, 1950; Texas Natural History Collection specimens 6317-6321). Adults grow to approximately 2.5 to 3 inches (63-76 mm) in total length. Adult body morphology includes reduced eyes and elongate, spindly limbs indicative of a semi-subterranean lifestyle. The head is relatively broad and deep in lateral view, and the snout appears somewhat truncate when viewed from above. Three bright red, feathery gills are present on either side of the base of the head. The coloration on the salamander's upper body varies from light to dark brown, purple, reddish brown, yellowish cream, or orange. The characteristic mottled salt-and-pepper color pattern on the upper body surface is due to brown or black melanophores (cells containing pigments called melanin) and silvery-white iridiophores (cells containing pigments containing guanine). The arrangement of these pigment cells is highly variable and can be widely dispersed in some Barton Springs salamanders, causing them to have an overall pale appearance. In other individuals, the melanophores may be dense, resulting in a dark brown appearance. The ventral side (underside) of the body is cream-colored and translucent, allowing some internal organs and developing eggs in females to be visible. The tail is relatively short with a well-developed dorsal (upper) fin and poorly developed ventral (lower) fin. The upper and lower mid-lines of the tail usually exhibit some degree of orange-yellow pigmentation. Juveniles closely resemble adults (Chippindale et al., 1993). Newly hatched larvae are about 0.5 inches (12 mm) in total length and may lack fully developed limbs or pigment (Chamberlain and O'Donnell, 2003).

D.3 Population Status and Distribution

The Barton Spring salamander has been found only at the four spring outlets that make up Barton Springs complex (Figure D.2). This species is considered to have one of the smallest geographical ranges of any vertebrate species in North America (Chippindale et al., 1993; Conant and Collins, 1998).

The salamander was first observed in Barton Springs Pool and Eliza Springs in the 1940s, Sunken Garden Springs in 1993 (Chippindale et al., 1993), and the intermittent Upper Barton Springs in 1997 (City of Austin, 1998).

The extent of the Barton Spring salamander's range within the Barton Springs Segment of the Edwards Aquifer, and the degree of subsurface connection among these spring populations is unknown. However, observations of salamanders actively swimming into high flow areas from the spring openings, including Main Springs in Barton Springs Pool (USFWS, 2005), and the discovery of a more cave-adapted species (Austin blind salamander, *Eurycea waterlooensis*), suggest that the Barton Springs salamander is not entirely subterranean (triglobotic). The Barton Springs salamander appears to reproduce primarily in subterranean areas (*i.e.,* within the aquifer). Although salamander larvae are present in surface water year-round, very few eggs have been observed on the surface (Chamberlain and O'Donnell, 2003).

D.3.1 Survey Results

The City of Austin initiated salamander surveys in (1) Barton Springs Pool in 1993, (2) Old Mill Springs and Eliza Springs in 1995, and (3) Upper Barton Springs in 1997 (City of Austin, 1998, City of Austin, 1993-2003, unpublished data). Due to the inaccessibility of the aquifer and spring orifices, survey counts reflect the number of individuals observed in the spring pools and spring runs rather than total population census estimates (City of Austin, 2005a). Survey methods have varied to some degree, mainly in Barton Springs Pool, where the survey area gradually shifted from transects to the immediate area around the spring outlets where salamanders are most abundant (USFWS, 2005).

The results of the adult and juvenile salamander survey data are depicted in Figures D.3 and D.4, respectively. From 1997 to 2005 (years in which there are survey data for all four springs), the mean number of adult salamanders observed per year at all four springs combined ranged between 5 and 80. Further examination of the data shows a marked increase in the number of observed adults and juveniles in Eliza Spring, relative to the other springs, from mid-2003 to 2005. From 1997 until 2003, the largest mean number of adult and juvenile salamanders (15 and 14, respectively) were observed in Barton Springs Pool, followed by Old Mill Spring (13 and 8, respectively). However, in 2004 and 2005, the largest average number of adult and juvenile salamanders were observed in Eliza Springs (252 and 91, respectively), followed by Barton Springs Pool (35 and 21, respectively).

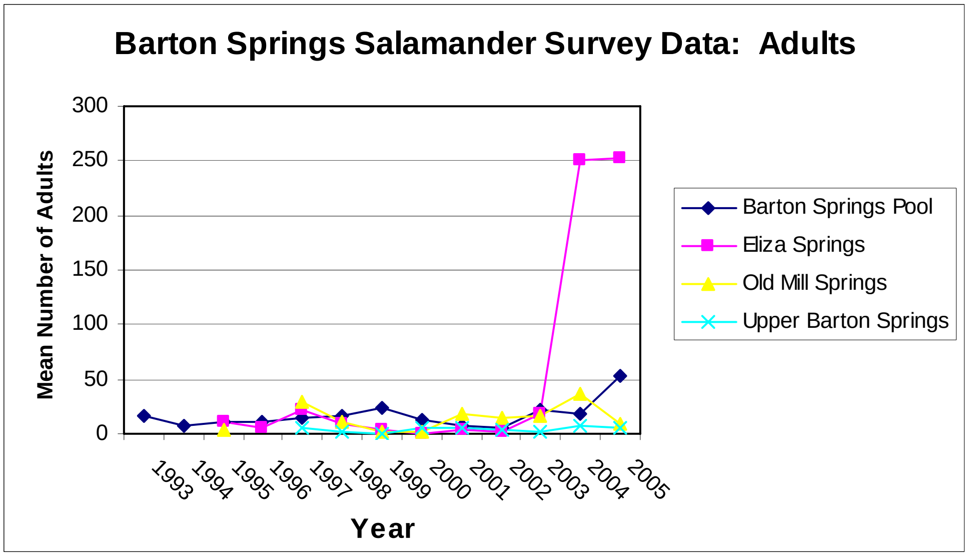

Figure D.3. Barton Springs Salamander Survey Data: Adults

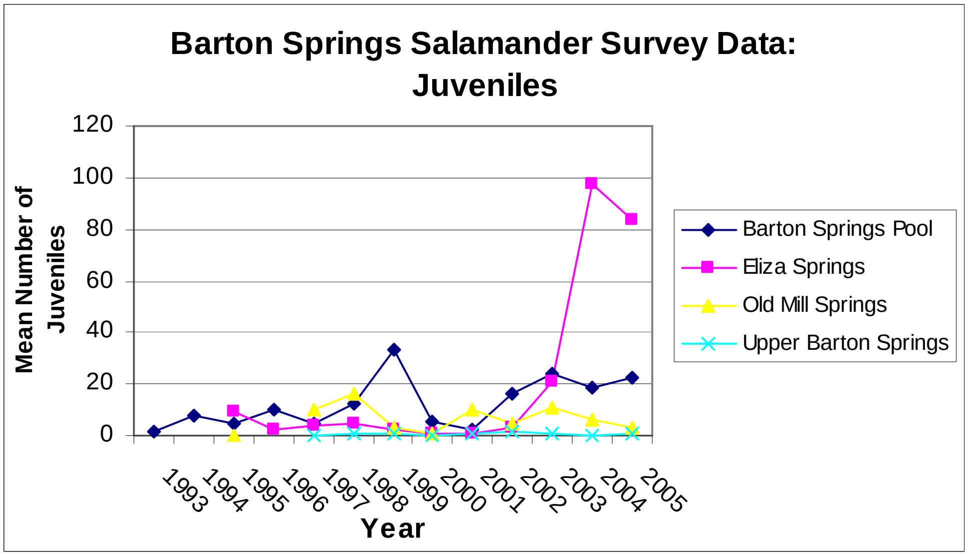

Figure D.4. Barton Springs Salamander Survey Data: Juveniles

Increased numbers of observed adult and juvenile salamanders in Eliza Springs from 2003 to 2005 are believed to be due to habitat restoration efforts, initiated in Eliza Springs by the City of Austin biologists in the fall of 2002 (City of Austin, 2003). Following habitat restoration, observed numbers of salamanders began to increase in July 2003. The habitat restoration efforts at Eliza Springs included removal of debris from the drainage infrastructure to increase flow across the bottom of the spring pool and allow for more natural flushing and draining of the spring ecosystem. Removal of fine sediment exposed a layer of gravel and cobble that had previously been obscured, making it available as habitat for the salamanders. Several species of native aquatic plants, including water primrose (*Ludwegia* sp.), rush (*Eleocharis* sp.), and water hyssop (*Bacopa* sp.) were also successfully transplanted from Barton Creek into Eliza Springs to serve as cover and promote invertebrate prey species. In addition, mosquitofish and crayfish, predators to the salamander, were removed from Eliza Springs. The net impact of the restoration efforts at Eliza Springs was the following: (1) to increase lateral water flow across the spring pool, thus reducing the amount of sediment and increasing the amount of loose rock substrate (habitat) available for the salamander and its forage base; and (2) to decrease the number of predators and other species that compete for available food. As a result of these efforts, mean numbers of adults and juveniles collected from Eliza Springs during 2004 increased by approximately 13-fold and 5-fold, respectively, as compared to total numbers collected during 2003. With the exception of an increase in the number of juvenile salamanders in Eliza Spring over the past two years, there does not appear to be any clear pattern in the number of young salamanders recorded by year or month over the past decade of survey results.

The majority of salamanders in Barton Springs Pool are found primarily in the immediate area of the spring outlets (USFWS, 2005). They have also been found to a lesser extent in the "beach" area, which includes an underwater concrete bench immediately adjacent to a pedestrian sidewalk on the north side of Barton Springs Pool. Salamanders are rarely seen in the deep end of the pool, which is often covered by sediment, or in the shallow end, which is almost entirely limestone and/or concrete, and thus not considered suitable habitat. Based on observations of salamanders in water depths ranging from <1 inch to >15 feet, it appears that water depth is not a determining factor in habitat selection. Although Barton Springs salamanders do not appear to have an obvious depth preference, constant water flow, stable temperatures, and rock substrates free of sediment are needed for suitable habitat. The survey area in Barton Springs Pool has gradually shifted from transects that included the beach and the deep end, to the intermediate area around the spring outlets where salamanders appear to be most abundant. Based on the comprehensive surveys conducted by the City of Austin and the Service, the number of estimated salamanders inhabiting the surface habitat in Barton Springs Pool may be negatively biased, with actual expected numbers of individuals that are three to five times greater than the number of individuals counted during the regular monthly surveys (City of Austin, 1998).

The Barton Springs Salamander Recovery Plan (USFWS, 2005) notes that numbers of salamanders at Old Mill Springs appear to be related to flow patterns and the presence of predatory fish. For example, a decrease in salamander numbers observed during the winter of 2002-2003 may have been due to the presence of Mexican tetras (*Asyanax mexicanus*), a non-native predatory fish (City of Austin, 2003). Review of the survey data also indicates a drop in numbers in Old Mill Springs in 2000, which is believed to be due to reduced water flow within the spring. According the City of Austin (2003), flow was extremely low in 2000; in fact, much of Old Mill Springs was dry in the spring/summer of 2000.

In 1997, biologists from the City of Austin and the USFWS discovered 14 adult salamanders at Upper Barton Springs, which flows intermittently. The number of salamanders found at this site in subsequent surveys has ranged from 0 to 14 (City of Austin, unpublished data). Given that salamanders are absent when this spring is dry, survey data indicate that salamander numbers are directly affected by surface flow. However, some monthly surveys at Upper Barton Springs have not found salamanders, even during periods when the spring was flowing (USFWS, 2005).

D.4 Habitat

All available information indicates that the Barton Springs salamander is restricted to the immediate vicinity of the four spring outlets of Barton Springs. Because 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, the salamander may be affected by changes in water quality and quantity occurring in the Barton Springs watershed[2].

"Surface" habitat for the Barton Springs salamander refers to the spring pools and spring runs where the salamander is observed, as opposed to its potential subsurface aquifer habitat. The Barton Springs salamander experiences relatively stable aquatic environmental conditions. These conditions consist of perennially flowing spring water that is generally clear, has a neutral pH (~7), and cool average annual temperatures of 21 to 22 ºC (~70-72 ºF) (USFWS, 2005). As is typical of groundwater dominated systems, the springs exhibit a narrow temperature range (stenothermal). Flows of clean spring water with a relatively constant, cool temperature are essential to maintaining well-oxygenated water necessary for salamander respiration and survival (USFWS, 2005). Dissolved oxygen (DO) concentrations in Barton Springs average approximately 6 mg/L (USFWS, 2005) and are directly related to springflow. Higher DO concentrations occur during periods of high spring discharge (USFWS, 2005).

The subterranean component of the Barton Springs salamander's habitat may provide a location for reproduction, serve as refugium during high flow events or high sediment loads from surface sources in the surface habitat, and/or provide a migration pathway between the surface habitat areas (USFWS, 2005).

Based on the survey results, Barton Springs salamanders appear to prefer clean, loose substrate for cover. They are found primarily under boulder, cobble, and gravel substrates, but may also be found in the vicinity of aquatic plants, leaf litter, and woody debris (USFWS, 2005). In the main pool, City of Austin surveys indicate that salamanders are found primarily near the spring outlets. To a lesser extent, Barton Springs salamanders are also found in aquatic moss (*Amblystegium riparium*) that grows on bare rocks and on the walls surrounding Barton Springs Pool, Eliza Springs, and Old Mill Springs (City of Austin, 2003).

Historical records indicate a diversity of macrophytes once resided in Barton Springs Pool, including arrowhead (*Sagittaria platyphylla*), water primrose (*Ludwigia* spp.), wild celery (*Vallisneria americana*), cabomba (*Cabomba caroliniana*), water stargrass (*Heteranthera sp.*), southern naiad (*Najas guadalupensis*), and pondweed (*Potamogeton* sp.) (Alan Plummer Associates Inc., 2000 in USFWS, 2005). In 1992, the dominant aquatic plant in the pool was the moss (*A. riparium*), an aquatic bryophyte ubiquitous in Central Texas springs. In addition to providing cover, moss and other aquatic plants harbor a variety and abundance of the aquatic invertebrates that salamanders eat.

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[2] The "Barton Springs watershed" includes the contributing zone and recharge zone of the Barton Springs segment of Edwards Aquifer.

During the 1980s and 1990s, the majority of aquatic macrophytes disappeared from the Barton Springs Pool (USFWS, 2005), leaving primarily unvegetated limestone substrate and sediment as habitat. The disappearance of the aquatic macrophytes in the deep end of the pool appears to have resulted from the combined effects of flooding, dredging, and the mechanical dragging of the deep end with chains for sediment removal (USFWS, 2005). However, it is unclear how these activities and the related disappearance of aquatic macrophytes in Barton Springs Pool may have affected the salamander numbers because they pre-dated the survey efforts, which were initiated in 1993.

In addition to restoration efforts for Eliza Springs (previously discussed in Section D.3.1), efforts to reintroduce endemic plant species in Barton Springs Pool were initiated by the City of Austin in 1993. At that time, aquatic vegetation in Barton Springs Pool was limited to two small patches of *Potamogeton*, one patch of *Sagittaria* in the far deep end of the pool, and areas of *Amblystegium* near the discharge points. *Sagittaria*, *Ludwigia*, and *Cabomba* have been introduced into Barton Springs Pool in June 1993 and again in the fall of 1994. It is not possible to gauge the effect of these activities on salamander numbers because there were no historical survey data. Aquatic macrophytes currently found in Barton Springs Pool are limited to *Sagittaria*. *Amblystegium* is also common on limestone surfaces in the general vicinity of the main springs and various side springs.

Salamanders are most frequently found around the main spring outflows, hidden within a 2-8 cm (0.8 – 3.1 inches) deep zone of gravel and small rocks overlying a coarse sandy or bare limestone substrate (USFWS, 2005). These areas are visibly clear of fine silt or decomposed organic debris and appear to be kept clean by flowing spring water during medium to high aquifer levels. Abundant prey species for the salamander also inhabit these areas. Piles of woody debris in the vicinity of the main springs provide habitat for the salamander, as well as its prey base, after floods, when normal habitat may be covered with sediment. Suitable habitat can increase or decrease depending on a number of factors including springflows, abundance of aquatic macrophytes, sedimentation rates, and frequency of floods.

In addition, pool cleanings may affect the salamander and its habitat. During the cleanings, full drawdowns of the pool (removal of 4-5 feet of water) are limited to four times/year, when spring discharge exceeds 53 cfs (cubic feet/second) and Barton Creek floods. For the past two years, the water level has been partially lowered (by 18-24") once per month when the flow exceeds 53 cfs. During this time, biologists clean sediment and debris from salamander habitat with garden hoses. Salamander habitat in Barton Springs Pool that is exposed during full drawdowns includes the area of fissures on the bedrock above the main spring outlets. The main spring outlets, which are located 10-16 feet below the top of the bedrock fissures, are not exposed during drawdowns as spring water continues to flow.

When discharge from Barton Springs Pool is lower than 54 cfs, the water level in Eliza Springs has the potential to drop below the surface substrate during a full drawdown. This is partially due to the presence of a concrete slab at the bottom of Eliza Springs, beneath the gravel and cobble. Flowing spring water into Eliza Springs must have adequate pressure to discharge through holes in the concrete bottom. When discharge is low and Barton Springs Pool is drawn down, the water level in Eliza Springs drops to below the surface substrate and salamanders are stranded at the surface. The habitat beneath this concrete slab is dark and sediment laden, and thus considered as poor habitat. In general, the water level in Old Mill Springs does not drop below the surface substrate when the Pool is drawn down, unless there is very low discharge from the aquifer.

D.5 Life History and Ecology

Information on the life history and ecology of the Barton Springs salamander, including diet, respiration, reproduction, longevity, diseases, and predators is provided in Sections D.5.1 through D.5.6.

D.5.1 Diet

Barton Springs salamanders appear to be opportunistic predators of small, live aquatic invertebrates (USFWS, 2005). Chippindale et al. (1993) found amphipod remains in the stomachs of wild-caught salamanders. The gastro-intestinal tracts of 18 adult and juvenile Barton Springs salamanders and fecal pellets from 11 adult salamanders collected from Eliza Springs, Barton Springs Pool, and Sunken Garden Springs contained ostracods, copepods, chironomids, snails, amphipods, mayfly larvae, leeches, and adult riffle beetles. The most prevalent organisms found in these samples were ostracods, amphipods, and chironomids (USFWS, 2005). The types of invertebrates found in the pools at Barton Springs are documented in the City of Austin's Habitat Conservation Plan (1998).

D.5.2 Respiration

Primary respiration in neotenic salamanders is through the gills; however, a substantial amount of gas exchange occurs through the skin (Boutilier et al. 1992; Hillman and Withers 1979). They require moving water across their gills and bodies for respiration. Metabolic rates and oxygen consumption are highest in juveniles and decrease with increasing body size (Norris et al., 1963). Oxygenation of salamander eggs is critical to embryonic development since gas exchange and waste elimination occur through semipermeable membranes surrounding the embryo (Duellman and Trueb 1986).

D.5.3 Reproduction

Little is known about the reproductive biology of the Barton Springs salamander in the wild. The ability to view Barton Springs salamanders in their natural environment is limited because of the animal's propensity to inhabit interstitial spaces under rocks and subterranean environments. Therefore, information regarding the reproductive biology of the Barton Springs salamander is based primarily on captive breeding populations maintained by the City of Austin, and extrapolations from closely related species. Although some aspects of the reproductive biology may be affected by the artificial environment in which they are maintained, information collected on the captive breeding population represents the best available information. When field data are available, the differences and similarities between the wild and captive populations are compared.

Barton Springs salamanders are not sexually dimorphic; however, gravid females can sometimes be distinguished by the presence of eggs which are visible through the translucent skin of the underside. Recent studies with captive individuals indicate that salamander eggs are 1.5 to 2.0 mm (0.06 to 0.08 inches) in diameter when they are laid. Young larvae develop and hatch in approximately 16 to 39 days (USFWS, 2005). Captive raised female salamanders have developed eggs within 11 to 17 months after hatching. One male also displayed courtship behavior (tail undulation) at one year from hatching (Chamberlain and O'Donnell, 2003). At sexual maturity, salamanders are generally at least 50 mm in total length (Chamberlain and O'Donnell, 2003). No clear pattern of reproductive activity has been recorded in the field or in the laboratory. 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). No relationship between breeding activity and environmental factors has been established to date.

The captive breeding program has observed clutch sizes ranging from 5 to 39 eggs, with an average of 22 eggs based on 32 clutches; individual captive females have produced up to 6 clutches per year (Chamberlain and O'Donnell, 2003). Of the 34 egg-laying events at the Dallas Aquarium, clutch size ranged from 10 to 55 (Lynn Ables, Dallas Aquarium, pers. comm., 2000). Females may lay all or only a few of their eggs, and in some cases, females may reabsorb their unlaid eggs within a few weeks after egg-laying (Chamberlain and O'Donnell, 2003). Currently, specific cues and/or environmental factors associated with clutch size and timing of courtship and reproduction have not been identified (USFWS, 2005).

Data regarding development and hatching of eggs are based almost exclusively on observations of the captive populations. In spite of relatively intensive survey efforts, only four eggs have been located in the wild. In four separate instances, a single egg was found near a spring orifice (USFWS, 2005). These observations combined with the visibility of the eggs to predators due to their lack of pigment (eggs are white) suggest the eggs are laid in the subterranean portion of the salamander's habitat. Eggs are laid singly and receive no parental care (USFWS, 2005). Hatching of eggs in captivity has occurred within 16 to 39 days after eggs have been laid (Chamberlain and O'Donnell, 2003). Hatching success of a clutch is variable (10 - 100%), with means ranging from 26 to 57 percent (Chamberlain and O'Donnell, 2003). Based on information summarized in USFWS (2005), egg mortality in captivity has been attributed to (1) fungus (Chamberlain and O'Donnell, 2002 and 2003), (2) hydra (small invertebrates with stinging tentacles) (Lynn Ables, Dallas Aquarium, pers. comm., 2000), and (3) other factors, including infertility (Chamberlain and O'Donnell, 2003). Environmental conditions, water quality, adequate space, habitat heterogeneity, and food availability may also influence egg laying (Chamberlain and O'Donnell, 2003).

At hatch, juveniles measure 13 mm in total length (snout to tip of tail). After 4 months, juveniles ranged in total length from 13 to 38 mm (Chamberlain and O'Donnell, 2003). Growth rates in the wild, based on a limited mark-recapture dataset of 11 Barton Springs salamanders, ranged from 0.14 to 0.50 mm per day over a 30- to 57-day period (City of Austin, unpublished data). The available data suggest that Barton Springs salamanders could potentially reach full maturity within six months from hatching, although the sample size upon which these data are based is limited and additional research is warranted.

City of Austin biologists have generally found the first three months following hatching to be a critical period for juvenile survival (Chamberlain and O'Donnell, 2003). Of the 285 eggs laid in one breeding study, only 12 (4%) survived the first three months (Chamberlain and O'Donnell, 2003). Newly hatched larvae have sufficient yolk to sustain their nutritional needs for several days after hatch. Larvae feeding on prey items have been observed 11 to 15 days after hatching (Lynn Ables, Dallas Aquarium, pers. comm., 1999).

D.5.4 Longevity

The longevity of the Barton Springs salamander in the wild is unknown; however, salamanders in captivity have survived to at least 12 years (USFWS, 2005).

D.5.5 Diseases

A limited number of physiological infections have been reported in the wild for the Barton Springs salamanders. Adult Barton Springs salamanders have been infected with trematodes (*Clinostomum* sp.) that invaded tissue near the salamander's vent (Chamberlain and O'Donnell, 2002).

D.5.6 Predators

Predation on adult Barton Springs salamanders in the wild is expected to be minimal when adequate cover is available (USFWS, 2005). Most of the potential predators native to the Barton Springs ecosystem are opportunistic feeders, and predation is unlikely unless the salamanders become exposed. Crayfish (*Procambarus clarkii*) and other large predatory invertebrates may prey on salamanders or on their larvae and eggs (Gamradt and Kats, 1996). Crayfish have been reported to be extremely abundant at times, with an apparent "crayfish bloom" occurring in the spring of 1995, when thousands of crayfish were found throughout the pool (USFWS, 2006). Predatory fish found at Barton Springs include mosquitofish (*Gambusia affiinis*), longear sunfish (*Lepomis megalotis*), and largemouth bass (*Micropterus salmoides*). Mosquitofish have been known to prey on frog and salamander larvae in areas where the fish have been introduced (Gamradt and Kats, 1996; Goodsell and Kats, 1999; Lawler et al., 1999). Longear sunfish are known to prey on aquatic vertebrates, and largemouth bass are opportunistic predators that feed primarily on smaller fishes and crayfish. Mexican tetras are non-native fish and aggressive generalist predators that are occasionally found in Barton Creek, Barton Springs Pool, Upper Barton Springs, and Sunken Garden Springs (USFWS, 2005). In addition, green-throat darters (*Etheostoma lepidum*) have been known to prey upon small juvenile salamanders when no cover is available.

D.6 References

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Appendix E. Stepwise Modeling Approach for the Barton Springs Salamander Endangered Species Assessment for Diazinon.

1. Modify the PE4v01.pl shell to indicate daily time series (TSER) instead of the standard cumulative (TCUM) output in Record 40 of przm3.inp files.

2. Remove irrigation parameters from the TX_BSSNursury and TX_BSSResidential scenarios by setting the IRFLAG input in Record 20 to "0".

3. Use the modified PE4 shell to run the TX_BSSNursery scenario with the maximum ornamental use pattern, the TX_BSSOrchard scenario with the maximum peach use pattern, and the TX_BSSResidential scenario with any use pattern.

4. Open the *.zts files with Microsoft Office Excel, fixing each column width to capture the appropriate data (allow eight character spaces beyond the decimal). Save the result as a Microsoft Office Excel Workbook (*.xls).

5. On a separate worksheet, list the values (expressed in hectares) for area of contributing and recharge zones (see cells E5 to E6 in **Figure E1**).

6. List the values (expressed in hectares) for area of each use scenario in the contributing zone (see cells B9 to B10 in **Figure E1**).

7. List the values (expressed in hectares) for area of each use scenario in the recharge zone (see cells B13 to B14 in **Figure E1**).

8. Calculate (imbedded in cell) the values (expressed in hectares) for non-cropped area in each zone (see cells B20 to B21 in **Figure E1**; formula *e.g.* B20=E5-B9-B10).

9. Insert the value (expressed in µg/L) for the average monitored base flow concentration (see cell B17 in **Figure E1**).

10. Insert the value for fraction of stream flow attributed to base flow (see cell B18 in **Figure E1**).

11. Copy the pesticide mass flux in runoff (RFLX; expressed as 10^{-5} g/cm^2 or kg/ha) outputs for each use scenario from the respective *.xls files converted from *.zts and paste them on the worksheet (see columns F and I in **Figure E1**).

12. Copy the runoff flux (RUNF; expressed as cm) outputs for each use scenario and the residential scenario from the respective *.xls files converted from *.zts and paste them on the worksheet (see columns E, H, and K in **Figure E1**).

13. Calculate daily residue mass in runoff (µg) from each use area in the contributing zone (CZ) in separate columns, one for each use (see columns M and N in **Figure E1**) using the formula:

$$\text{Daily Mass in Runoff } (\mu g) = \text{RFLX (kg/ha) x Use Area (ha) x } 10^9 \ \mu g/kg$$

(*e.g.* M25=F25*$B$9*1000000000)

14. Calculate daily runoff mass (µg) from each use area in the recharge zone (RZ) in separate columns, one for each use (see columns Q and R in **Figure E1**) using the formula above (formula *e.g.* Q25=F25*$B$13*1000000000).

15. Calculate mass totals (µg) for each aquifer zone in separate columns (see columns O and S in **Figure E1**; formula *e.g.* O25=SUM(M25:N25)).

16. Calculate daily runoff (L) from each use and non-use area in the CZ in separate columns, one for each scenario (see columns U, V, and W in **Figure E2**) using the formula:

$$\text{Daily Runoff (L)} = \text{RUNF (cm) x Use/Non-use Area (ha) x } 10^8 \text{ cm}^2\text{/ha x } 10^{-3} \text{ L/cm}^3$$

(*e.g.* U25=E25*$B$9*100000000/1000)

17. Calculate daily runoff (L) from each use and non-use area in the RZ in separate columns, one for each scenario (see columns Z, AA, and AB in **Figure E2**) using the formula above (formula *e.g.* Z25=E25*$B$13*100000000/1000).

18. Calculate runoff totals (L) for each aquifer zone in separate columns (see columns X and AC in **Figure E2**; formula *e.g.* X25=SUM(U25:W25)).

19. In order to estimate base stream flow in the contributing zone:

    a. Calculate the sum of total runoff (L) in the CZ (see cell N7 in **Figure E1**; formula *e.g.* N7=SUM($X$25:$X$10981)).
    b. Calculate the number of days modeled (see cell N8 in **Figure E1**; formula *e.g.* N8=COUNT($C$25:$C$10981)).
    c. Calculate the average daily flow in runoff (L/d) from the contributing zone (see cell N9 in **Figure E1**; formula *e.g.* N9=N7/N8).
    d. Calculate base stream flow (L/d) (see cell N10 in **Figure E1**) using the formula:

$$\text{Base Stream Flow (L/d)} = \text{Base Stream Fraction x Mean CZ Runoff Flow (L/d)} \, / \, \text{CZ Runoff Fraction}$$

[*e.g.* N10 =$B$18*N9/(1-$B$18)]

20. Calculate daily runoff EECs (µg/L) for each aquifer zone in separate columns (see columns AE and AJ in **Figure E2**) using the formula:

$$\text{Daily Runoff EEC } (\mu g/L) = \text{Daily Total Mass in Zone Runoff } (\mu g) \, / \, \text{Daily Zone Runoff (L)}$$

[*e.g.* AE25=IF(X25=0, 0, O25/X25)]

21. Calculate the total daily CZ stream flow (L) in a separate column by summing the total daily runoff in the CZ (L) and the base stream flow (L) (see column AF in **Figure E2**; formula *e.g.* AF25 =$N$10+X25).

22. Calculate the daily stream flow fraction from runoff (Stream Dilution Factor) in a separate column (see column AG in **Figure E2**; formula *e.g.* AG25=X25/AF25).

23. Calculate daily stream EECs (µg/L) in the contributing zone (see column AH in **Figure E2**) using the formula:

Daily CZ Stream EEC (µg/L) = [Stream Dilution Factor x CZ Runoff EEC (µg/L)] + [Base Flow Dilution Factor x Mean Base Flow Concentration (µg/L)]

[*e.g.* AH25=AG25*AE25+(1-AG25)*$B$17]

24. Calculate the total daily flow into the Barton Springs (L) by summing the total daily CZ stream flow (L) and the total RZ runoff (L) (see column AL in **Figure E3**; formula *e.g.* AL25=AF25+AC25).

25. Calculate the fraction of flow in the Barton Springs from RZ runoff (RZ Flow Fraction; see column AM in **Figure E3**; formula *e.g.* AM25=AC25/AL25).

26. Calculate the fraction of flow in the Barton Springs from CZ stream flow (CZ Stream Flow Fraction; see column AN in **Figure E3**; formula *e.g.* AN25 =AF25/AL25).

27. Calculate daily EECs (µg/L) in the Barton Springs (see column AO in **Figure E3**) using the formula:

Daily Barton Springs EEC (µg/L) = [RZ Flow Fraction x Daily RZ Runoff EEC (µg/L)] + [CZ Stream Flow Fraction x Daily CZ Stream EEC (µg/L)]

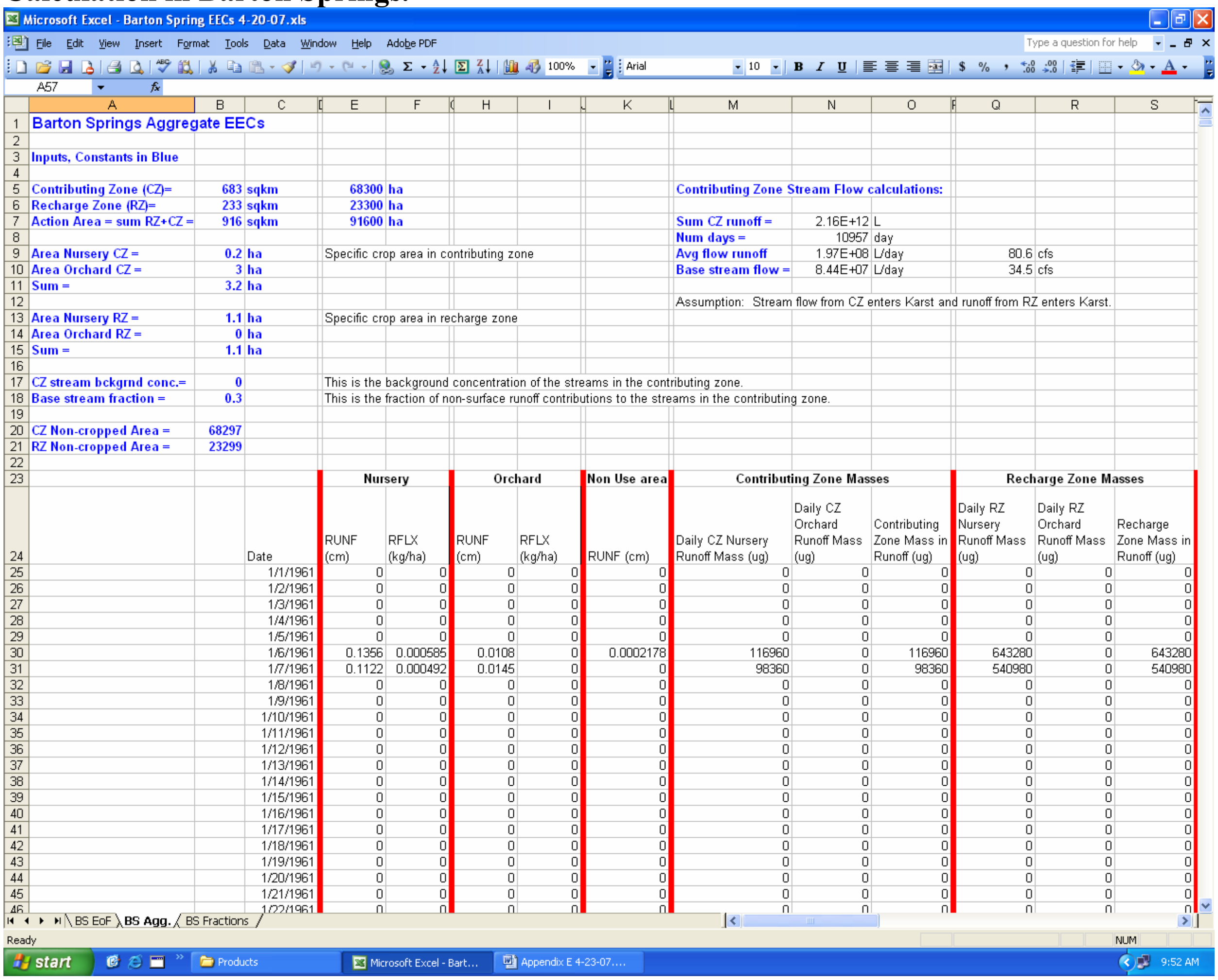
(*e.g.* AO25=AM25*AJ25+AN25*AH25)

28. Calculate rolling time weighted averages for the appropriate durations including 14-day (see column AQ in **Figure E3**), 21-day (see column AR), 30-day (see column AS), 60-day (see column AT), and 90-day (see column AU) durations. Time weighted averages are calculated using the daily values from half of the duration preceding the day of interest and half of the duration after the day of interest. For example, the 14-day average on January 14 is calculated by averaging the daily values from January 8 to January 21. This calculation is repeated for each day and for each duration for the entire 30 years of daily values.

29. List the peak EEC and rolling 14-day, 21-day, 30-day, 60-day, and 90-day average EEC for each year between 1961 and 1990 [see columns AX to BC in **Figure E3**; formula *e.g.* AX25 =MAX(AO25:AO389)].

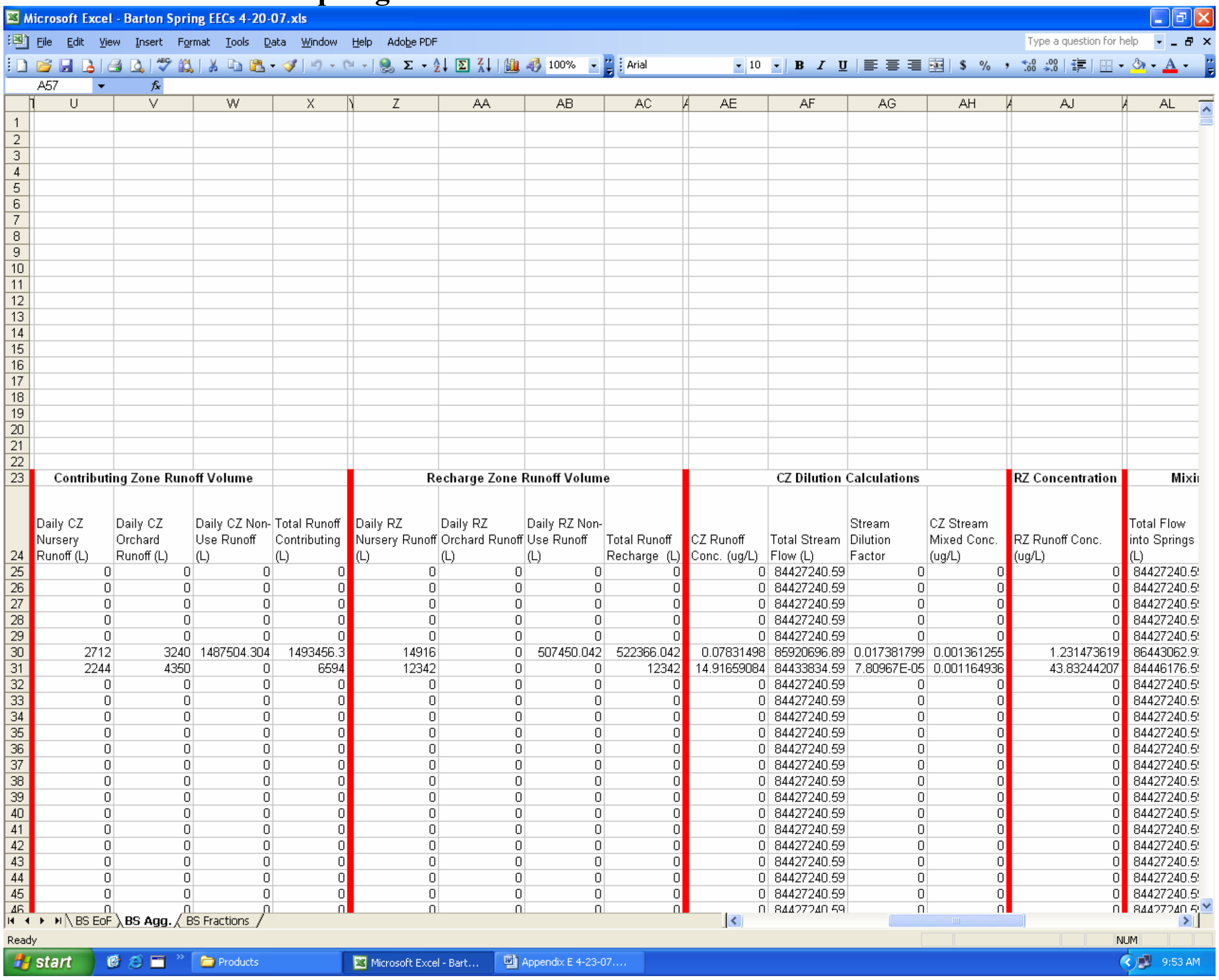
30. Calculate the 1-in-10-year return frequency for each duration [see row 57, AX to BC in **Figure E3**; formula *e.g.* AX57=PERCENTILE(AX25:AX54,0.9)].

Figure E1. Screen Shot of Columns A to S of an Example Excel Worksheet for Estimate Calculation in Barton Springs.



	A	B	C	E	F	H	I	K	M	N	O	Q	R	S
1	Barton Springs Aggregate EECs													
2														
3	Inputs, Constants in Blue													
4														
5	Contributing Zone (CZ)=	683	sqkm	68300	ha				Contributing Zone Stream Flow calculations:					
6	Recharge Zone (RZ)=	233	sqkm	23300	ha									
7	Action Area = sum RZ+CZ =	916	sqkm	91600	ha				Sum CZ runoff =	2.16E+12	L			
8									Num days =	10957	day			
9	Area Nursery CZ =	0.2	ha	Specific crop area in contributing zone					Avg flow runoff	1.97E+08	L/day	80.6	cfs	
10	Area Orchard CZ =	3	ha						Base stream flow =	8.44E+07	L/day	34.5	cfs	
11	Sum =	3.2	ha											
12									Assumption: Stream flow from CZ enters Karst and runoff from RZ enters Karst.					
13	Area Nursery RZ =	1.1	ha	Specific crop area in recharge zone										
14	Area Orchard RZ =	0	ha											
15	Sum =	1.1	ha											
16														
17	CZ stream bckgrnd conc.=	0		This is the background concentration of the streams in the contributing zone.										
18	Base stream fraction =	0.3		This is the fraction of non-surface runoff contributions to the streams in the contributing zone.										
19														
20	CZ Non-cropped Area =	68297												
21	RZ Non-cropped Area =	23299												
22														
23				Nursery		Orchard		Non Use area	Contributing Zone Masses			Recharge Zone Masses		
24			Date	RUNF (cm)	RFLX (kg/ha)	RUNF (cm)	RFLX (kg/ha)	RUNF (cm)	Daily CZ Nursery Runoff Mass (ug)	Daily CZ Orchard Runoff Mass (ug)	Contributing Zone Mass in Runoff (ug)	Daily RZ Nursery Runoff Mass (ug)	Daily RZ Orchard Runoff Mass (ug)	Recharge Zone Mass in Runoff (ug)
25			1/1/1961	0	0	0	0	0	0	0	0	0	0	0
26			1/2/1961	0	0	0	0	0	0	0	0	0	0	0
27			1/3/1961	0	0	0	0	0	0	0	0	0	0	0
28			1/4/1961	0	0	0	0	0	0	0	0	0	0	0
29			1/5/1961	0	0	0	0	0	0	0	0	0	0	0
30			1/6/1961	0.1356	0.000585	0.0108	0	0.0002178	116960	0	116960	643280	0	643280
31			1/7/1961	0.1122	0.000492	0.0145	0	0	98360	0	98360	540980	0	540980
32			1/8/1961	0	0	0	0	0	0	0	0	0	0	0
33			1/9/1961	0	0	0	0	0	0	0	0	0	0	0
34			1/10/1961	0	0	0	0	0	0	0	0	0	0	0
35			1/11/1961	0	0	0	0	0	0	0	0	0	0	0
36			1/12/1961	0	0	0	0	0	0	0	0	0	0	0
37			1/13/1961	0	0	0	0	0	0	0	0	0	0	0
38			1/14/1961	0	0	0	0	0	0	0	0	0	0	0
39			1/15/1961	0	0	0	0	0	0	0	0	0	0	0
40			1/16/1961	0	0	0	0	0	0	0	0	0	0	0
41			1/17/1961	0	0	0	0	0	0	0	0	0	0	0
42			1/18/1961	0	0	0	0	0	0	0	0	0	0	0
43			1/19/1961	0	0	0	0	0	0	0	0	0	0	0
44			1/20/1961	0	0	0	0	0	0	0	0	0	0	0
45			1/21/1961	0	0	0	0	0	0	0	0	0	0	0
46			1/22/1961	0	0	0	0	0	0	0	0	0	0	0

Figure E2. Screen Shot of Columns U to AK of an Example Excel Worksheet for Estimate Calculation in Barton Springs.



Microsoft Excel - Barton Spring EECs 4-20-07.xls

Row	U	V	W	X	Z	AA	AB	AC	AE	AF	AG	AH	AJ	AL
23	Contributing Zone Runoff Volume				Recharge Zone Runoff Volume				CZ Dilution Calculations				RZ Concentration	Mixi
24	Daily CZ Nursery Runoff (L)	Daily CZ Orchard Runoff (L)	Daily CZ Non-Use Runoff (L)	Total Runoff Contributing (L)	Daily RZ Nursery Runoff (L)	Daily RZ Orchard Runoff (L)	Daily RZ Non-Use Runoff (L)	Total Runoff Recharge (L)	CZ Runoff Conc. (ug/L)	Total Stream Flow (L)	Stream Dilution Factor	CZ Stream Mixed Conc. (ug/L)	RZ Runoff Conc. (ug/L)	Total Flow into Springs (L)
25	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
26	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
27	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
28	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
29	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
30	2712	3240	1487504.304	1493456.3	14916	0	507450.042	522366.042	0.07831498	85920696.89	0.017381799	0.001361255	1.231473619	86443062.9
31	2244	4350	0	6594	12342	0	0	12342	14.91659084	84433834.59	7.80967E-05	0.001164936	43.83244207	84446176.5
32	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
33	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
34	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
35	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
36	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
37	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
38	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
39	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
40	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
41	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
42	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
43	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
44	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
45	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5
46	0	0	0	0	0	0	0	0	0	84427240.59	0	0	0	84427240.5

BS EoF | BS Agg. | BS Fractions

Figure E3. Screen Shot of Columns AL to BD of an Example Excel Worksheet for Estimate Calculation in Barton Springs.

Microsoft Excel - Barton Spring EECs 4-20-07.xls

	AL	AM	AN	AO	AQ	AR	AS	AT	AU	AW	AX	AY	AZ	BA	BB	BC	BD
23	Mixing of Stream and Direct Recharge				Running Averages (ppb)					Yearly Peaks and Averages (ppb)							
24	Total Flow into Springs (L)	Recharge Fraction	Stream CZ Fraction	Daily Barton Springs EEC (ug/L)	14 day Avg	21 day Avg	30 day Avg	60 day Avg	90 day Avg	Year	Max Peak	Max 14 day	Max 21 day	Max 30 day	Max 60 day	Max 90 day	Annual average
25	84427240.59	0	1	0						1961	3.97E-02	6.04E-03	4.03E-03	3.05E-03	2.19E-03	1.66E-03	6.90E-04
26	84427240.59	0	1	0						1962	5.17E-02	4.15E-03	3.42E-03	2.67E-03	2.13E-03	1.90E-03	9.69E-04
27	84427240.59	0	1	0						1963	4.54E-02	4.15E-03	3.09E-03	3.29E-03	2.02E-03	1.77E-03	9.51E-04
28	84427240.59	0	1	0						1964	5.34E-02	5.19E-03	3.50E-03	2.82E-03	2.12E-03	1.65E-03	8.73E-04
29	84427240.59	0	1	0						1965	5.16E-02	4.13E-03	3.29E-03	2.55E-03	2.19E-03	1.57E-03	9.39E-04
30	86443062.93	0.006042891	0.993957109	0.008794691						1966	4.68E-02	3.92E-03	3.58E-03	2.50E-03	2.15E-03	1.53E-03	8.03E-04
31	84446176.59	0.000146152	0.999853848	0.007570976	0.00					1967	6.33E-02	7.41E-03	4.94E-03	4.01E-03	2.65E-03	2.91E-03	1.51E-03
32	84427240.59	0	1	0	0.00					1968	3.24E-02	5.13E-03	3.44E-03	2.59E-03	2.12E-03	1.81E-03	1.07E-03
33	84427240.59	0	1	0	0.00					1969	4.27E-02	4.02E-03	3.35E-03	2.76E-03	2.06E-03	1.86E-03	1.03E-03
34	84427240.59	0	1	0	0.00					1970	5.94E-02	7.72E-03	6.89E-03	5.92E-03	3.98E-03	2.66E-03	1.16E-03
35	84427240.59	0	1	0	0.00	0.00				1971	5.56E-02	5.57E-03	3.71E-03	3.48E-03	2.55E-03	2.23E-03	1.14E-03
36	84427240.59	0	1	0	0.00	0.00				1972	5.19E-02	7.70E-03	5.62E-03	4.15E-03	2.72E-03	2.12E-03	1.35E-03
37	84427240.59	0	1	0	0.00	0.00				1973	4.72E-02	6.51E-03	4.49E-03	4.44E-03	3.90E-03	3.05E-03	1.41E-03
38	84427240.59	0	1	0	0.00	0.00				1974	3.59E-02	5.48E-03	4.59E-03	4.08E-03	2.45E-03	2.08E-03	1.19E-03
39	84427240.59	0	1	0	0.00	0.00	0.00			1975	2.61E-02	6.39E-03	4.51E-03	3.75E-03	2.28E-03	1.62E-03	6.25E-04
40	84427240.59	0	1	0	0.00	0.00	0.00			1976	6.13E-02	5.29E-03	3.70E-03	3.58E-03	2.78E-03	2.16E-03	1.59E-03
41	84427240.59	0	1	0	0.00	0.00	0.00			1977	5.27E-02	6.46E-03	5.56E-03	3.89E-03	2.65E-03	2.27E-03	1.17E-03
42	84427240.59	0	1	0	0.00	0.00	0.00			1978	3.96E-02	6.82E-03	4.55E-03	3.19E-03	2.30E-03	1.77E-03	1.07E-03
43	84427240.59	0	1	0	0.00	0.00	0.00			1979	3.73E-02	6.49E-03	4.92E-03	4.12E-03	3.36E-03	2.87E-03	1.23E-03
44	84427240.59	0	1	0	0.00	0.00	0.00			1980	4.33E-02	4.26E-03	3.61E-03	2.62E-03	2.03E-03	1.67E-03	9.20E-04
45	84427240.59	0	1	0	0.00	0.00	0.00			1981	4.71E-02	5.11E-03	4.13E-03	3.17E-03	2.56E-03	2.10E-03	1.14E-03
46	84427240.59	0	1	0	0.00	0.00	0.00			1982	5.94E-02	5.04E-03	4.16E-03	3.74E-03	2.60E-03	2.27E-03	1.23E-03
47	84427240.59	0	1	0	0.00	0.00	0.00			1983	4.24E-02	4.84E-03	3.26E-03	3.43E-03	2.40E-03	2.18E-03	1.38E-03
48	84427240.59	0	1	0	0.00	0.00	0.00			1984	8.44E-02	6.66E-03	7.42E-03	5.19E-03	2.65E-03	2.23E-03	1.13E-03
49	84427240.59	0	1	0	0.00	0.00	0.00			1985	4.20E-02	5.94E-03	5.01E-03	3.96E-03	2.67E-03	2.28E-03	1.12E-03
50	84427240.59	0	1	0	0.00	0.00	0.00			1986	4.59E-02	5.70E-03	4.54E-03	3.45E-03	3.03E-03	2.25E-03	1.20E-03
51	84427240.59	0	1	0	0.00	0.00	0.00			1987	5.33E-02	6.62E-03	5.75E-03	5.28E-03	3.59E-03	2.75E-03	1.55E-03
52	84427240.59	0	1	0	0.00	0.00	0.00			1988	6.02E-02	5.23E-03	5.10E-03	4.36E-03	3.02E-03	2.70E-03	1.04E-03
53	84427240.59	0	1	0	0.00	0.00	0.00			1989	4.02E-02	4.30E-03	3.94E-03	2.89E-03	2.06E-03	1.73E-03	9.34E-04
54	84427240.59	0	1	0	0.00	0.00	0.00	0.00		1990	4.82E-02	5.53E-03	4.06E-03	3.47E-03	1.94E-03	1.53E-03	9.34E-04
55	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
56	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
57	84427240.59	0	1	0	0.00	0.00	0.00	0.00		90th %-ile	0.060272	0.006878	0.005636	0.004519	0.00338	0.002765	0.001423
58	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
59	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
60	12093457001	0.252598824	0.747401176	0.007034382	0.00	0.00	0.00	0.00									
61	84429351.79	2.11585E-05	0.999978841	0.002554443	0.00	0.00	0.00	0.00									
62	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
63	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
64	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
65	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
66	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
67	84427240.59	0	1	0	0.00	0.00	0.00	0.00									
68	84427240.59	0	1	0	0.00	0.00	0.00	0.00									

BS EoF | BS Agg. | BS Fractions

PRZM Input Files for the Barton Springs Salamander Endangered Species Assessment of Diazinon.

Ornamentals Input File

Output File: Diaz_nursery_NoIrrig_Apr12				
Metfile:	w13958.dvf			
PRZM scenario:	TX_BSSNursery_NoIrrig.txt			
EXAMS environment file:	pond298.exv			
Chemical Name:	Diazinon			
Description	Variable Name		Value	Units Comments
Molecular weight	mwt	304.3	g/mol	
Henry's Law Const.	henry	1.40e-6	atm-m^3/mol	
Vapor Pressure	vapr	1.40e-4	torr	
Solubility	sol	400	mg/L	
Kd	Kd		mg/L	
Koc	Koc	616	mg/L	
Photolysis half-life	kdp	37	days	Half-life
Aerobic Aquatic Metabolism	kbacw	77.4	days	Halfife
Anaerobic Aquatic Metabolism	kbacs	0	days	Halfife
Aerobic Soil Metabolism	asm	38.7	days	Halfife
Hydrolysis:	pH 5	12	days	Half-life
Hydrolysis:	pH 7	138	days	Half-life
Hydrolysis:	pH 9	77	days	Half-life
Method:	CAM	2	integer	See PRZM manual
Incorporation Depth:	DEPI	0	cm	
Application Rate:	TAPP	1.121	kg/ha	
Application Efficiency:	APPEFF	0.99	fraction	
Spray Drift	DRFT	0.01	fraction of application rate applied to pond	
Application Date	Date	02-01	dd/mm or dd/mmm or dd-mm or dd-mmm	
Interval 1	interval	14	days	Set to 0 or delete line for single app.
Interval 2	interval	14	days	Set to 0 or delete line for single app.
Interval 3	interval	14	days	Set to 0 or delete line for single app.
Interval 4	interval	14	days	Set to 0 or delete line for single app.
Interval 5	interval	14	days	Set to 0 or delete line for single app.
Interval 6	interval	14	days	Set to 0 or delete line for single app.
Interval 7	interval	14	days	Set to 0 or delete line for single app.
Interval 8	interval	14	days	Set to 0 or delete line for single app.
Interval 9	interval	14	days	Set to 0 or delete line for single app.
Interval 10	interval	14	days	Set to 0 or delete line for single app.
Interval 11	interval	14	days	Set to 0 or delete line for single app.
Interval 12	interval	14	days	Set to 0 or delete line for single app.
Interval 13	interval	14	days	Set to 0 or delete line for single app.
Interval 14	interval	14	days	Set to 0 or delete line for single app.
Interval 15	interval	14	days	Set to 0 or delete line for single app.
Interval 16	interval	14	days	Set to 0 or delete line for single app.
Interval 17	interval	14	days	Set to 0 or delete line for single app.
Interval 18	interval	14	days	Set to 0 or delete line for single app.
Interval 19	interval	14	days	Set to 0 or delete line for single app.
Interval 20	interval	14	days	Set to 0 or delete line for single app.
Interval 21	interval	14	days	Set to 0 or delete line for single app.
Interval 22	interval	14	days	Set to 0 or delete line for single app.
Interval 23	interval	14	days	Set to 0 or delete line for single app.
Interval 24	interval	14	days	Set to 0 or delete line for single app.
Interval 25	interval	14	days	Set to 0 or delete line for single app.
Record 17:	FILTRA			

	IPSCND	2		
	UPTKF			
Record 18:	PLVKRT			
	PLDKRT			
	FEXTRC	0.5		
Flag for Index Res. Run	IR	Pond		
Flag for runoff calc.	RUNOFF	none	none, monthly or total(average of entire run)	

Peaches Input File

Output File: Diaz_orchard_Apr12

Metfile:	w13958.dvf			
PRZM scenario:	TX_BSSOrchard.txt			
EXAMS environment file:	pond298.exv			
Chemical Name:	Diazinon			
Description	Variable Name		Value	Units Comments
Molecular weight	mwt	304.3	g/mol	
Henry's Law Const.	henry	1.40e-6	atm-m^3/mol	
Vapor Pressure	vapr	1.40e-4	torr	
Solubility	sol	400	mg/L	
Kd	Kd		mg/L	
Koc	Koc	616	mg/L	
Photolysis half-life	kdp	37	days	Half-life
Aerobic Aquatic Metabolism	kbacw	77.4	days	Halfife
Anaerobic Aquatic Metabolism	kbacs	0	days	Halfife
Aerobic Soil Metabolism	asm	38.7	days	Halfife
Hydrolysis:	pH 5	12	days	Half-life
Hydrolysis:	pH 7	138	days	Half-life
Hydrolysis:	pH 9	77	days	Half-life
Method:	CAM	2	integer	See PRZM manual
Incorporation Depth:	DEPI	0	cm	
Application Rate:	TAPP	2.242	kg/ha	
Application Efficiency:	APPEFF	0.99	fraction	
Spray Drift	DRFT	0.01	fraction of application rate applied to pond	
Application Date	Date	15-01	dd/mm or dd/mmm or dd-mm or dd-mmm	
Interval 1	interval	120	days	Set to 0 or delete line for single app.
Record 17:	FILTRA			
	IPSCND	3		
	UPTKF			
Record 18:	PLVKRT			
	PLDKRT			
	FEXTRC	0.5		
Flag for Index Res. Run	IR	Pond		
Flag for runoff calc.	RUNOFF	none	none, monthly or total(average of entire run)	

Residential Input File (for runoff estimates)

Output File: Diaz_res_noirrig_Mar13

Metfile:	w13958.dvf			
PRZM scenario:	TX_BSSResidential_NoIrrig.txt			
EXAMS environment file:	pond298.exv			
Chemical Name:	Diazinon			
Description	Variable Name		Value	Units Comments
Molecular weight	mwt	304.3	g/mol	
Henry's Law Const.	henry	1.40e-6	atm-m^3/mol	
Vapor Pressure	vapr	1.40e-4	torr	
Solubility	sol	400	mg/L	

Kd	Kd		mg/L	
Koc	Koc	616	mg/L	
Photolysis half-life	kdp	37	days	Half-life
Aerobic Aquatic Metabolism	kbacw	77.4	days	Halfife
Anaerobic Aquatic Metabolism	kbacs	0	days	Halfife
Aerobic Soil Metabolism	asm	38.7	days	Halfife
Hydrolysis:	pH 5	12	days	Half-life
Hydrolysis:	pH 7	138	days	Half-life
Hydrolysis:	pH 9	77	days	Half-life
Method:	CAM	2	integer	See PRZM manual
Incorporation Depth:	DEPI	0	cm	
Application Rate:	TAPP	1.121	kg/ha	
Application Efficiency:	APPEFF	0.99	fraction	
Spray Drift	DRFT	0.01	fraction of application rate applied to pond	
Application Date	Date	15-05	dd/mm or dd/mmm or dd-mm or dd-mmm	
Interval 1	interval	7	days	Set to 0 or delete line for single app.
Interval 2	interval	7	days	Set to 0 or delete line for single app.
Interval 3	interval	7	days	Set to 0 or delete line for single app.
Interval 4	interval	7	days	Set to 0 or delete line for single app.
Record 17:	FILTRA			
	IPSCND	3		
	UPTKF			
Record 18:	PLVKRT			
	PLDKRT			
	FEXTRC	0.5		
Flag for Index Res. Run	IR	Pond		
Flag for runoff calc.	RUNOFF	none	none, monthly or total(average of entire run)	

Appendix F. Species Sensitivity Distribution Data.

Tables F.1-F.4 contain the 96-hour LC_{50} data for fish and associated calculations used to derive the quantitative species sensitivity distribution shown in Figure 5.2 (of risk assessment). Tables F.5-F.8 contain the 96-hour LC_{50} data for fish and associated calculations used to derive the qualitative species sensitivity distribution shown in Figure 5.3 (of risk assessment). Tables F.9-F.12 contain the 48 to 96-hour EC_{50} data for invertebrates and associated calculations used to derive the quantitative species sensitivity distribution shown in Figure 5.4 (of risk assessment). Tables F.13-F.16 contain the 48 to 96-hour EC_{50} data for invertebrates and associated calculations used to derive the qualitative species sensitivity distribution shown in Figure 5.5 (of risk assessment). References are located in **Appendix H**.

Table F.1. Summary of 96 hour LC50 data for effects of diazinon on freshwater fish (quantitative data).

Common Name	Species Name	Mean LC50 (ppb)	Log 10 LC50	Test Subst. (% a.i.)	MRID/ Accession	ECOTOX Number	Comments
Bluegill sunfish	*Lepomis macrochirus*	136	2.134	91.0	104923	NA	cited in RED
Bluegill sunfish	*Lepomis macrochirus*	460	2.663	92.5	ROODI007	NA	cited in RED
Bluegill sunfish	*Lepomis macrochirus*	168	2.225	92.0	40094602	NA	cited in RED
Brook trout	*Salelinus fontinalis*	770	2.886	92.5	ROODI007	NA	cited in RED
Cutthroat trout	*Oncorhynchus clarki*	1700	3.230	92.0	40094602	NA	cited in RED
Fathead Minnow	*Pimephales promeals*	7800	3.892	92.5	ROODI007	NA	cited in RED
Flagfish	*Jordanella floridae*	1600	3.204	92.5	ROODI007	NA	cited in RED
Guppy	*Lebistes reticulatus*	1100	3.041	NR	5000811	NA	cited in RED
Lake trout	*Salevelinus namaychus*	602	2.780	92.0	40094602	NA	cited in RED
Rainbow trout	*Oncorhynchus gairdneri*	90	1.954	89.0	40094602	NA	cited in RED
Rainbow trout	*Oncorhynchus sp.*	400	2.602	91.0	104923	NA	cited in RED
NR = not reported, NA = not applicable							

Table F.2. Species Mean Acute Values (SMAVs) for freshwater fish (quantitative).		
Common Name	**Species Name**	**Log10 SMAV**
Bluegill sunfish	*Lepomis macrochirus*	2.3405
Brook trout	*Salelinus fontinalis*	2.8865
Cutthroat trout	*Oncorhynchus clarki*	3.2304
Fathead Minnow	*Pimephales promeals*	3.8921
Flagfish	*Jordanella floridae*	3.2041
Guppy	*Lebistes reticulatus*	3.0414
Lake trout	*Salevelinus namaychus*	2.7796
Rainbow trout	*Oncorhynchus gairdneri*	1.954
Rainbow trout	*Oncorhynchus sp.*	2.602

Table F.3. Genus Mean Acute Values (GMAVs) for freshwater fish (quantitative).					
Common Name	**Species Name**	**Log10 GMAV**	**GMAV LC50**	**Sensitivity Rank**	**Rank on curve**
sunfish	*Lepomis*	2.3405	219	1	0.00
Brook trout	*Salelinus*	2.8865	770	4	0.50
Trout	*Oncorhynchus*	2.5956	394	2	0.17
Fathead Minnow	*Pimephales*	3.8921	7800	7	1.00
Flagfish	*Jordanella*	3.2041	1600	6	0.83
Guppy	*Lebistes*	3.0414	1100	5	0.67
Lake trout	*Salevelinus*	2.7796	602	3	0.33
Genus Mean for All:		**2.9628**	**1784**		
Genus Standard Deviation for all:		**0.4982**	**2693**		

Table F.4. Calculation of species sensitivity distribution curve for freshwater fish (quantitative).			
Proportion	**Z_P**	**Log10 point**	**Point Estimate**
0.05	-1.645	2.143229	139
0.10	-1.282	2.324089	211
0.20	-0.842	2.543314	349
0.25	-0.675	2.626761	423
0.30	-0.524	2.701754	503
0.40	-0.253	2.836776	687
0.50	0	2.962831	918
0.60	0.253	3.088885	1227
0.70	0.524	3.223907	1675
0.75	0.675	3.299141	1991
0.80	0.842	3.382347	2412
0.90	1.282	3.601572	3996
0.95	1.645	3.782432	6059
Z_P = (Log10 LC50 - fish mean GMAV)/(fish std GMAV)			

Table F.5. Summary of 96 hour LC50 data for effects of diazinon on freshwater fish (qualitative data).							
Common Name	Species Name	Mean LC50 (ppb)	Log 10 LC50	Test Substance (% a.i.)	MRID/Accession	ECOTOX Number	Comments
Common eel	*Anguilla anguilla*	80	1.903	95.0	NA	7004	
Common eel	*Anguilla anguilla*	85	1.929	95.0	NA	15687	
Common eel	*Anguilla anguilla*	85	1.929	95.0	NA	6728	
Goldfish	*Carassius auratus*	9000	3.954	91.0	NA	13000	
Hawk Fish Carp	*Cirrhinus mrigala*	1002	3.001	100.0	NA	45088	
Flagfish	*Jordanella floridae*	1600	3.204	92.5	ROODI007	NA	cited in RED
Flagfish	*Jordanella floridae*	1500	3.176	92.5	NA	664	
Flagfish	*Jordanella floridae*	1800	3.255	92.5	NA	664	
Guppy	*Lebistes reticulatus*	1100	3.041	NR	5000811	NA	cited in RED
Bluegill sunfish	*Lepomis macrochirus*	136	2.134	91.0	104923	NA	cited in RED
Bluegill sunfish	*Lepomis macrochirus*	460	2.663	92.5	ROODI007	NA	cited in RED
Bluegill sunfish	*Lepomis macrochirus*	168	2.225	92.0	40094602	NA	cited in RED
Bluegill sunfish	*Lepomis macrochirus*	400	2.602	100.0	NA	13005	
Bluegill sunfish	*Lepomis macrochirus*	440	2.643	92.5	NA	664	
Bluegill sunfish	*Lepomis macrochirus*	480	2.681	92.5	NA	664	
Eastern rainbow fish	*Melanotaenia duboulayi*	8850	3.947	90.2	NA	85626	
Eastern rainbow fish	*Melanotaenia duboulayi*	11520	4.061	90.2	NA	85626	
Eastern rainbow fish	*Melanotaenia duboulayi*	6440	3.809	90.2	NA	85626	
Golden shiner	*Notemingonus crysoleucas*	400	2.602	100.0	NA	13005	
Cutthroat trout	*Oncorhynchus clarki*	1700	3.230	92.0	40094602	NA	cited in RED
Rainbow trout	*Oncorhynchus gairdneri*	90	1.954	89.0	40094602	NA	cited in RED
Rainbow trout	*Oncorhynchus sp.*	400	2.602	91.0	104923	NA	cited in RED
Chinook salmon	*Oncorhynchus tshawytscha*	29500	4.470	97.0	NA	82750, 84761	
Chinook salmon	*Oncorhynchus tshawytscha*	545000	5.736	97.0	NA	82750, 84761	
Fathead	*Pimephales*	7800	3.892	92.5	ROODI007	NA	cited in

Minnow	*promeals*						RED
Fathead Minnow	*Pimephales promeals*	6100	3.785	87.1	NA	15462	
Fathead Minnow	*Pimephales promeals*	10000	4.000	92.5	NA	664	
Fathead Minnow	*Pimephales promeals*	9350	3.971	87.1	NA	12859	
Fathead Minnow	*Pimephales promeals*	6900	3.839	87.1	NA	15462	
Fathead Minnow	*Pimephales promeals*	6800	3.833	92.5	NA	664	
Fathead Minnow	*Pimephales promeals*	6600	3.820	92.5	NA	664	
Fathead Minnow	*Pimephales promeals*	6000	3.778	100.0	NA	64773	
Fathead Minnow	*Pimephales promeals*	4300	3.633	87.1	NA	15462	
Sacromento splittail	*Pogonichthys macrolepidot*	7500	3.875	100.0	NA	64773	
Brook trout	*Salelinus fontinalis*	770	2.886	92.5	ROODI007	NA	cited in RED
Brook trout	*Salelinus fontinalis*	400	2.602	411.0	NA	13005	
Brook trout	*Salelinus fontinalis*	450	2.653	92.5	NA	664	
Brook trout	*Salelinus fontinalis*	1050	3.021	92.5	NA	664	
Brook trout	*Salelinus fontinalis*	800	2.903	92.5	NA	664	
Lake trout	*Salevelinus namaychus*	602	2.780	92.0	40094602	NA	cited in RED
Mozambique tilapia	*Tilapia mossambica*	15850	4.200	90.0	NA	66476	

NR = not reported, NA = not applicable

Table F.6. Species Mean Acute Values (SMAVs) for freshwater fish (qualitative).		
Common Name	**Species Name**	**Log10 SMAV**
Common eel	*Anguilla anguilla*	1.921
Goldfish	*Carassius auratus*	3.954
Hawk Fish Carp	*Cirrhinus mrigala*	3.001
Flagfish	*Jordanella floridae*	3.2118
Guppy	*Lebistes reticulatus*	3.0414
Bluegill sunfish	*Lepomis macrochirus*	2.4914
Eastern rainbow fish	*Melanotaenia duboulayi*	3.939
Golden shiner	*Notemingonus crysoleucas*	2.602
Cutthroat trout	*Oncorhynchus clarki*	3.2304
Rainbow trout	*Oncorhynchus gairdneri*	1.954
Rainbow trout	*Oncorhynchus sp.*	2.602
Chinook salmon	*Oncorhynchus tshawytscha*	5.103
Fathead Minnow	*Pimephales promeals*	3.8390
Sacromento splittail	*Pogonichthys macrolepidot*	3.875
Brook trout	*Salelinus fontinalis*	2.8132
Lake trout	*Salevelinus namaychus*	2.7796
Mozambique tilapia	*Tilapia mossambica*	4.200

Table F.7. Genus Mean Acute Values (GMAVs) for freshwater fish (qualitative).					
Common Name	**Species Name**	**Log10 GMAV**	**GMAV LC50**	**Sensitivity Rank**	**Rank on curve**
Eel	*Anguilla*	1.9206	83	1	0.00
Goldfish	*Carassius*	3.9542	9000	13	0.92
Carp	*Cirrhinus*	3.0009	1002	6	0.38
Flagfish	*Jordanella*	3.2118	1629	8	0.54
Guppy	*Lebistes*	3.0414	1100	7	0.46
sunfish	*Lepomis*	2.4914	310	2	0.08
rainbow fish	*Melanotaenia*	3.9391	8691	12	0.85
shiner	*Notemingonus*	2.6021	400	3	0.15
Trout	*Oncorhynchus*	3.2225	1669	9	0.62
Fathead Minnow	*Pimephales*	3.8390	6902	10	0.69
splittail	*Pogonichthys*	3.8751	7500	11	0.77
Brook trout	*Salelinus*	2.8132	650	5	0.31
Lake trout	*Salevelinus*	2.7796	602	4	0.23
tilapia	*Tilapia*	4.2000	15850	14	1.00
Genus Mean for All:		**3.2065**	**3956**		
Genus Standard Deviation for all:		**0.6718**	**4814**		

Table F.8. Calculation of species sensitivity distribution curve for freshwater fish (qualitative).			
Proportion	**Z_P**	**Log10 point**	**Point Estimate**
0.05	-1.645	2.101405	126
0.10	-1.282	2.345263	221
0.20	-0.842	2.640848	437
0.25	-0.675	2.753361	567
0.30	-0.524	2.854475	715
0.40	-0.253	3.036528	1088
0.50	0	3.20649	1609
0.60	0.253	3.376451	2379
0.70	0.524	3.558504	3618
0.75	0.675	3.659944	4570
0.80	0.842	3.772132	5917
0.90	1.282	4.067717	11687
0.95	1.645	4.311574	20492
Z_P = (Log10 LC50 - fish mean GMAV)/(fish std GMAV)			

Table F.9. Summary of 48-96 hour EC50 data for effects of diazinon on freshwater invertebrates (quantitative).							
Common Name	**Species Name**	**Mean EC50 (ppb)**	**Log 10 EC50**	**Test Substance (% a.i.)**	**MRID**	**ECOTOX Number**	**Comments**
waterflea	*Ceriodaphnia dubia*	**0.21**	-0.678	**NA**		**76752**	
waterflea	*Ceriodaphnia dubia*	**0.45**	-0.347	**NA**		**76752**	
daphnid	*Simocephalus serrulatus*	1.34	0.127	89.0	40094602	NA	cited in RED, updated by 10-5-05 memo*
daphnid	*Simocephalus serrulatus*	1.67	0.223			NA	cited in 10-5-05 memo*
daphnid	*Daphnia pulex*	0.79	-0.102	89.0	40094602	NA	cited in RED, updated by 10-5-05 memo*
daphnid	*Daphnia magna*	0.83	-0.081	>89.0	109022	NA	cited in RED
mosquito larvae	*Culex pipiens fatigans*	35.0	1.544	NR	5000811	NA	cited in RED
scud	*Gammarus fasciatus*	2.0	0.299	89.0	40094602	NA	cited in RED, updated by 10-5-05 memo*
stonefly	*Pteronarcys californica*	20.49	1.312	89.0	40094602	NA	cited in RED, updated by 10-5-05 memo*
NR = not reported, NA = not applicable							

Table F.10. Species Mean Acute Values (SMAVs) for freshwater invertebrates (quantitative).		
Common Name	**Species Name**	**Log10 SMAV**
waterflea	*Ceriodaphnia dubia*	**-0.512**
daphnid	*Simocephalus sp.*	0.1749
daphnid	*Daphnia pulex*	-0.1024
daphnid	*Daphnia magna*	-0.0809
mosquito larvae	*Culex pipiens fatigans*	1.5441
scud	*Gammarus fasciatus*	0.2989
stonefly	*Pteronarcys sp.*	1.3115

Table F.11. Genus Mean Acute Values (GMAVs) for freshwater invertebrates (quantitative).					
Common Name	**Species Name**	**Log10 GMAV**	**GMAV EC50**	**Sensitivity Rank**	**Rank on curve**
waterflea	*Ceriodaphnia dubia*	**-0.512**	0.31	1	0.00
daphnid	*Simocephalus*	0.1749	1.50	3	0.40
daphnid	*Daphnia*	-0.0916	0.81	2	0.20
mosquito larvae	*Culex*	1.5441	35.00	6	1.00
scud	*Gammarus*	0.2989	1.99	4	0.60
stonefly	*Pteronarcys*	1.3115	20.49	5	0.80
Genus Mean for All:		**0.4542**	**10**		
Genus Standard Deviation for all:		**0.8071**	**14**		

Table F.12. Calculation of species sensitivity distribution curve for freshwater invertebrates (quantitative).			
Proportion	**Z_P**	**Log10 point**	**Point Estimate**
0.05	-1.645	-0.87343	0.13
0.10	-1.282	-0.58046	0.26
0.20	-0.842	-0.22533	0.60
0.25	-0.675	-0.09016	0.81
0.30	-0.524	0.031322	1.07
0.40	-0.253	0.250045	1.78
0.50	0	0.45424	2.85
0.60	0.253	0.658436	4.55
0.70	0.524	0.877159	7.54
0.75	0.675	0.99903	9.98
0.80	0.842	1.133816	13.61
0.90	1.282	1.488938	30.83
0.95	1.645	1.781914	60.52
Z_P = (Log10 LC50 - fish mean GMAV)/(fish std GMAV)			

Table F.13. Summary of 48-96 hour EC50 data for effects of diazinon for freshwater invertebrates (qualitative).								
Common Name	**Species Name**	**Mean EC50 (ppb)**	**Log 10 EC50**	**Test Subst. (% a.i.)**	**Duration of exposure (hrs)**	**MRID**	**ECOTOX Number**	**Comments**
waterflea	*Ceriodaphnia dubia*	0.21	-0.678	99.8	48	NA	76752	
waterflea	*Ceriodaphnia dubia*	0.25	-0.602	85.0	48	NA	16043	
waterflea	*Ceriodaphnia dubia*	0.26	-0.585	99.0	48	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.29	-0.538	99.0	48	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.32	-0.495	99.0	96	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.33	-0.481	99.0	72	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.33	-0.481	85.0	48	NA	16043	
waterflea	*Ceriodaphnia dubia*	0.33	-0.481	99.0	48	NA	62060	
waterflea	*Ceriodaphnia dubia*	0.35	-0.456	85.0	48	NA	16043	
waterflea	*Ceriodaphnia dubia*	0.35	-0.456	99.0	72	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.35	-0.456	99.0	96	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.36	-0.444	85.0	48	NA	16043	
waterflea	*Ceriodaphnia dubia*	0.38	-0.420	99.0	48	NA	62060	
waterflea	*Ceriodaphnia dubia*	0.4	-0.398	99.0	72	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.4	-0.398	100.0	96	NA	65773	
waterflea	*Ceriodaphnia dubia*	0.41	-0.387	100.0	96	NA	16844	
waterflea	*Ceriodaphnia dubia*	0.43	-0.367	99.0	72	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.43	-0.367	85.0	48	NA	16043	
waterflea	*Ceriodaphnia dubia*	0.47	-0.328	100.0	96	NA	16844	
waterflea	*Ceriodaphnia dubia*	0.48	-0.319	99.0	48	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.52	-0.284	99.0	48	NA	18190	
waterflea	*Ceriodaphnia dubia*	0.57	-0.244	85.0	48	NA	16043	
waterflea	*Ceriodaphnia dubia*	0.58	-0.237	99.0	48	NA	18190	
waterflea	*Ceriodaphnia*	0.59	-0.229	85.0	48	NA	16043	

	dubia							
waterflea	*Ceriodaphnia dubia*	0.66	-0.180	85.0	48	NA	16043	
waterflea	*Ceriodaphnia dubia*	0.8	-0.097	99.0	48	NA	6449	
midge	*Chironomus riparius*	22.8	1.358	99.7	96	NA	54582	
midge	*Chironomus riparius*	32	1.505	99.7	48	NA	54582	
midge	*Chironomus riparius*	167	2.223	99.7	96	NA	54582	
midge	*Chironomus riparius*	450	2.653	100.0	48	NA	61180	
mosquito larvae	*Culex pipiens fatigans*	35.0	1.544	NR		5000811	NA	cited in RED
caddisfly	*Cyrnus trimaculatus*	1.1	0.041	99.7	96	NA	55077	
daphnid	*Daphnia magna*	0.83	-0.081	>89.0		109022	NA	cited in RED
daphnid	*Daphnia magna*	0.7	-0.155	99	48	NA	6449	
daphnid	*Daphnia magna*	1.5	0.176	99	48	NA	6449	
daphnid	*Daphnia pulex*	0.79	-0.102	89.0		40094602	NA	cited in RED, updated by 10-5-05 memo*
mayfly	*Ephoron virgo*	11.8	1.072	99.7	96	NA	55077	
mayfly	*Ephoron virgo*	2.4	0.380	99.7	96	NA	66378	
mayfly	*Ephoron virgo*	1.1	0.041	99.7	96	NA	66378	
scud	*Gammarus fasciatus*	2.0	0.299	89.0		40094602	NA	cited in RED, updated by 10-5-05 memo*
scud	*Gammarus pseudolimnaeus*	16.82	1.226	100.0	96	NA	85464	
scud	*Hyalella azteca*	4.3	0.633	>98	96	NA	64955	
caddisfly	*Hydropsyche angustipennis*	29.4	1.468	99.7	96	NA	54582	
caddisfly	*Hydropsyche angustipennis*	1.3	0.114	99.7	96	NA	54582	
caddisfly	*Hydropsyche angustipennis*	2.9	0.462	99.7	48	NA	54582	
FW shrimp	*Paratya compressa*	2.33	0.367	100.0	96	NA	18945	
stonefly	*Pteronarcys californica*	20.49	1.312	89.0		40094602	NA	cited in RED, updated by 10-5-05 memo*
daphnid	*Simocephalus serrulatus*	1.34	0.127	89.0		40094602	NA	cited in RED, updated by 10-5-05

								memo*
daphnid	*Simocephalus serrulatus*	1.67	0.223			Need source	NA	cited in 10-5-05 memo*

NR = not reported, NA = not applicable
In cases where the same value (and duration) were reported multiple times by ECOTOX for the same source and species, only one entry was considered.
*USEPA 2005. Memorandum: Revaluation of acute aquatic toxicity data on diazinon. EFED to SRRD. October 5, 2005.

Table F.14. Species Mean Acute Values (SMAVs) for freshwater invertebrates (qualitative).

Common Name	Species Name	Log10 SMAV
waterflea	*Ceriodaphnia dubia*	-0.4003
midge	*Chironomus riparius*	1.9348
mosquito larvae	*Culex pipiens fatigans*	1.5441
caddisfly	*Cyrnus trimaculatus*	0.0414
daphnid	*Daphnia magna*	-0.0199
daphnid	*Daphnia pulex*	-0.1024
mayfly	*Ephoron virgo*	0.4978
scud	*Gammarus fasciatus*	0.2989
scud	*Gammarus pseudolimnaeus*	1.2258
scud	*Hyalella azteca*	0.6335
caddisfly	*Hydropsyche angustipennis*	0.6816
FW shrimp	*Paratya compressa*	0.3674
stonefly	*Pteronarcys californica*	1.3115
daphnid	*Simocephalus serrulatus*	0.1749

Table F.15. Genus Mean Acute Values (GMAVs) for freshwater invertebrates (qualitative).

Common Name	Species Name	Log10 GMAV	GMAV EC50	Sensitivity Rank	Rank on curve
waterflea	*Ceriodaphnia*	-0.4003	0.40	1	0.00
midge	*Chironomus*	1.9348	86.05	12	1.00
mosquito larvae	*Culex*	1.5441	35.00	11	0.91
caddisfly	*Cyrnus*	0.0414	1.10	3	0.18
daphnid	*Daphnia*	-0.0611	0.87	2	0.09
mayfly	*Ephoron*	0.4978	3.15	6	0.45
scud	*Gammarus*	0.7623	5.79	9	0.73
scud	*Hyalella*	0.6335	4.30	7	0.55
caddisfly	*Hydropsyche*	0.6816	4.80	8	0.64
FW shrimp	*Paratya*	0.3674	2.33	5	0.36
stonefly	*Pteronarcys*	1.3115	20.49	10	0.82
daphnid	*Simocephalus*	0.1749	1.50	4	0.27
Genus Mean for All:		**0.6240**	**14**		
Genus Standard Deviation for all:		**0.6876**	**25**		

Table F.16. Calculation of species sensitivity distribution curve for freshwater invertebrates (qualitative).			
Proportion	**Z_P**	**Log10 point**	**Point Estimate**
0.05	-1.645	-0.50719	0.31
0.10	-1.282	-0.25757	0.55
0.20	-0.842	0.04499	1.11
0.25	-0.675	0.160158	1.45
0.30	-0.524	0.263659	1.84
0.40	-0.253	0.45001	2.82
0.50	0	0.623983	4.21
0.60	0.253	0.797956	6.28
0.70	0.524	0.984306	9.65
0.75	0.675	1.08814	12.25
0.80	0.842	1.202976	15.96
0.90	1.282	1.505537	32.03
0.95	1.645	1.755151	56.91
Z_P = (Log10 LC50 - fish mean GMAV)/(fish std GMAV)			

Appendix G. The Risk Quotient Method and Levels of Concern.

The Risk Quotient Method is the means used by EFED to integrate the results of exposure and ecotoxicity data. For this method, Risk Quotients (RQs) are calculated by dividing exposure estimates by the acute and chronic ecotoxicity values (i.e., RQ = EXPOSURE/TOXICITY). These RQs are then compared to OPP's levels of concern (LOCs). These LOCs are criteria used by OPP to indicate potential risk to non-target organisms and the need to consider regulatory action. EFED has defined LOCs for acute risk, potential restricted use classification, and for endangered species.

The criteria indicate that a pesticide used as directed has the potential to cause adverse effects on non-target organisms. LOCs currently address the following risk presumption categories:

(1) acute - there is a potential for acute risk; regulatory action may be warranted in addition to restricted use classification;

(2) acute restricted use - the potential for acute risk is high, but this may be mitigated through restricted use classification;

(3) acute endangered species - the potential for acute risk to endangered species is high, regulatory action may be warranted; and

(4) chronic risk - the potential for chronic risk is high, regulatory action may be warranted.

Currently, EFED does not perform assessments for chronic risk to plants, acute or chronic risks to non-target insects, or chronic risk from granular/bait formulations to mammalian or avian species.

The ecotoxicity test values (i.e., measurement endpoints) used in the acute and chronic RQs are derived from required studies. Examples of ecotoxicity values derived from short-term laboratory studies that assess acute effects are: (1) LC50 (fish and birds), (2) LD50 (birds and mammals), (3) EC50 (aquatic plants and aquatic invertebrates), and (4) EC25 (terrestrial plants). Examples of toxicity test effect levels derived from the results of long-term laboratory studies that assess chronic effects are: (1) the Lowest Observed Adverse Effect Concentration (LOAEC) (birds, fish, and aquatic invertebrates), and (2) the No Observed Adverse Effect Concentration (NOAEC) (birds, fish and aquatic invertebrates). The NOAEC is generally used as the ecotoxicity test value in assessing chronic effects. Risk presumptions, along with the corresponding RQs and LOCs are summarized in Table G-1.

Table G-1. Agency risk quotient (RQ) metrics and levels of concern (LOC) per risk class.			
Risk Class	**Risk Description**	**RQ**	**LOC**
Aquatic Animals (fish and invertebrates)			
Acute	Potential for effects to non-listed animals from acute exposures	Peak EEC/LC_{50}[1]	0.5
Acute Restricted Use	Potential for effects to animals from acute exposures Risks may be mitigated through restricted use classification	Peak EEC/LC_{50}[1]	0.1
Acute Listed Species	Listed species may be potentially affected by acute exposures	Peak EEC/LC_{50}[1]	0.05
Chronic	Potential for effects to non-listed and listed animals from chronic exposures	60-day EEC/NOEC (fish) 21-day EEC/NOEC (invertebrates)	1
Terrestrial Animals (mammals and birds)			
Acute	Potential for effects to non-listed animals from acute exposures	EEC[2]/LC_{50} (Dietary) EEC/LD_{50} (Dose)	0.5
Acute Restricted Use	Potential for effects to animals from acute exposures Risks may be mitigated through restricted use classification	EEC[2]/LC_{50} (Dietary) EEC/LD_{50} (Dose)	0.2
Acute Listed Species	Listed species may be potentially affected by acute exposures	EEC [2]/LC_{50} (Dietary) EEC/LD_{50} (Dose)	0.1
Chronic	Potential for effects to non-listed and listed animals from chronic exposures	EEC [2]/NOAEC	1
Plants			
Non-Listed	Potential for effects to non-target, non-listed plants from exposures	EEC/ EC_{25}	1
Listed Plant	Potential for effects to non-target, listed plants from exposures	EEC/ NOEC EEC/ EC_{05}	1

[1] LC_{50} or EC_{50}. [2] Based on upper bound Kenaga values.

Appendix H. List of citations accepted and rejected by ECOTOX criteria.

The citations in this appendix were accepted by ECOTOX. Citations include the ECOTOX Reference number. References in section H.1 those relevant to diazinon which were cited within this risk assessment. References in section H.2 were those relevant to diazinon which were not cited within the risk assessment. References in section H.3 those relevant to degredates of diazinon which were cited within this risk assessment. References in section H.4 were those relevant to degredates of diazinon which were not cited within the risk assessment. In order to be included in the ECOTOX database, papers must meet the following minimum criteria:

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

Section H.5 includes the list of exclusion terms and descriptions for citations not accepted by ECOTOX. For diazinon, there were hundreds of references that were not accepted by ECOTOX for one or more of the reasons included in section H.5. A full list of the citations reviewed and rejected by the criteria for ECOTOX is listed in section H.6.

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H.5. List of exclusion terms utilized for reviewing studies considered for ECOTOX database

Review--all toxicity tests reported elsewhere. If the publication is applicable to one of the ECOTOX databases, the bibliography is skimmed and any applicable articles are ordered.

Methods--no usable toxicity tests. Reports of methods of conducting tests, determination or purification of chemicals, etc. Methods publications are selected to be ordered for the ECOTOX toxicology methods information file (Methfile).

Modeling only, no new organism exposure data. Modeling studies may report original toxicity tests performed as comparisons or as a basis for extrapolation; order the paper if it is not clear from the abstract.

Other ambient conditions--effects on organisms from changes in conditions other than addition of chemicals, including radioactivity, ultraviolet light (UV), temperature, pH, salinity, dissolved oxygen (DO), or other water, air, or soil parameters.

Biological Toxicant--includes venoms, fungal toxins, *Bacillus thuringiensis*, other plant, animal, or microbial extracts or toxins.

Drug--testing for drug effects and side-effects .

Effluent, sewage, or polluted runoff.

Mixture--no single chemical tests reported.

Nutrient studies--in situ chemicals tested as nutrients.

No Species--no organism present or tested or unable to verify a species or exposure of dead organism.

In Vitro studies, including exposure of cell cultures and excised tissues.

Bacteria as test organism, including **Microtox** tests, or other microbial organisms.

Yeast as a test organism is historically not coded in ECOTOX.

No Toxicity **Data**--publications which are not toxicology studies.

Human Health effects; studies with human subjects or with animal subjects as surrogates for human health risk assessment.

No Concentration--no usable dose or concentration reported; identified after examination of full paper. Includes lead-shot studies which lack dose information or give only number of pellets. Concentrations reported only in log units are not coded.

Sediment Concentration--chemical concentration reported in sediment only. Sediment studies are coded for AQUIRE only if a water concentration of the added chemical is also reported; order the publication if unclear from the abstract.

No Duration reported, identified after examination of full paper.

Incident papers--reports of animal deaths by poison, etc. Lacks usable concentration or duration or both.

Survey studies--measuring amounts of chemical present, but no usable quantification of exposure. Lacks either usable concentration or duration or both.

Fate: Studies reporting only what happens to the chemical in abiotic matrices

Food Studies, no chemical and effects information are reported

PUBL AS, author has results were published in a different format. For example, may be used for a Ph.D. dissertation when the same results were also published in a peer-reviewed journal.

NON-ENGLISH or **FORE**, paper was published in a foreign language.

Appendix I. Individual Effect Analysis.

As discussed in the effects assessment section of the chapter, OPP conducted an analysis of U.S.G.S. data used to support the Mayer and Ellerseick data set. The analysis included 48-hr acute toxicity data for freshwater aquatic invertebrates including *Simocephalus serrulatus*, *Daphnia pulex*, *Gammarus fasciatus* and *Pteronarcys californica* (**Table I1**). Across the four species, the 48-hr probit dose response slope ranged from 5.74 to 6.90; the mean slope and standard error of the mean were 6.34 and 0.21, respectively. Since a probit dose-response slope is not available for the most the most sensitive species, *i.e.*, *Ceriodaphnia dubia*, the mean slope of 6.34 will be used in the analysis of potential individual effects discussed below.

Table I1. Acute 48-hr and 96-hr LC_{50} values for freshwater aquatic invertebrates based on USGS data used in support of Mayer and Ellerseick.

Species	48-hr LC_{50} (95% CI)	Slope	96-hr LC_{50}	Slope
Simocephalus serrulatus	1.34 (1.00 – 1.71)	6.9	no data	--
S. serrulatus	1.67 (1.31 – 2.16)	6.71	no data	--
Daphnia pulex	0.79 (0.58 – 1.02)	6.20	no data	--
Gammarus fasciatus	4.71 (3.69 – 6.11)	6.13	1.99 (1.48 – 2.63)	4.67
Pteronarcys califonica	59.4 (42.5 – 83.3)	5.74	20.5	22.7

Likelihood of individual acute effects to freshwater invertebrates based on maximum application rate with 26 application per year.

IEC V1 - Individual Effect Chance Model Version 1
Predictor of chance of individual effect using probit dose-response curve slope and median lethal estimate

Enter LC_{50} or LD_{50}	0.21	Note: This is not used in calculation, just serves as a reminder to user
Enter desired threshold	0.27	Note: This is either the RQ fraction of the toxicity endpoint, the EEC or dose fraction of the dose/concentration at tox endpoint, or the LOC
Enter slope of dose-response	6.3	Note: This is the slope of the dose response relationship from the study providing the above endpoint
z score result	-3.58240829	z is the standard normal deviate
Probability associated with z	0.00017022	Uses Excel NORMDIST function to estimate P
Chance of individual effect, ~1 in . . .	5.87E+03	Calculated as 1/P rounded to 0 decimals

This is based on the formula $\log LC_k = \log LC_{50} + (z/b)$
where: z is the standard normal deviate and b equals slope
Works for dose-response models based on a probit assumption (i.e. log normal distribution of individual sensitivity)
Note: Probability asociated with z value may be reported as "0". This is due to the inability of Excel to handle extremes in z scores beyond -8.2
In such cases the chance of individual effect is defaulted to 1 in 10^{16}, which is the limit of Excel reporting.

Ed Odenkirchen, May 28, 2003 EFED/OPP/USEPA

Figure I1. Estimation of likelihood on individual mortality based on risk quotients for freshwater invertebrates (RQ=0.27) following 26 applications per year to ornamentals. . Estimated dose-response slope is 6.3.

Likelihood of an individual acute effects to freshwater invertebrates based on maximum application rate and a single application per year.

IEC V1 - Individual Effect Chance Model Version 1
Predictor of chance of individual effect using probit dose-response curve slope and median lethal estimate

Enter LC_{50} or LD_{50}	0.21	Note: This is not used in calculation, just serves as a reminder to user
Enter desired threshold	0.08	Note: This is either the RQ fraction of the toxicity endpoint, the EEC or dose fraction of the dose/concentration at tox endpoint, or the LOC
Enter slope of dose-response	6.3	Note: This is the slope of the dose response relationship from the study providing the above endpoint
z score result	-6.91053308	z is the standard normal deviate
Probability associated with z	2.4142E-12	Uses Excel NORMDIST function to estimate P
Chance of individual effect, ~1 in . . .	4.14E+11	Calculated as 1/P rounded to 0 decimals

This is based on the formula $\log LC_x = \log LC_{50} + (z/b)$
where: z is the standard normal deviate and b equals slope
Works for dose-response models based on a probit assumption (i.e. log normal distribution of individual sensitivity)
Note: Probability asociated with z value may be reported as "0". This is due to the inability of Excel to handle extremes in z scores beyond -8.2
In such cases the chance of individual effect is defaulted to 1 in 10^{16}, which is the limit of Excel reporting.

Ed Odenkirchen, May 28, 2003 EFED/OPP/USEPA

Figure I2. Estimation of likelihood of individual mortality based on risk quotients for freshwater invertebrates (RQ=0.08) following a single application of diazinon per year to ornamentals. Estimated dose-response slope is 6.3.

Appendix J. The Generalized Barton Springs Refined Modeling Approach.

J.1 Background

The Barton Springs are supplied predominantly with water discharging from fractures and conduits formed in the Barton Springs Segment of the Edwards Aquifer (BSSEA) as a result of dissolution of the fractured limestone aquifer over time. Slade *et al*. (1986) estimated that approximately 85% of the water that recharges this aquifer infiltrates through the beds of six creeks that cross the recharge zone (Slade *et al*. 1985, Barrett and Charbeneau 1996), with the remaining approximately 15 % of the recharge derived from precipitation and recharge in interbed areas in the recharge zone. In the BSSEA, natural ground water discharge occurs primarily at Barton Springs (Lindgren *et al*., 2004). Recharge features in creek bottoms overlying the recharge zone allow only a limited flow of water during a storm event; therefore, water that is in excess of the flow capacities of recharge features leaves the recharge zone as creek flow. The contributing zone encompasses the watersheds of the upstream portions of the six major creeks that cross the recharge zone and therefore provides the source for most of the water that enters the BSSEA as recharge. These streams gain water, as they flow across the land surface in the contributing zone, from the lower-permeability Glen Rose limestone of the adjacent Trinity aquifer (Lindgren *et al*., 2004). Kuniansky (1989) estimated baseflow discharge from the Trinity aquifer to streams and creeks in this area ranging from 25% to 90% of total flow. In the portion of the Trinity aquifer nearest the contributing zone this was loosely estimated at 30%. The remainder of water in creeks in the contributing zone is derived from precipitation and runoff.

J.2 Model Outline

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

Masses and volumes of runoff are determined for this assessment from modeling scenarios developed specifically for the various land uses (*e.g.*, orchards, nurseries, vineyards, residential) found in the Barton Springs Salamander action area. Similar to the Agency's standard ecological risk assessment methodology described above, 30 years of meteorological data were linked to these specific scenarios to estimate 1-in-10-year edge-of-field exposure to potential diazinon uses.

J.3 Determination of Runoff Concentrations and Volume

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

J.3.1 Contributing Zone

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

$$V_{CZ} = \sum_{t=1}^{n}\left(\sum_{i=1}^{m}\left(V_{CZ,i,t}\right)\right) \tag{J.1}$$

where V_{CZ} = 30-year simulated cumulative runoff [volume]
$V_{CZ,i,t}$ = runoff from area i on day t [volume]
n = number of days in simulation
m= number of different areas (*e.g.*, crop areas) in simulation

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

$$V_{base} = \frac{V_{CZ}}{n}\left(\frac{0.30}{0.70}\right) \tag{J.2}$$

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

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

$$V_{stream,t} = \sum_{i=1}^{m}\left(V_{CZ,i,t}\right) + V_{base} \tag{J.3}$$

where $V_{stream,t}$ = the total stream volume on day t [volume]

Daily stream concentrations were calculated directly from the PRZM out put, the area of the scenario, and the stream base flow as follows:

$$C_{stream,t} = \frac{\sum_{i=1}^{n}\left(M_{CZ,i,t}\right) + M_{base}}{V_{stream,t}} \tag{J.4}$$

where $C_{stream,t}$ = the daily stream concentration [mass/volume]
$M_{CZ,i,t}$ = mass of runoff for scenario i on day t in contributing zone [mass]
M_{base} = daily average mass in stream base flow [mass]

The above calculated stream volume ($V_{stream,t}$) in **equation J.3** along with its associated concentration ($C_{stream,t}$) in **equation J.4** are assumed to be delivered to the recharge zone where they mix with recharge zone runoff as described next.

J.3.2 Recharge Zone

Runoff originating in the recharge zone was determined in a similar manner as for the contributing zone using PRZM output as follows:

$$V_{RZ,t} = \sum_{i=1}^{m} \left(V_{RZ,i,t} \right) \quad \textbf{(J.5)}$$

where $V_{RZ,t}$ = total daily runoff in recharge zone [volume]
$V_{RZ,i,t}$ = runoff from area i on day t [volume]
m = number of different areas (*e.g.*, crop areas) in simulation

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

$$C_{RZ,t} = \frac{\sum_{i=1}^{n} \left(M_{i,t} \right)}{V_{RZ,t}} \quad \textbf{(J.6)}$$

where $C_{RZ,t}$ = daily recharge zone runoff concentration [mass/volume]
$M_{RZ,i,t}$ = mass of runoff for scenario i on day t in recharge zone [mass]

J.4 Barton Springs Daily Concentrations

It is assumed that the stream flow from the contributing area and the runoff from the recharge area mix and flow through the Karst and into Barton Springs. The spring concentration is determined from:

$$C_{Barton,t} = \frac{C_{RZ,t} V_{RZ,t} + C_{stream,t} V_{stream,t}}{V_{RZ,t} + V_{stream,t}} \quad \textbf{(J.7)}$$

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

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

J.5 Special Case: Use area hydrologically similar to non-use area

In the case where a pesticide use area has the same hydrological characteristics as the non-use area, a simplification can be made that gives approximately identical results as the more complicated model described above. For example, in the Barton Springs area of interest, the non-crop use area is modeled with a residential PRZM scenario (predominantly characterized by a curve number of 85). If a sole use area is also modeled with the same residential scenario, then runoff would occur from both the use area and the non-use areas in an identical manner.

Consider now, the Barton Springs calculation (**equation J.7** above). This equation can be rewritten as:

$$C_{Barton,t} = \frac{M_{RZ,non-use,t} + M_{RZ,use,t} + M_{CZ,non-use,t} + M_{CZ,use,t} + M_{base,t}}{V_{RZ,non-use,t} + V_{RZ,use,t} + V_{CZ,non-use,t} + V_{CZ,use,t} + V_{base,t}} \qquad \textbf{(J.8)}$$

For the 30-year simulation of the watershed area, less than 9 of the 569 runoff events produced runoff from the area that had a volume of less than 10 times the calculated stream base flow. This means that the volume of the base stream flow is negligible in nearly every event in comparison to runoff volume. In the unlikely case that a high pesticide concentration would occur from one of these rare events (1.6% of runoff events) then such an event would be screened out by the EPA practice of selecting the 90th percentile reoccurrence event. Therefore for practical purposes, the base volume can be eliminated from the above equation. Additionally, since all the runoff volumes are generated from the same scenario with only area differing among them and if base stream concentrations can be assumed to be negligible, then **equation A.8** can be rewritten as

$$C_{Barton,t} = \frac{\left(M_{A,t}\right)\left(A_{CZ,use} + A_{RZ,use}\right)}{D_t\left(A_{CZ,non-use} + A_{CZ,use} + A_{RZ,non-use} + A_{RZ,use}\right)} \qquad \textbf{(J.9)}$$

where $M_{A,t}$ = daily PRZM output for pesticide mass [mass/area]
D_t = daily PRZM output for runoff depth [length]
$A_{CZ,i}$ = extent of i area in contributing zone [area]
$A_{RZ,i}$ = extent of i area in recharge zone [area]

Therefore, the Barton Springs concentration can be determined by the PRZM edge-of-field concentration times the ratio of use area to total area:

$$C_{Barton,t} = C_{edge}\frac{A_{use}}{A_{total}} \qquad \textbf{(J.10)}$$

where C_{edge} = PRZM edge of field concentration [mass/volume]
A_{use} = total use area [area]
A_{total} = total Barton Springs watershed area [area]

The above simplified model equation (**J.10**) can be used where the use and non-use areas can be described by the same PRZM scenario and where background concentrations are not present.