Risks of Atrazine Use to Three Federally Listed Endangered Freshwater Mussels

August 31, 2007

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Fat Pocketbook Pearly Mussel (*Potamilus capax*),
Purple Cat's Paw Pearlymussel (*Epioblasma obliquata obliquata*), and
Northern Riffleshell (*Epioblasma torulosa rangiana*)

Pesticide Effects Determination

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1. Executive Summary

The purpose of this assessment is to make an "effects determination" by evaluating the potential direct and indirect effects of the herbicide atrazine on the survival, growth, and reproduction of the following three Federally listed species of freshwater mussels: fat pocketbook pearly mussel (*Potamilus capax*), purple cat's paw pearlymussel (*Epioblasma obliquata obliquata*) (PCPP mussel), and northern riffleshell (*Epioblasma torulosa rangiana*). 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).

Atrazine is used throughout the United States on a number of agricultural commodities (primarily corn and sorghum) and on non-agricultural sites (including residential uses, forestry, and turf). Although the action area is likely to encompass a large area of the United States, given its use, the scope of this assessment limits consideration of the overall action area to those portions that are applicable to the protection of the three listed mussels. As such, the action area includes the current range of the species, which occur in streams and rivers within a wide geographic range from Louisiana, north along the Mississippi River Valley, to the lower Missouri River Valley, northwest into Iowa, and east along the Ohio River Valley extending into Pennsylvania and West Virginia. In general, the species are found in streams and rivers within the Mississippi, Missouri, and Ohio River watersheds.

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

Throughout the assessment, a semi-quantitative comparison is made between Strahler stream order, stream flow rates, and species' locations. In general, the fat pocketbook mussel is found in streams ranging from 1^{st} to 7^{th} order (this is uncertain due to poor location information) with flow rates ranging from approximately 100 ft^3/s to 600,000 ft^3/s (using all mean stream flow data from the Enhanced Reach File version 1_2 used to create species maps). The PCPP mussel is found in streams ranging from 3^{rd} to 5^{th} order with flow rates between approximately 5,000 ft^3/s and 17,000 ft^3/s. The northern riffleshell mussel is found in streams ranging between 2^{nd} and 4^{th} order and with flow rates between approximately 100 ft^3/s and 16,000 ft^3/s.

Although an increase in flow rate is generally observed within a watershed as the stream order increases, there is no direct relationship between stream order and flow rate. For example, flow rates in two first order streams in different watersheds may have vastly different flow rates. Although both flow rate and stream order are discussed in this assessment, in the context of relating exposures to species' location, flow rate (not stream order) is the primary predictor of where exposures are expected to be above or below the LOC.

In estimating potential exposure to the listed mussels, a combination of modeling and monitoring data were considered. Modeling was conducted as part of this assessment using both static and flowing waters. The screening-level static water estimated environmental concentrations (EECs) were used for risk estimation, while refined EECs using flow rates from occupied streams were used to characterize risk in the risk description. Flow rates used in the refined modeling represent the range of occupied streams described above and generally predict long term concentrations that are orders of magnitude lower than those seen in modeling with the static water body. In addition, available monitoring data indicate that LOCs may be exceeded in watersheds where flow rates are less than 200 ft^3/s and atrazine use is high (e.g. vulnerable watersheds sampled as part of the Atrazine Ecological Monitoring Program or AEMP).

In accordance with the methodology specified in the Agency's Overview Document (U.S. EPA, 2004), screening-level estimated environmental concentrations (EECs), based on the PRZM/EXAMS static water body scenario, were used to derive RQs for all relevant agricultural and non-agricultural atrazine uses within the action area. RQs based on screening-level EECs were used to distinguish "no effect" from "may effect" determinations for direct/indirect effects to the listed mussels. However, screening-level EECs based on the static water body are not considered to be representative of flowing waters where the assessed mussels occur. For "may affect" determinations, screening-level EECs were further refined and characterized, as follows, based on site-specific flow information and the location of the assessed mussels within or outside the boundary of vulnerable watersheds.

- The most vulnerable watersheds to atrazine runoff are defined as the top 20th percentile based on model predictions using the WARP model.[1] These watersheds represent the locations where atrazine exposures in 2nd and 3rd order streams are expected to be highest. Preliminary results from the Atrazine Ecological Monitoring Program (AEMP) indicate that some proportion of waters within this area are above the LOC. Targeted monitoring data from the AEMP were used to refine the screening-level EECs for fat pocketbook and northern riffleshell mussels in occupied streams within the vulnerable watershed boundary that have flow rates < 200 ft^3/sec or for which no flow rate information is available.

- Flow-adjusted EECs and available non-targeted monitoring data (i.e., the study design is not specifically targeted to detect peak atrazine exposures in high use areas) were used to refine screening-level EECs for the PCPP mussel (because

[1] Watershed Regression of Pesticides model (USGS 2005) at http://pubs.usgs.gov/circ/2005/1291/

stream flow data, which is available for all occupied streams, suggests that this species requires a higher flow rate than those represented by data from the targeted AEMP), and fat pocketbook and northern riffleshell mussels that are either located outside the boundary of vulnerable watersheds and/or within vulnerable watersheds in larger rivers and streams with flow rates > 200 ft^3/sec.

Therefore, separate effects determinations were derived for direct/indirect endpoints based on flow requirements and the location of the assessed species within highly vulnerable and less vulnerable watersheds of the action area. A flow rate of 200 ft^3/sec was chosen as the threshold for use of targeted versus non-targeted monitoring data because the targeted AEMP data are representative of only a small subset of occupied streams with flow below the 15^{th} percentile of flow from occupied streams (or occupied streams with flow rates < 200 ft^3/sec).

The assessment endpoints for the listed mussels include direct toxic effects on survival, reproduction, and growth of individual mussels, as well as indirect effects, such as reduction of the food source or perturbation of host fish, and/or modification of habitat. Acute toxicity data on freshwater mussels are available and were utilized for RQ calculations. However, chronic RQs were derived using data on the closest taxonomic group with available toxicity data (freshwater invertebrates).

Given that the mussel's food source and habitat requirements are dependant on the availability of freshwater fish, aquatic plants, freshwater invertebrates, and terrestrial plants (i.e., riparian habitat), toxicity information for these taxonomic groups is also discussed. In addition to the registrant-submitted and open literature toxicity information, indirect effects, via impacts to aquatic plant community structure and function, are also evaluated based on time-weighted threshold concentrations that correspond to potential aquatic plant community-level effects.

Effects determinations for direct/indirect effects to the three listed mussels, by assessment endpoint, are presented in Table 1.1. In addition, Table 1.2 provides a summary of the direct and indirect effects determinations for each of the three listed mussels. Effects determinations for this assessment are summarized below.

- An "LAA" determination was concluded for indirect prey and habitat effects to fat pocketbook and northern riffleshell mussels that occur in streams within the vulnerable watershed boundary that have flow rates < 200 ft^3/sec or for which no flow rate information is available, based on potential direct aquatic plant community-level effects.
    - The "LAA" determination is based on the results of recently submitted atrazine monitoring data from vulnerable watersheds; however, the degree to which this targeted monitoring data represents exposures in occupied streams (for the fat pocketbook and northern riffleshell) that co-occur with lower flowing vulnerable watersheds is not available. For the purposes of this assessment, it is conservatively assumed that detected concentrations of atrazine from the AEMP monitoring data may be representative of

exposures in lower flow (i.e., < 200 ft^3/sec) vulnerable watersheds of the action area.

  - If further analysis reveals that the AEMP monitoring data are not representative of atrazine concentrations in vulnerable watersheds where the fat pocketbook and northern riffleshell mussels occur, the "LAA" effects determination will be revisited and could be changed to "NLAA" for these species.

- An "LAA" determination was concluded for the fat pocketbook mussel based on indirect effects to habitat and water quality via direct effects to herbaceous/grassy riparian vegetation. However, atrazine is not likely to adversely affect the fat pocketbook mussel in watersheds with predominantly forested riparian areas because woody shrubs and trees are generally not sensitive to environmentally-relevant concentrations of atrazine. In addition, atrazine-related impacts to riparian areas adjacent to large rivers occupied by the fat pocketbook are expected to be insignificant, based on a spatial analysis of land cover data adjacent to occupied rivers. Potential indirect effects to the PCPP mussel and northern riffleshell via atrazine-related impacts to riparian vegetation adjacent to the occupied streams/rivers are also not expected, based on an analysis of land cover and county-level use data[2], as well as aerial satellite photography. Therefore, the effects determination for the fat pocketbook mussel located in watersheds with predominantly forested vegetation (including big rivers), and for the PCPP mussel and northern riffleshell in all occupied watersheds is "NLAA".

[2] County-level data was obtained from http://www.fedstats.gov, and http://www.ams.usda.gov/statesummaries.

Table 1.1 Effects Determination Summary for the Assessed Listed Mussels (by Assessment Endpoint)				
Direct and Indirect Effects to Listed Mussels				
Assessment Endpoints for Aquatic Animals and Plants	**Effects Determination and Basis for PCPP Mussel (in all occupied streams) and Fat Pocketbook and Northern Riffleshell Mussels (located in less vulnerable watersheds and larger river/streams with flow > 200 ft^3/sec in vulnerable watersheds)**		**Effects Determination and Basis for Fat Pocketbook and Northern Riffleshell (located in highly vulnerable watersheds with stream flow < 200 ft^3/sec or for which no flow data is available)**	
	Effects Determination[a]	**Basis**	**Effects Determination[a]**	**Basis**
1. Survival, growth, and reproduction of assessed mussel individuals via direct acute or chronic effects	Acute direct effects: NE	No acute LOCs are exceeded.	Acute direct effects: NE	No acute LOCs are exceeded.
	Chronic direct effects: NLAA	Chronic LOCs are exceeded based on screening-level EECs; however, RQs based on flow-adjusted EECs and non-targeted monitoring data are less than concentrations shown to cause adverse effects in freshwater mollusks. This finding is based on discountable effects (i.e., chronic effects at refined levels of exposure are not likely to occur and/or result in "take" of a single listed mussel).	Chronic direct effects: NLAA	Chronic LOCs are exceeded based on screening-level EECs; however detected concentrations of atrazine in monitoring data from vulnerable watersheds are less than those shown to cause adverse effects in freshwater mollusks. This finding is based on discountable effects (i.e., chronic effects to atrazine at refined levels of exposure are not likely to result in "take" of a single fat pocketbook and northern riffleshell mussel located in highly vulnerable watersheds).
2. Indirect effects to assessed mussel individuals via reduction in food items (i.e., freshwater phytoplankton and zooplankton)	Phytoplankton: NLAA	Individual aquatic plant species may be affected. However, refined 14-, 30-, 60- and 90-day EECs, which consider the impact of flow and non-targeted monitoring data, are less than the threshold concentrations representing community-level effects. This finding is based on insignificance of effects (i.e., community-level effects to aquatic plants cannot be meaningfully measured, detected, or evaluated in the context of a "take" of a single listed mussel via a reduction in food items).	Phytoplankton: LAA[b]	Individual aquatic plant species within vulnerable watersheds of the action area may be affected. 14-, 30-, 60-, and 90- day rolling averages, based on the AEMP data, exceed their respective threshold concentrations for 5 to 12.5% of the sampled vulnerable watersheds. Therefore, community-level effects are possible for phytoplankton, resulting in indirect effects to the food supply of the fat pocketbook and northern riffleshell mussels, within lower flow (< 200 ft^3/sec) vulnerable watersheds of the action area.

	Acute direct effects to zooplankton: NLAA	Acute LOCs are exceeded based on screening-level EECs and the most sensitive freshwater invertebrate toxicity data. Based on the refined analysis, which considered flow-adjusted EECs, non-targeted monitoring data, and effects data specific to zooplankton, acute effects to zooplankton are not likely to occur at refined levels of exposure. Effects are discountable because refined exposures are not likely to cause adverse effects to zooplankton and the probability of an individual effect to zooplankton is low (i.e., 0.03%). Effects are also insignificant because the level of effect at predicted levels of exposure is low (i.e., <2%) and zooplankton are not the primary food source for listed mussels. Therefore, "take" of a single listed mussel is not expected to occur).	Acute direct effects to zooplankton: NLAA	Acute LOCs are exceeded based on screening-level EECs and the most sensitive freshwater invertebrate toxicity data. Based on the refined analysis, which considered flow-adjusted EECs, non-targeted monitoring data, and effects data specific to zooplankton, acute effects to zooplankton are not likely to occur at refined levels of exposure. Effects are discountable because refined exposures are not likely to cause adverse effects to zooplankton and the probability of an individual effect to zooplankton is low (i.e., 0.03%). Effects are also insignificant because the level of effect at predicted levels of exposure is low (i.e., <2%) and zooplankton are not the primary food source for listed mussels. Therefore, "take" of a single listed fat pocketbook and northern riffleshell mussel is not expected to occur).
	Chronic direct effects to zooplankton: NLAA	Chronic LOCs are exceeded based on screening-level EECs and the most sensitive freshwater invertebrate toxicity data. However, all refined measures of exposure (21-day flow-adjusted EECs and non-targeted monitoring data) are well below levels of chronic effects in cladocerons. This finding is based on discountable effects (i.e., chronic effects to atrazine at refined levels of exposure are not likely to occur and/or result in a "take" of a single listed mussel via a reduction in zooplankton as food items).	Chronic direct effects to zooplankton: NLAA	Chronic LOCs are exceeded based on screening-level EECs and the most sensitive freshwater invertebrate toxicity data. However, 21-day rolling averages based on the ecological monitoring data are well below levels of chronic effects in cladocerons. This finding is based on discountable effects (i.e., chronic effects to atrazine in highly vulnerable watersheds are not likely to occur and/or result in a "take" of a single fat pocketbook and northern riffleshell mussel via a reduction in zooplankton as food items).

3. Indirect effects to assessed mussel individuals via reduction in host fish for mussel glochidia (i.e., larvae)	Acute direct effects to host fish: NE	No acute LOCs are exceeded.	Acute direct effects to host fish: NE	No acute LOCs are exceeded.
	Chronic direct effects to host fish: NLAA	Chronic LOCs are exceeded based on screening-level EECs; however refined flow-adjusted EECs and non-targeted monitoring data are not likely to result in adverse chronic effects to fish. This finding is based on discountable effects (i.e., chronic exposure to atrazine is not likely to result in "take" of a single listed mussel because direct chronic effects to host fish are unlikely to occur).	Chronic direct effects to host fish: NLAA	Chronic LOCs are exceeded based on screening-level EECs; however, detected concentrations of atrazine in monitoring data from vulnerable watersheds are not likely to result in adverse chronic effects to fish. This finding is based on discountable effects (i.e., chronic exposure to atrazine is not likely to result in "take" of a single fat pocketbook and northern riffleshell because direct chronic effects to host fish in vulnerable watersheds are unlikely to occur).
4. Indirect effects to assessed mussel individuals via direct effects to aquatic plants (i.e., reduction of habitat and/or primary productivity)	Direct effects to aquatic plants: NLAA	Individual aquatic plant species may be affected. However, flow-adjusted 14-, 30-, 60-, and 90-day EECs and similar durations of exposure based on non-targeted monitoring data, are less than the threshold concentrations representing community-level effects. This finding is based on insignificance of effects (i.e., community-level effects to aquatic plants cannot be meaningfully measured, detected, or evaluated in the context of a "take" of a single listed mussel via direct effects on habitat and primary productivity).	Direct effects to aquatic plants: LAA[b]	Individual aquatic plant species within vulnerable watersheds of the action area may be affected. 14-, 30-, 60-, and 90- day rolling averages based on the AEMP data from vulnerable watersheds exceed their respective threshold concentrations for a small percentage of the data set. Therefore, community-level effects are possible for phytoplankton, resulting in indirect effects to the fat pocketbook and northern riffleshell, via direct effects on habitat and primary productivity, within lower flow (< 200 ft^3/sec) vulnerable watersheds of the action area.
Assessment Endpoints for Terrestrial Plants	**Effects Determination[a]**	**Basis**	**Effects Determination[a]**	**Basis**
5a. Indirect effects to *fat pocketbook* individuals via reduction of terrestrial vegetation (i.e., riparian habitat)	Direct effects to forested riparian vegetation: NLAA	Riparian vegetation may be affected because terrestrial plant RQs are above LOCs. However, woody shrubs and trees are generally not sensitive to atrazine; therefore, listed mussels in watersheds with predominantly forested	Direct effects grassy/herbaceous riparian vegetation: LAA	Riparian vegetation may be affected because terrestrial plant RQs are above LOCs. The LAA effects determination for listed mussels that are in close proximity to grassy/herbaceous riparian areas is based on the sensitivity of herbaceous vegetation to atrazine. Until further analysis on specific land

required to maintain acceptable water quality and habitat[c]		riparian vegetation (i.e., woody shrubs and trees) are not likely to adversely affected. This finding is based on insignificance of effects (i.e., effects to forested riparian vegetation in the action area are not likely to result in "take" of a single listed mussel).		management practices and sensitivity of grassy riparian vegetation adjacent to fat pocketbook mussel habitat is completed, potential indirect effects via sedimentation are presumed to adversely affect the fat pocketbook.
	Indirect effects to fat pocketbook mussels that occur in big rivers: NLAA	Land cover data from seven example watersheds (i.e., big rivers including the Big Sunflower River in Mississippi, the Wabash River in Illinois, the White River and Lower Ohio River in Indiana, the Upper Ohio River in Kentucky, and the St. Francis and White Rivers in Arkansas) indicates that the majority of riparian vegetation directly adjacent to occupied rivers is comprised of deciduous forest and woody wetlands that are not sensitive to atrazine at environmentally relevant concentrations. Therefore, potential indirect effects via atrazine-related impacts to riparian areas adjacent to large rivers occupied by the fat pocketbook are expected to be insignificant (i.e., cannot be meaningfully measured, detected or evaluated in the context of a level of effects where "take" occurs for a single fat pocketbook).		
5b. Indirect effects to *PCPP mussel* and *northern riffleshell* individuals via reduction of terrestrial vegetation (i.e., riparian habitat) required to maintain acceptable water quality and habitat[d]	NLAA	Land cover and land use data (as well as aerial satellite imagery) surrounding the occupied streams/rivers of the PCPP mussel and northern riffleshell suggest that the predominant riparian area adjacent to occupied watersheds is not likely to be sensitive to atrazine and/or riparian vegetation exposure to atrazine is expected to be minimal. Therefore, potential indirect effects via atrazine-related impacts to riparian areas adjacent to occupied streams/rivers are expected to be insignificant, such that they cannot be meaningfully measured, detected or evaluated in the context of a level of effects where "take" occurs for a single PCPP mussel or northern riffleshell.		

[a] NE = "no effect"; NLAA ‘ "may affect, but not likely to adversely affect"; and LAA = "may affect and likely to adversely affect".

[b] Further analysis of the AEMP data is required to determine the representativeness of the data to other watersheds within vulnerable areas where the listed mussel species occur. If the analysis suggests that the AEMPg data are representative of atrazine concentrations in vulnerable watersheds where the fat pocketbook and northern riffleshell mussels occur, the effects determination will remain as "LAA." However, if further analysis reveals that the monitoring data are not representative of atrazine concentrations in vulnerable watersheds where these listed mussels occur, the effects determination will be revised to "NLAA".

[c] The effects determinations for indirect effects to the fat pocketbook mussel based on direct impacts to riparian habitat is applicable to its entire action area including riparian areas adjacent to both vulnerable and less vulnerable watersheds. Separate effects determinations are based on the presence of forested or herbaceous/grassy riparian vegetation adjacent to the streams and rivers within the fat pocketbook mussel's action area. In addition, a separate effects determination for fat pocketbook mussels located in big rivers was made, based on available land cover data.

[d] Given the limited range of the PCPP mussel and northern riffleshell, an analysis of land cover and county-level use data was completed as part of the effects determination for this endpoint.

<table>
<tr><th colspan="11">Table 1.2 Effects Determination Summary for Each of the Three Assessed Listed Mussels[a]</th></tr>
<tr><th rowspan="3">Assessed Mussel Species</th><th colspan="2">Direct Effects</th><th colspan="8">Indirect Effects</th></tr>
<tr><th rowspan="2">Acute</th><th rowspan="2">Chronic</th><th colspan="2">Food Items</th><th colspan="2">Host Fish</th><th rowspan="2">Aquatic Habitat: community-level effects</th><th colspan="2">Riparian Vegetation</th><th rowspan="2">Big Rivers[b]</th></tr>
<tr><th>Phytoplankton</th><th>Zooplankton</th><th>Acute</th><th>Chronic</th><th>Herbaceous/ Grassy</th><th>Forested</th></tr>
<tr><td>Fat Pocketbook</td><td>NE</td><td>NLAA</td><td>LAA[c]</td><td>NLAA</td><td>NE</td><td>NLAA</td><td>LAA[c]</td><td>LAA</td><td>NLAA</td><td>NLAA</td></tr>
<tr><td>Purple Cat’s Paw Pearlymussel</td><td>NE</td><td>NLAA</td><td>NLAA</td><td>NLAA</td><td>NE</td><td>NLAA</td><td>NLAA</td><td colspan="3">NLAA</td></tr>
<tr><td>Northern Riffleshell</td><td>NE</td><td>NLAA</td><td>LAA[c]</td><td>NLAA</td><td>NE</td><td>NLAA</td><td>LAA[c]</td><td colspan="3">NLAA</td></tr>
</table>

[a] NE = “no effect”; NLAA ‘ “may affect, but not likely to adversely affect”; and LAA = “may affect and likely to adversely affect”. See Table 1.1 for the basis of the effects determinations for each of the assessed mussel species.

[b] Big Rivers include the Big Sunflower River in Mississippi, the Wabash River in Illinois, the White River and Lower Ohio River in Indiana, the Upper Ohio River in Kentucky, the St. Francis and White Rivers in Arkansas, and other similarly sized watersheds where the fat pocketbook mussel occurs.

[c] This LAA determination applies to populations of the fat pocketbook and northern riffleshell that are located in highly vulnerable watersheds with stream flow < 200 ft^3/sec or for which no data are available. Further analysis of the AEMP data is required to determine the representativeness of the data to other watersheds within vulnerable areas where the fat pocketbook and northern riffleshell mussels occur. If the analysis suggests that the AEMP monitoring data are representative of atrazine concentrations in vulnerable watersheds where these listed mussels occur, the effects determination will remain as “LAA.” However, if further analysis reveals that the AEMP monitoring data are not representative of atrazine concentrations in vulnerable watersheds where these listed mussels occur, the effects determination will be revised to “NLAA”.

2. Problem Formulation

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

2.1 Purpose

The purpose of this endangered species risk assessment is to evaluate the potential direct and indirect effects resulting from the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) registered uses of the herbicide atrazine (6-chloro-N-ethyl-N-isopropyl-1, 3, 5-triazine-2, 4-diamine) on the survival, growth, and/or reproduction of individuals of the following three federally listed species of freshwater mussels: (1) fat pocketbook mussel (*Potamilus capax*); (2) purple cat's paw pearlymussel (*Epioblasma obliquata obliquata)* (hereafter referred to as PCPP mussel); and (3) northern riffleshell (*Epioblasma torulosa rangiana*). A summary of the listing status for these species is provided in Table 2.1, and a brief summary of key biological and ecological components related to the assessment of these species is provided in Section 2.5. No critical habitat has been designated for any of the three assessed mussel species. This ecological risk assessment is a component of the settlement for the *Natural Resources Defense Council, Civ. No: 03-CV-02444 RDB (filed March 28, 2006).*

Table 2.1 Identification and Listing Status of Three Listed Freshwater Mussel Species Included in This Assessment

Species	Status[1]	Date Listed
Fat pocketbook pearly mussel (*Potamilus capax*)	Endangered 41 FR 24062-24067	June 14, 1976
Purple cat's paw pearly mussel (*Epioblasma obliquata obliquata*)	Endangered 55 FR 28209-28213	July 10, 1990
Northern riffleshell (*Epioblasma torulosa rangiana*)	Endangered 58 FR 5638-5642	January 22, 1993

[1] All assessed species were listed by the U.S. Fish and Wildlife Service (USFWS).

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

In accordance with the Overview Document, provisions of the Endangered Species Act (ESA), and the Services' *Endangered Species Consultation Handbook*, the assessment of effects of the FIFRA regulatory action is based on a defined action area and the extent of association of this action area with locations of the assessed listed mussels. It is acknowledged that the action area for a national-level FIFRA regulatory decision involving a potentially widely used pesticide may potentially involve numerous areas throughout the United States and its Territories. However, for the purposes of his assessment, attention will be focused on those parts of the action area with the potential to be associated with locations of the assessed listed mussels.

As part of the "effects determination", the Agency will reach one of the following three conclusions regarding the potential for FIFRA regulatory actions regarding atrazine to directly or indirectly affect individuals of the three listed freshwater mussels:

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

If the results of the initial screening-level assessment methods show no direct or indirect effects upon individual listed mussels, a "no effect" determination is made for the FIFRA regulatory action regarding atrazine as it relates to these listed species. If, however, direct or indirect effects to individual listed mussels are anticipated, the Agency concludes a preliminary "may affect" determination for the FIFRA regulatory action regarding atrazine.

If a determination is made that use of atrazine within the action area(s) "may affect" the listed mussels, additional information is considered to refine the potential for exposure at the predicted levels and for effects to the listed mussels and other taxonomic groups upon which these species depend (i.e., freshwater fish and invertebrates, aquatic plants, riparian vegetation). Based on the refined information, the Agency uses the best available information to distinguish those actions that "may affect, but are not likely to adversely affect" ("NLAA") from those actions that are "likely to adversely affect" ("LAA") the three listed mussels. This information is presented as part of the Risk Characterization in Section 5.

2.2 Scope

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

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

This ecological risk assessment is for currently registered uses of atrazine in portions of the action area reasonably assumed to be biologically relevant to the assessed mussel species. Further discussion of the action area(s) for the three listed mussels is provided in Section 2.7.

Degradates of atrazine include hydroxyatrazine (HA), deethylatrazine (DEA), deisopropylatrazine (DIA), and diaminochloroatrazine (DACT). Comparison of available toxicity information for degradates of atrazine indicates lesser aquatic toxicity than the parent for fish, aquatic invertebrates, and aquatic plants. Specifically, the available degradate toxicity data for HA indicate that it is not toxic to freshwater fish and invertebrates at the limit of its solubility in water. In addition, no adverse effects were observed in fish or daphnids at DACT concentrations up to 100 mg/L. Acute toxicity values for DIA are 8.5- and 36-fold less sensitive than acute toxicity values for atrazine in fish and daphnids, respectively. In addition, available aquatic plant degradate toxicity data for HA, DEA, DIA, and DACT report non-definitive EC_{50} values (i.e., 50% effect was not observed at the highest test concentrations) at concentrations that are at least 700 times higher than the lowest reported aquatic plant EC_{50} value for parent atrazine. Although degradate toxicity data are not available for terrestrial plants, lesser toxicity is assumed, given the available ecotoxicological information for other taxonomic groups including aquatic plants and the likelihood that degradates of atrazine may lose efficacy as an herbicide. Therefore, given the lesser toxicity of degradates as compared to the parent, and the relatively small proportion of degradates expected to be in the environment and available for exposure relative to atrazine, the focus of this assessment is parent atrazine. Additional details on available toxicity data for degradates are provided in Section 4 and Appendix A.

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

Atrazine has a number of registered products that contain multiple active ingredients. Analysis of the available open literature and acute oral mammalian LD_{50} data for multiple active ingredient products relative to the single active ingredient is provided in Appendix

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

The results of available toxicity data for mixtures of atrazine with other pesticides are presented in Section A.7 of Appendix A. According to the available data, other pesticides may combine with atrazine to produce synergistic, additive, and/or antagonistic toxic effects. According to the available data, other pesticides may combine with atrazine to produce synergistic or additive toxic effects. Based on the results of the available data, study authors claim that synergistic effects with atrazine may occur for a number of organophosphate insecticides including diazinon, chlorpyrifos, and methyl parathion, as well as herbicides including alachlor. If chemicals that show synergistic effects with atrazine are present in the environment in combination with atrazine, the toxicity of atrazine may be increased, offset by other environmental factors, or even reduced by the presence of antagonistic contaminants if they are also present in the mixture. The variety of chemical interactions presented in the available data set suggest that the toxic effect of atrazine, in combination with other pesticides used in the environment, can be a function of many factors including but not necessarily limited to: (1) the exposed species, (2) the co-contaminants in the mixture, (3) the ratio of atrazine and co-contaminant concentrations, (4) differences in the pattern and duration of exposure among contaminants, and (5) the differential effects of other physical/chemical characteristics of the receiving waters (e.g. organic matter present in sediment and suspended water). Quantitatively predicting the combined effects of all these variables on mixture toxicity to any given taxa with confidence is beyond the capabilities of the available data. However, a qualitative discussion of implications of the available pesticide mixture effects data involving atrazine on the confidence of risk assessment conclusions for the freshwater mussels is addressed as part of the uncertainty analysis for this effects determination.

2.3 Previous Assessments

A summary of the Agency's ecological risk assessments for atrazine is provided in previously submitted effects determinations for 16 listed species (U.S. EPA, 2006c, 2007a, 2007b, and 2007c). In addition, ecological risks associated with exposure of non-target animals and plants to atrazine were evaluated in a 2003 Interim Reregistration Decision (IRED) for atrazine (U.S. EPA, 2003a and b; http://www.epa.gov/oppsrrd1/REDs/0001.pdf).

The Agency also conducted an evaluation of the submitted studies regarding the potential effects of atrazine on amphibian gonadal development and presented its assessment in the form of a white paper for external peer review to a FIFRA Scientific Advisory Panel (SAP) in June 2003[3]. In the white paper dated May 29, 2003, the Agency summarized seventeen studies consisting of both open literature and registrant-submitted laboratory and field studies involving both native and non-native species of frogs (U.S. EPA, 2003d). The Agency concluded that none of the studies fully accounted for

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

environmental and animal husbandry factors capable of influencing endpoints that the studies were attempting to measure. The Agency also concluded that the current lines-of-evidence did not show that atrazine produced consistent effects across a range of exposure concentrations and amphibian species tested.

Based on this assessment, the Agency concluded and the SAP concurred that there was sufficient evidence to formulate a hypothesis that atrazine exposure may impact gonadal development in amphibians, but there were insufficient data to confirm or refute the hypothesis (http://www.epa.gov/oscpmont/sap/2003/June/junemeetingreport.pdf). Because of the inconsistency and lack of reproducibility across studies and an absence of a dose-response relationship in the currently available data, the Agency determined that the data did not alter the conclusions reached in the January 2003 IRED regarding uncertainties related to atrazine's potential effects on amphibians. The SAP supported EPA in seeking additional data to reduce uncertainties regarding potential risk to amphibians. Subsequent data collection has followed the multi-tiered process outlined in the Agency's white paper to the SAP (U.S. EPA, 2003d). In addition to addressing uncertainty regarding the potential use of atrazine to cause these effects, these studies are expected to characterize the nature of any potential dose-response relationship. A data call-in for the first tier of amphibian studies was issued in 2005. The results of these studies, as well as other recent open literature data which focus on the potential effects of atrazine on amphibian gonadal development, are being reviewed. This information will be presented and discussed as part of a second SAP to be held in October 2007.

The Agency has completed four separate effects determinations for atrazine as it relates to 16 of the listed species included in the Natural Resources Defense Counsel settlement agreement and one listed species included in a second settlement agreement with the Center for Biological Diversity and Save Our Springs Alliance. These effects determinations, which are available on the web at www.epa.gov/espp, review atrazine's potential direct and indirect effects to the following listed species: 1) Barton Springs salamander (*Eurycea sosorum*) (U.S. EPA, 2006c); 2) shortnose sturgeon (*Acipenser brevirostrum*), dwarf wedgemussel (*Alasmidonta heterodon*), loggerhead turtle (*Caretta caretta*), Kemp's ridley turtle (*Lepidochelys kempii*), leatherback turtle *(Dermochelys coriacea*), and green turtle (*Chelonia mydas*) in the Chesapeake Bay (U.S. EPA, 2007a); 3) Alabama sturgeon (*Scaphirhynchus suttkusi*) (U.S. EPA, 2007b); and 4) eight listed freshwater mussels including the pink mucket pearly mussel (*Lampsilis abrupta*), rough pigtoe mussel (*Pleurobema plenum*), shiny pigtoe pearly mussel (*Fusconaia edgariana*), fine-rayed pigtoe mussel (*F. cuneolus*), heavy pigtoe mussel (*P. taitianum*), ovate clubshell mussel (*P. perovatum*), southern clubshell mussel (*P. decisum*), and stirrup shell mussel (*Quadrula stapes*) (U.S. EPA, 2007c). The freshwater mussel effects determination also evaluates the potential for atrazine use to result in the destruction or adverse modification of designated critical habitat for the ovate clubshell and southern clubshell mussels. Based on the results of the Barton Springs salamander, Chesapeake Bay, and Alabama sturgeon endangered species risk assessments, atrazine effects determinations for the eight aforementioned listed species are either "no effect" or "may affect, but not likely to adversely affect." In the freshwater mussel assessment, an "LAA" determination was concluded for aquatic plant community-level effects to the pink pearly mucket, rough pigtoe, and fine-rayed pigtoe mussels that occur in highly vulnerable watersheds of the action area. In addition, an "LAA" determination was concluded for the critical habitat impact and indirect effects analysis for all mussels, with the exception of the stirrupshell, based on indirect effects to habitat and water quality via direct effects to herbaceous/grassy riparian vegetation.

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

2.4 Stressor Source and Distribution

2.4.1 Environmental Fate and Transport Assessment

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

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

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

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

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

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

2.4.2 Mechanism of Action

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

2.4.3 Use Characterization

Atrazine is widely used to control broadleaf and many other weeds, primarily in corn, sorghum and sugarcane (U.S. EPA, 2003a). As a selective herbicide, atrazine is applied pre-emergence and post-emergence**.** Figure 2.1 presents the national distribution of use of atrazine (Kaul and Jones, 2006). Table 3.1 presents a summary of all atrazine uses being assessed quantitatively in this assessment.

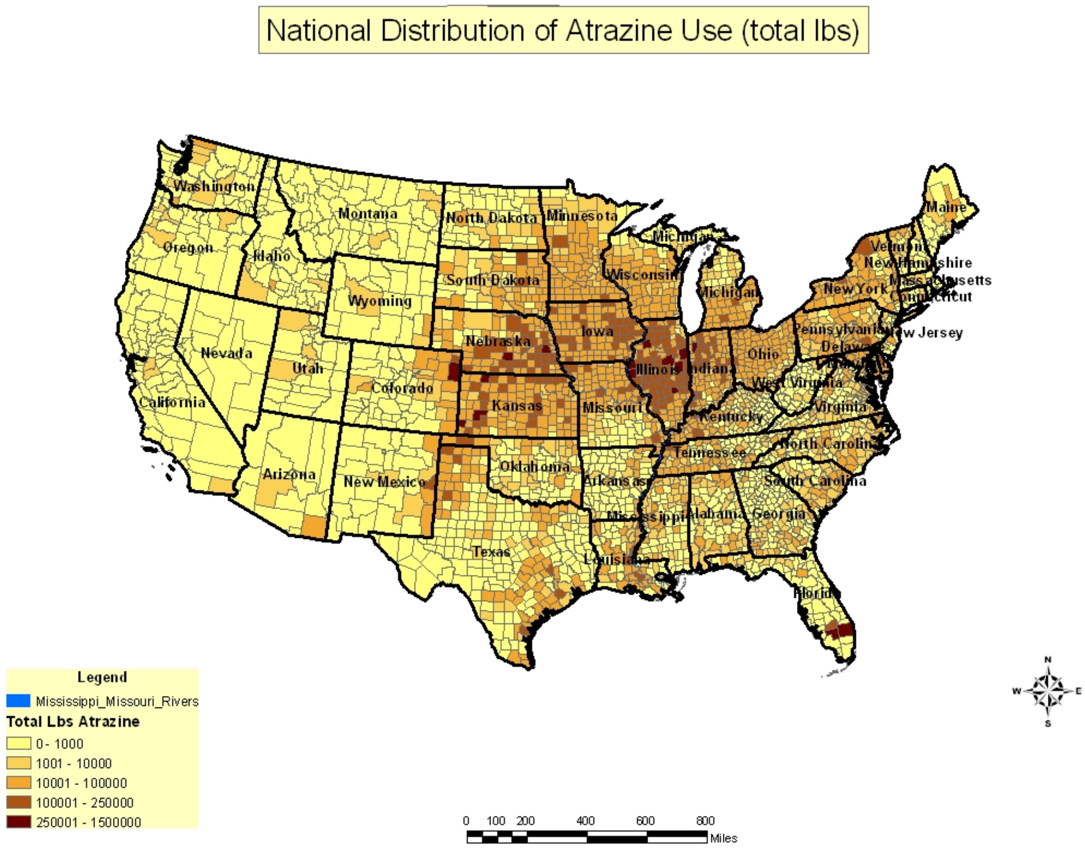

Figure 2.1 National Extent of Atrazine Use (lbs)

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

Assessment of the use information is critical to the development of appropriate modeling scenarios and evaluation of the appropriate model inputs (Kaul and Jones, 2006). Information on the agricultural uses of atrazine in the states comprising the regionalized exposure assessment approach (see Section 3.2.2 for more details) for the three listed mussels (Arkansas, Illinois. Indiana, Iowa, Kentucky, Louisiana, Michigan, Mississippi, Missouri, Ohio, Pennsylvania, Tennessee, and West Virginia), as defined in Section 2.6 of this assessment, was gathered (Kaul and Jones, 2006). In addition, typical atrazine crop use information was considered (Kaul, et al, 2005). Use information within the action area is utilized to determine which uses should be modeled, while the application methods, intervals, and timing are critical model inputs. While the modeling described in Section 3.2 relies initially on maximum label application rates and numbers of applications, information on typical ranges of application rates and number of applications is also presented to characterize the modeling results. No state or county level usage information is available on non-agricultural uses (residential, rights-of-way, forestry, or turf) of atrazine.

Agricultural cropland (presented as cultivated cropland and hay/pasture) and atrazine use relative to the three listed mussel's action area are depicted in Figures 2.2 and 2.3, respectively. The landuse mapping presented in Figure 2.2 provides a breakout of aggregated turf uses (residential, recreational, and golf course). No consistent coverage is available for rights-of-way uses. Given the potential use pattern shown in Figure 2.2, atrazine could be used in close proximity to the species range.

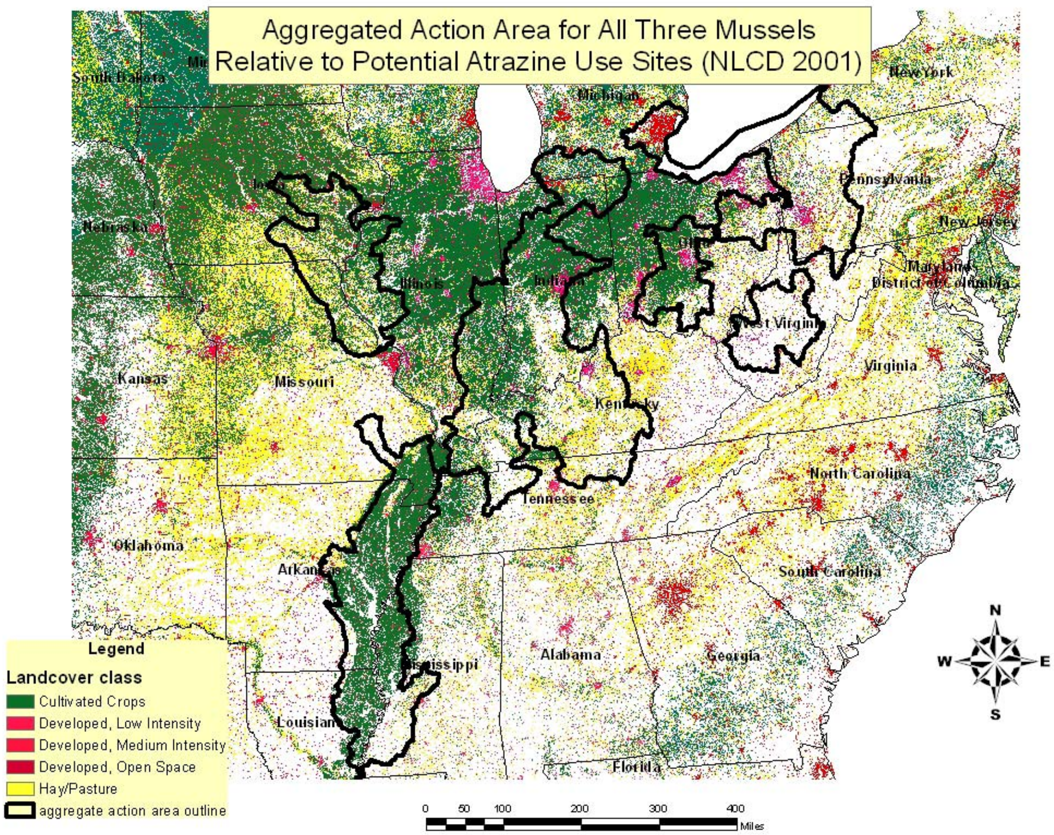

Figure 2.2 Agricultural Cropland Relative to Aggregated Action Area

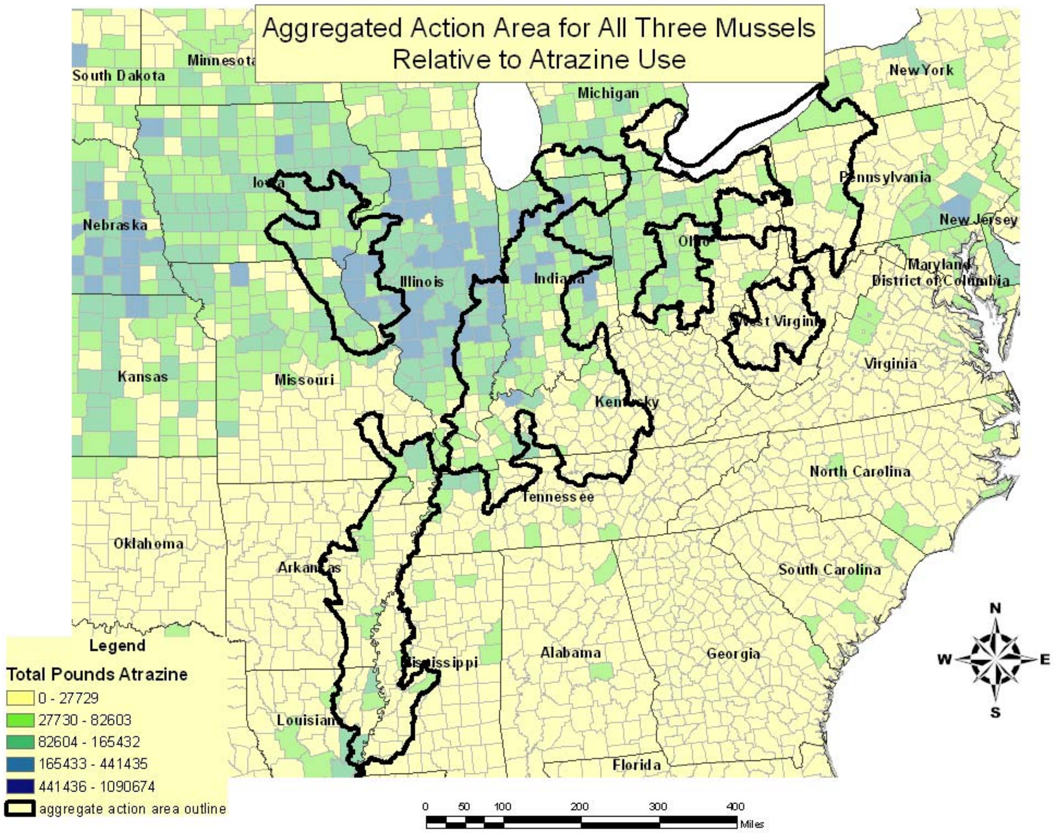

Figure 2.3 Atrazine Use Relative to Action Area

All agricultural use information for atrazine was considered in order to determine which uses occur within the action area for the three listed mussels (discussed further in Section 2.6). As noted above, information is not available for non-agricultural uses; therefore, they are presumed to occur within the action area and are included in this assessment. Agricultural uses of atrazine within the action area include corn, sweet corn, sorghum, and fallow/pasture. Specifically, county level data for the areas within and immediately surrounding the action area were used (Kaul and Jones, 2006). County level estimates of atrazine use were derived using state level estimates from USDA-NASS and data obtained from Doane (www.doane.com; the full dataset is not provided due to its proprietary nature). State level data from 1998 to 2004 were averaged together and extrapolated down to the county level based on apportioned county level crop acreage data from the 2002 USDA Agriculture of Census (AgCensus).

Of the thirteen principal states making up the regionalized approach for conducting the exposure assessment (several states far removed from the species location were not evaluated for use information because it is assumed that use in states in close proximity will have the greatest impact on the species) atrazine was used between 1998 and 2004 on average approximately 43,700,000 total pounds across all use sites in these 13 states.

The state with the highest use was Illinois with approximately 12,200,000 lbs used, and the least use was reported in West Virginia. The crop with the greatest use was corn with approximately 42,000,000 lbs. All other crops averaged less use than corn.

In general, this information suggests that the northern portion of the action area is located within the highest atrazine use area in Illinois, Iowa and Ohio. In general, atrazine use decreases in intensity further south and east of this area, with the lowest use in the far eastern portions of the action area in West Virginia. The atrazine use pattern within the action area is graphically presented in Figure 2.3. It should be noted, however, that information on non-agricultural use of atrazine is not available and, therefore, was not included in Figure 2.3.

Typical use information for atrazine is summarized in Table 2.2. The total average atrazine use per year from 1998 to 2004 was roughly 44,000,000 lbs within these states. Of this, roughly 42,350,000 lbs are used on corn or approximately 96% of total atrazine use. Of the remainder, only sorghum was used at amounts at or above 1,000,000 lbs. For all uses, the typical application rate and number of applications are fairly consistent across all states and all uses. For all uses, the average application rate is 1.2 lbs per acre, while the average number of applications is also 1.1. For corn, the average application rate is 1.2 lbs per acre, and the number of applications is also 1.1.

Table 2.2 Summary of Typical Atrazine Use Information Collected between 1998 and 2004 for all States in the Action Area

Crop	Total Pounds by Crop	Average Number of Applications by Crop	Average Application Rate (lbs/acre) by Crop
corn	42,352,000	1.1	1.2
Fallow/hay/pasture	32,000	1.1	1.1
sorghum	1,018,000	1.0	1.3
sweet corn	93,000	1.1	1.1
wheat	7,000	1.0	0.7

2.5 Assessed Species

General information on the following three listed freshwater mussels, including a summary of habitat requirements, food habits, and reproduction data relevant to this endangered species risk assessment is provided below:

- Fat pocketbook pearly mussel;
- Purple cat's paw pearlymussel; and
- Northern riffleshell.

All three of the assessed listed mussels are freshwater species that share similar general habitat requirements and reproductive cycles. In general, they live embedded in the bottom sand, gravel, and/or cobble substrates of rivers and streams. They also have a unique life cycle that involves a parasitic stage on host fish. Juvenile mussels require stable substrates with low to moderate amounts of sediment, low amounts of filamentous algae, and correct flow and water quality to continue to develop (USFWS, 2004). During the spawning period, males discharge sperm into the water column, and the sperm are taken in by females through their siphons during feeding and respiration. The females retain the fertilized eggs in their gills, until the larvae (glochidia) fully develop. The mussel glochidia are released into the water where they must attach to the gills and fins of appropriate host fishes, which they parasitize for a short time until they develop into juvenile mussels. The presence of suitable host fish is considered an essential element in the mussels' life cycles. Once the glochidia metamorphose to the juvenile stage, they drop to the substrate. If the environmental conditions are favorable, the juvenile mussel will survive and develop. Freshwater mussels are long lived, up to 50 years or more (USFWS, 1985). However, the northern riffleshell appears to have a relatively short life-span for a freshwater mussel (Rodgers et al., 2001). Freshwater mussels usually reach sexual maturity in 3-9 years.

All three listed species are members of the Unionidae family, which exhibit two reproductive cycles based on the length of time glochidia are retained in the gills of females. Fertilization occurs in the spring in tachytictic mussels (short-term brooders) and glochidia are released during spring and summer. In bradytictic species (long-term brooders), fertilization occurs in mid-summer and fall, and glochidia are released the following spring and summer (USFWS, 1976).

All adult freshwater mussels are filter-feeders, orienting themselves in the substrate to facilitate siphoning of the water column for oxygen and food (Kraemer, 1979). Phytoplankton is the principal food of bivalves, although mussels have also been reported to consume detritus, diatoms, zooplankton (microscopic animals that live suspended in the water), and other microorganisms (Ukeles, 1971; Coker et al., 1921; Churchill and Lewis, 1924; Fuller, 1974). Specific percentages of these food items within the mussel's diet are not known, although the available information indicates that adult mussels can clear and assimilate fine particulate organic matter (FPOM) particles ranging in size from 0.9 to 250 µm (Silverman et al., 1997; Wissing, 1997; and Nichols and Garling, 2000). This size range includes bacteria and algal cells, detritus, and soil particles (Allan, 1995). Juveniles up to two weeks old feed on bacteria, algae, and diatoms with small amounts of detrital and inorganic colloidal particles (Yeager et al., 1994). The diet of the glochidia comprises water (until encysted on a fish host) and fish body fluids (once encysted).

According to the USFWS (1985), the greatest single factor contributing to the decline of freshwater mussels is the alteration and destruction of stream habitat due to impoundments for flood control, navigation, hydroelectric power, and recreation. These dams and their impounded waters present physical barriers to the natural dispersal of mussels, including emigration (dispersal) of host fishes, and effectively isolate surviving mussel populations causing fragmentation in limited portions of their habitat range. Mussels are also susceptible to adverse effects caused by siltation in waterways. Specific biological impacts on mussels from excessive sediments include reduced feeding and respiratory efficiency from clogged gills, disrupted metabolic processes, reduced growth rates, increased substrata instability, limited burrowing activity and physical smothering (Ellis, 1936; Stansbery, 1971; Markings and Bills, 1979; Kat, 1982; Vannote and Minshall, 1982; Aldridge et al., 1987; and Waters, 1995).

A summary of the current range, habitat type, reproductive cycle, and glochidial hosts for each of the three assessed species is provided in Table 2.3. As shown in Table 2.3, the current range of the three assessed species spans various watersheds within 13 states, including Arkansas, Iowa, Illinois, Indiana, Louisiana, Kentucky, Michigan, Mississippi, Missouri, Ohio, Tennessee, Pennsylvania, and West Virginia. Information on the current habitat ranges of the listed mussels was obtained from USFWS recovery plans, which exist for all three assessed species (USFWS 1989, 1992, and 1994), species-specific information available on the USFWS website (http://www.fws.gov/endangered/; accessed in March 2007), locational information on the NatureServe website ((http://www.natureserve.org/; accessed March 2007), the draft 5-year review for the PCPP mussel (USFWS, 2007 draft), and personal communications with several known freshwater mussel experts (personal communications with Paul Hartfield [USFWS] 2007, Angela Zimmerman [USFWS] 2007, and Robert Anderson [USGS] 2007). Further detail on the general and specific status and life history information for the assessed mussels, including a diagram of the mussel's life cycle and species-specific maps depicting known occurrences, are provided in Appendix C.

Table 2.3 Summary of Current Distribution, Habitat Requirements, and Life History Information for the Three Assessed Mussels				
Assessed Species	**Current Range**	**Habitat Type (Stream order and range of flow rates)**	**Reproductive Cycle[a]**	**Known Glochidial Hosts**
Fat pocketbook mussel	(AR, IA, IL, IN, LA, KY, MS, MO): St. Francis River System (MO, AR); lower Wabash River (IN and IL); mouth of the Cumberland River and Ohio River (KY); Mississippi River (MS, AR, and MO); White River and Black River at Black Rock (AR); Gilliam Chute and St. Catherines Creek (MS); Cottonwood Chute at confluence with Lake Providence Harbor (LA)	Large rivers, streams, and chutes with flowing water and a mixture of sand, silt, and clay substrates (1^{st} and 7^{th} order streams with flow between 100 and 600,000 ft^3/s)	Long-term breeder (bradytictic)	Freshwater drum (*Aplodinotus grunniens*)
Purple cat's paw pearlymussel	(KY, OH, TN): Killibuck Creek and Walhonding River in Conshocton County (OH); Green River in Warren and Butler Counties (KY); and the middle Cumberland River in Smith County (TN)	Large rivers and streams with moderate to swift currents in sand/gravel substrate (3rd and 5^{th} order streams with flow between 5,000 and 17,000 ft^3/s)	Unknown	Unknown
Northern riffleshell	(KY, MI, OH, PA, WV): Fish Creek (OH) and Detroit River (MI) in the St. Lawrence River System; Green River (KY), Big Darby Creek (OH), Allegheny River (PA), Conewango Creek (PA), French Creek (PA), LeBeoeuf Creek (PA), Muddy Creek (PA), an Elk River (WV) in the Ohio River System	Large and small streams in riffles and runs with firmly packed sand and fine to coarse gravel substrate (also known to occur in slow-flowing, more lentic, deep run habitats) (2^{nd} and 4^{th} order streams with flow between 100 and 16,000 ft^3/s)	Long-term breeder (bradytictic)	Banded darter (*Etheostoma zonale*), bluebreast darter (*E. camurum*), Iowa darter (*E. exile*), Johnny darter (*E. nigrum*), brown trout (*Salmo trutta*), banded sculpin (*Cottus carolinae*) , and mottled sculpin (*C. bairdi*)

[a] Tachytictic species have a spring fertilization period, then the glochidia are incubated for a few months and expelled during the summer or early fall. Bradytictic species have a late summer or early fall fertilization period with the glochidia incubating overwinter, and expelled the following spring or summer.

2.6 Action Area

For listed species assessment purposes, the action area is considered to be the area affected directly or indirectly by the federal action and not merely the immediate area involved in the action (50 CFR 402.02). It is recognized that the overall action area for the national registration of atrazine uses is likely to encompass considerable portions of the United States based on the large array of both agricultural and non-agricultural uses. Based on the available atrazine monitoring data (discussed further in Section 3.2.6) and the toxicity data for the most sensitive non-vascular aquatic plant, the Agency's LOCs are likely to be exceeded in many watersheds that are in proximity to or downstream of atrazine use sites. Therefore, the overall action area for atrazine is likely to include many watersheds of the United States that co-occur and/or are in proximity to agricultural and non-agricultural atrazine use sites. However, in order to focus this assessment, the scope limits consideration of the overall action area to those geographic portions that may be applicable to the protection of the fat pocketbook, PCPP mussel, and northern riffleshell mussels (hereafter referred to as the "three listed mussels") included in this assessment. Based on the available information on potential atrazine use sites, none of the streams and rivers that are within the range of the three listed mussels could be excluded from the action area. Therefore, the portion of the atrazine action area that is assessed as part of this endangered species risk assessment includes the area within the boundary of the watersheds that drain to known current locations of the three listed mussels.

The three listed mussels are known to currently exist in a wide geographic range from Louisiana, north along the Mississippi River Valley, to the lower Missouri River Valley, northwest into Iowa, and east along the Ohio River Valley extending into Pennsylvania and West Virginia. In general, the species are found in streams and rivers within the Mississippi, Missouri, and Ohio River watersheds. Historically, the three listed mussels are presumed to have ranged over a much broader area; however, this assessment focuses on the current range of the species. In many instances, the location information (NatureServe; http://www.natureserve.org/explorer/) for the three listed mussels is non-specific and has been identified as county-level occurrences. The accuracy of current county-level occurrences and additional site-specific location information for the PCPP mussel (Angela Zimmerman, personal communication, 2007), the northern riffleshell (Robert Anderson, personal communication, 2007), and the fat pocketbook (Paul Hartfield, personal communication, 2007) have been provided and verified by the relevant expert within USFWS. These data have been used to identify locations where the species reside to focus the action area on those locations directly relevant to the species being assessed. The "action area" is the overall geographic scope where effects may occur. However, because this assessment is limited to reviewing potential effects of atrazine use to the three listed mussels, the action area is defined as the geographic scope where effects may occur, either directly or indirectly, to these species. Therefore, the initial definition of the action area for the three listed mussels is defined by the watersheds that drain to the known current range of these species.

In order to complete this task, watershed-based maps (Figure 2.5 to Figure 2.7) were created for each individual species. In addition, an aggregated locational map for all three mussel species was created (Figure 2.8). Maps depicting current locations of the three listed mussels were created using ArcMap GIS. Each of the streams, rivers, and counties identified as occupied using NatureServe data (and verified with USFWS freshwater mussel experts) were added to the map. Additional point locations not included in the NatureServe data were provided by the USFWS experts. Both the point locations (typically identified by stream reach) and county-level occurrences from NatureServe were assigned to a watershed (HUC8, or USGS hydrologic unit code) and added to the map. The USGS has defined watersheds within the entire United States into increasingly smaller HUCs, from coarse scales (Regions, or HUC2 watersheds) to subregions (HUC4 watersheds) to accounting units (HUC6 watersheds) to cataloging units (HUC8 watersheds). Those HUCs not draining to the streams where the three listed mussels occur were eliminated from the final map. Ultimately, the action area is defined by those HUC8 watersheds draining to the species' habitat range.

More detail on the Agency's enhanced reach file (ERF) stream data and the USGS' HUC classification scheme may be found at the following websites:

http://www.epa.gov/waters/doc/refs.html

http://water.usgs.gov/GIS/huc.html

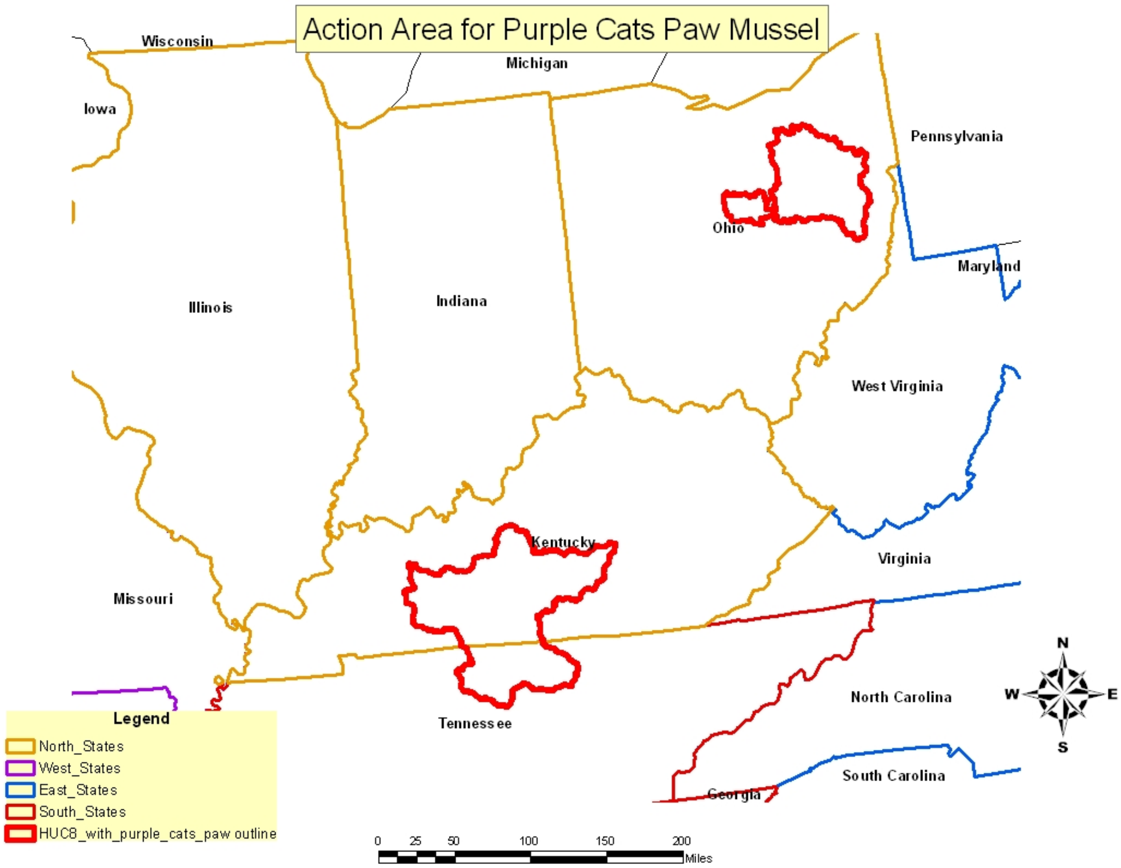

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Figure 2.5 Purple Cats Paw Mussel Action Area Defined by Hydrologic Unit Code (HUC8) Watersheds

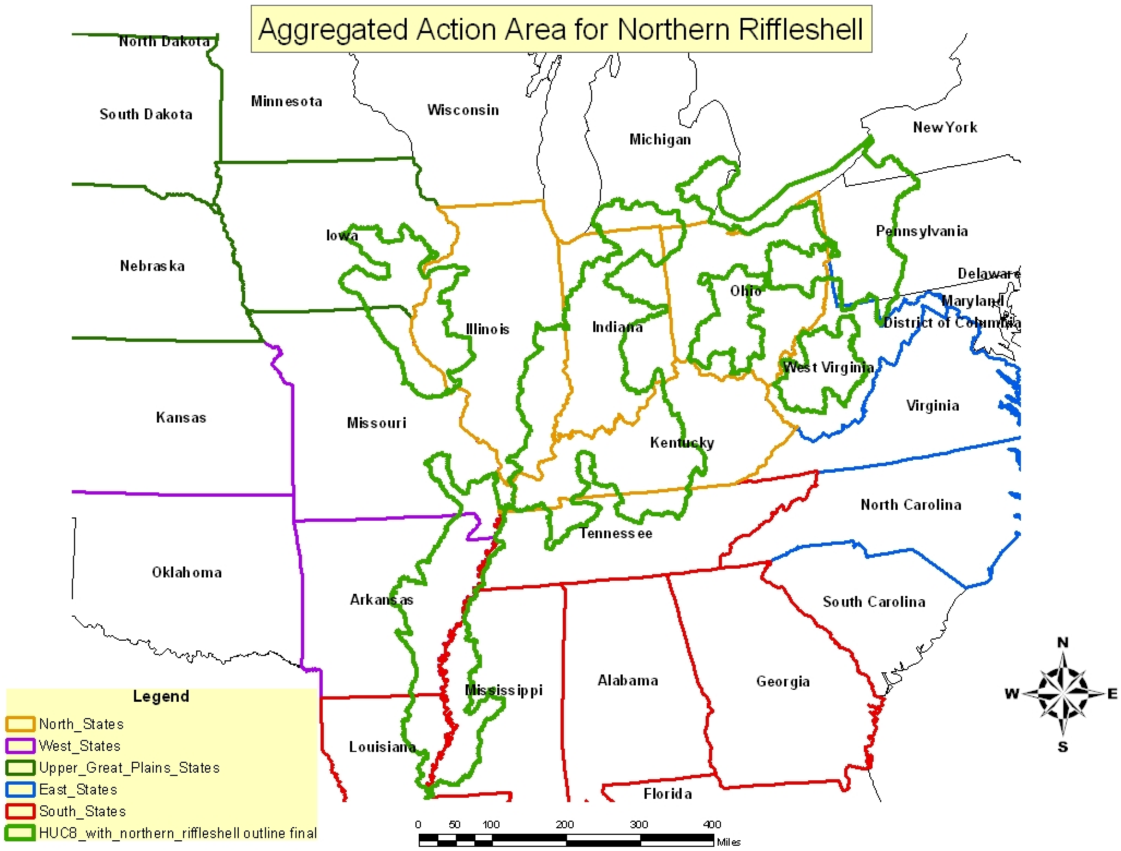

Figure 2.6 Northern Riffleshell Mussel Action Area Defined by Hydrologic Unit Code (HUC8) Watersheds

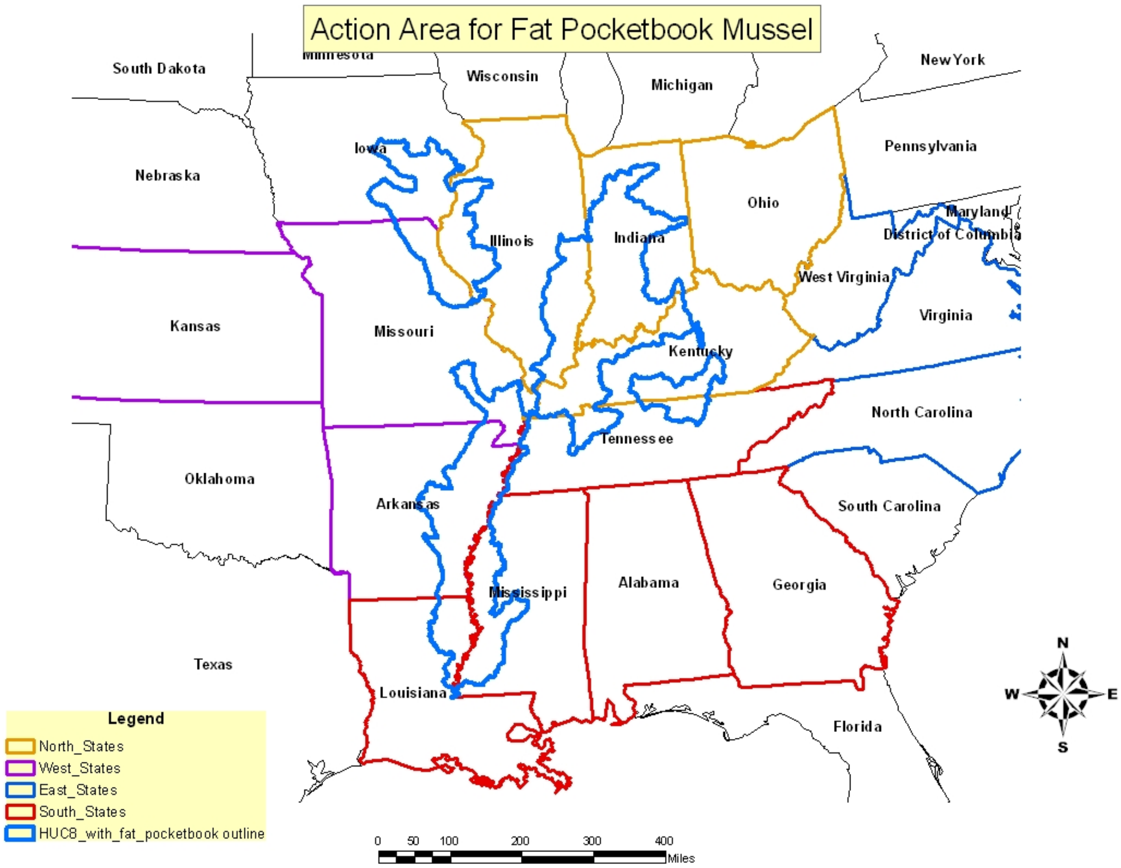

Figure 2.7 Fat Pocketbook Mussel Action Area Defined by Hydrologic Unit Code (HUC8) Watersheds

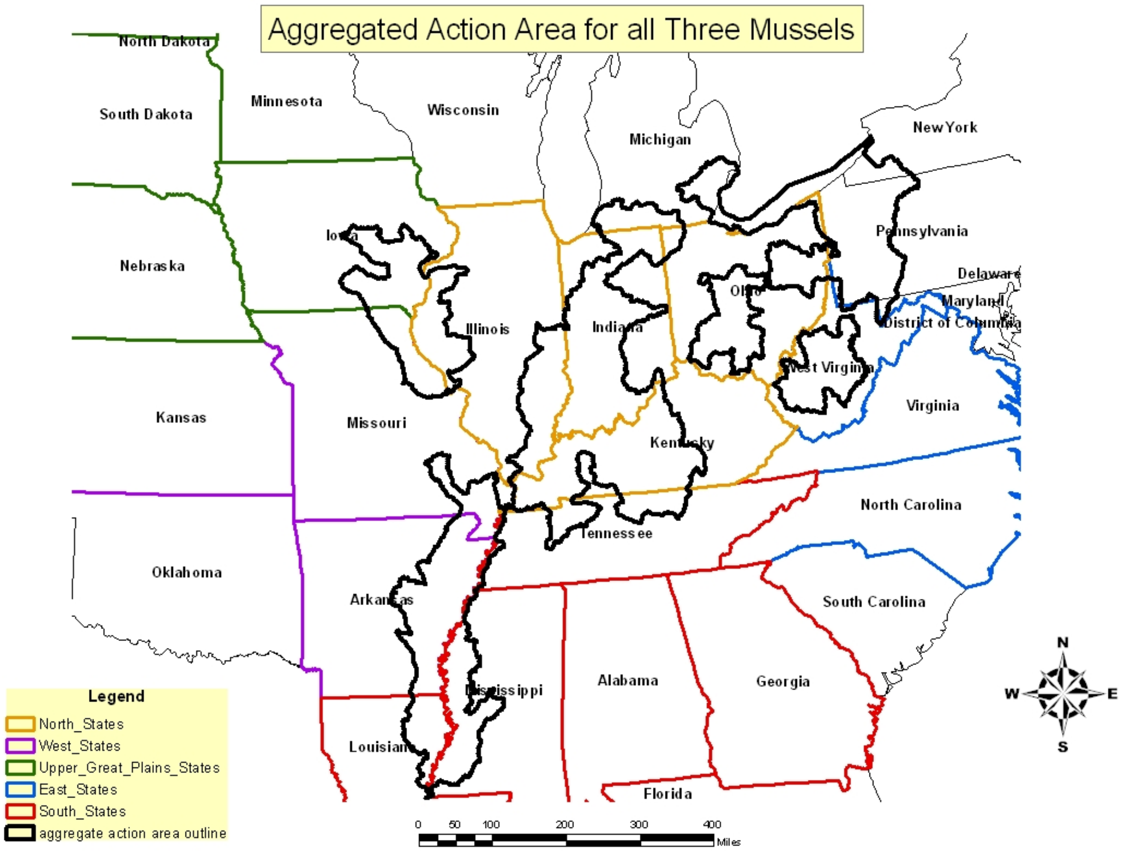

Figure 2.8 Aggregated Mussel's Action Area Defined by Hydrologic Unit Code (HUC8) Watersheds

Current labels were reviewed and local use information was evaluated to determine which atrazine uses could potentially be present within the defined area. This data suggest that extensive agricultural uses are present within the defined area and that the existence of non-agricultural uses cannot be precluded. Finally, local land cover data were considered to refine the characterization of potential atrazine use in the areas defined above. The overall conclusion of this analysis was that while certain agricultural uses could likely be excluded (i.e. sugarcane, guava and macadamia nuts) and some non-agricultural uses of atrazine were unlikely, none of the full extent depicted in the figures above could be excluded from the final action area based on usage and land cover data.

The environmental fate properties of atrazine were also evaluated to determine which routes of transport are likely to have an impact on the listed species included in this assessment. Review of the environmental fate data, as well as physico-chemical properties of atrazine, suggest that transport via runoff and spray drift are likely to be the dominant routes of exposure. In addition, long-range atmospheric transport of pesticides could potentially contribute to atrazine concentrations in the aquatic habitat used by the three listed mussels. Given the physico-chemical profile for atrazine and data showing that atrazine has been detected in both air and rainfall samples, the potential for long range transport from outside the area defined above cannot be precluded. However, the contribution of atrazine via long-range atmospheric transport is not expected to approach the concentrations predicted by modeling. The available data (U.S. EPA, 2003a) indicate that atrazine can enter the atmosphere via volatilization and spray drift. Atrazine is frequently found in rain samples and tends to be seasonal, related to application timing. Finally, the data suggest that although frequently detected, atrazine concentrations detected in rain samples are less than those seen in the monitoring data and modeling conducted as part of this assessment and support the contention that runoff and spray drift are the principal routes of exposure.

Atrazine transport away from the site of application by both spray drift and volatilization has been documented. Spray drift is addressed as a localized route of transport from the application site in the exposure assessment. However, quantitative models are currently unavailable to address the longer-range transport of pesticides from application sites. The environmental fate profile of atrazine, coupled with the available monitoring data, suggest that long-range transport of volatilized atrazine is a possible route of exposure to non-target organisms; therefore, the full extent of the action area could be influenced by this route of exposure. However, given the amount of direct use of atrazine within the immediate area surrounding the species, the magnitude of documented exposures in rainfall at or below available surface water and groundwater monitoring data (as well as modeled estimates for surface water), and the lack of modeling tools to predict the impact of long range transport of atrazine, the extent of the action area is defined by the transport processes of runoff and spray drift for the purposes of this assessment.

Based on this analysis, the action area for atrazine as it relates to the three listed mussels is defined by the entire watersheds depicted in the Figure 2.8.

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."[4] Selection of the assessment endpoints is based on valued entities (i.e., three listed mussels), the ecosystems potentially at risk (i.e., streams and rivers of the Mississippi River valley from Louisiana, north to the lower Missouri River Valley, northwest into Iowa, and east along the Ohio River Valley extending into Pennsylvania and West Virginia), the migration pathways of atrazine (i.e., runoff and spray drift), and the routes by which ecological receptors are exposed to atrazine-related contamination (i.e., direct contact).

Assessment endpoints for the three listed mussels include direct toxic effects on the survival, reproduction, and growth of the mussels, as well as indirect effects, such as reduction of the prey base, perturbation of host fish, 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 or changes in a

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

surrogate entity or attribute in response to exposure to a pesticide. Specific measures of ecological effect are evaluated based on a variety of data sources including registrant-submitted studies and information from the open literature. Acute and chronic toxicity information from registrant-submitted guideline tests are required to be conducted on a limited number of organisms. Additional ecological effects data from the open literature, including effects data on aquatic freshwater microcosm and mesocosm data, were also considered. Acute atrazine effects data for freshwater mussels are available; however, chronic data for freshwater mussels are not. Therefore, chronic toxicity data for surrogate species are used to assess potential direct effects to the assessed mussels.

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

A complete discussion of all the toxicity data available for this risk assessment, including use of CASM and associated aquatic community-level threshold concentrations, and the resulting measures of ecological effect selected for each taxonomic group of concern, is included in Section 4 of this document. A summary of the assessment endpoints and measures of ecological effect selected to characterize potential assessed mussel risks associated with exposure to atrazine are provided in Table 2.4.

Table 2.4 Summary of Assessment Endpoints and Measures of Ecological Effect for Three Listed Mussels

Assessment Endpoint	Measures of Ecological Effect
1. Survival, growth, and reproduction of mussel individuals via direct effects	1a. Freshwater mussel LC_{50} 1b. Freshwater invertebrate NOAEC
2. Survival, growth, and reproduction of mussel individuals via indirect effects on food source (i.e., phytoplankton, zooplankton) or host fish (i.e., freshwater fish)	2a. Freshwater fish, invertebrate, and aquatic plant EC_{50} or LC_{50} 2b. Freshwater fish and invertebrate NOAEC 2c. Microcosm/mesocosm threshold concentrations showing aquatic primary productivity community-level effects
3. Survival, growth, and reproduction of mussel individuals via indirect effects on habitat and/or primary productivity (i.e., aquatic plant community)	3a. Vascular plant (duckweed) acute EC_{50} 3b. Non-vascular plant (freshwater algae) acute EC_{50} 3c. Microcosm/mesocosm threshold concentrations showing aquatic primary productivity community-level effects
4. Survival, growth, and reproduction of mussel	4a. Monocot and dicot seedling emergence EC_{25}

individuals via indirect effects on terrestrial vegetation (riparian habitat) required to maintain acceptable water quality and habitat	4b. Monocot and dicot vegetative vigor EC_{25}

2.8 Conceptual Model

2.8.1 Risk Hypotheses

Risk hypotheses are specific assumptions about potential adverse effects (i.e., changes in assessment endpoints) and may be based on theory and logic, empirical data, mathematical models, or probability models (U.S. EPA, 1998). For this assessment, the risk is stressor-linked, where the stressor is the release of atrazine to the environment. Based on the results of the 2003 atrazine IRED (U.S. EPA, 2003a), the following risk hypotheses are presumed for this endangered species risk assessment:

- Atrazine in surface water and/or runoff/drift from treated areas within the action area may directly affect one or more of the assessed mussel species by causing mortality or adversely affecting growth or fecundity;
- Atrazine in surface water and/or runoff/drift from treated areas within the action area may indirectly affect one or more of the assessed mussel species by reducing or changing the composition of food supply and/or perturbing fish hosts required for the parasitic glochidial life stage of the assessed mussels;
- Atrazine in surface water and/or runoff/drift from treated areas within the action area may indirectly affect one or more of the assessed mussels by reducing or changing the composition of the aquatic plant community in the rivers and streams comprising the species' current range, thus affecting primary productivity and/or cover; and
- Atrazine in surface water and/or runoff/drift from treated areas within the action area may indirectly affect one or more of the assessed mussels by reducing or changing the composition of the terrestrial plant community (i.e., riparian habitat) required to maintain acceptable water quality and habitat in the rivers and streams comprising the species' current range.

2.8.2 Diagram

The conceptual model is a graphic representation of the structure of the risk assessment. It specifies the stressor (atrazine), release mechanisms, abiotic receiving media, biological receptor types, and effects endpoints of potential concern. The conceptual model for this endangered species risk assessment is shown in Figure 2.9. Exposure routes shown in dashed lines are not quantitatively considered because the contribution of those potential exposure routes to potential risks to the assessed mussel species is expected to be negligible.

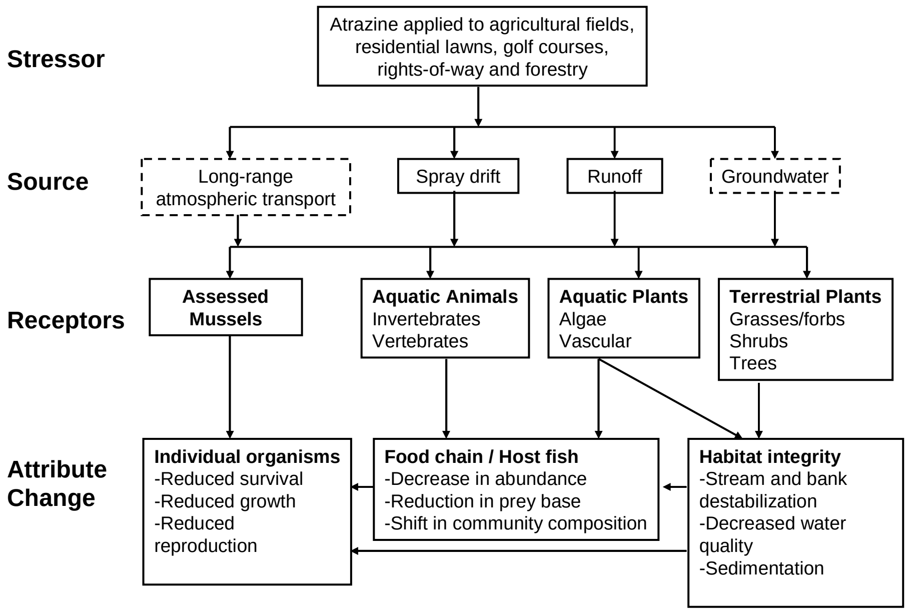

Figure 2.9 Conceptual Model for Three Assessed Mussel Species

The conceptual model provides an overview of the expected exposure routes for the assessed mussels within the atrazine action area previously described in Section 2.6. In addition to the mussel species included in this assessment, other aquatic receptors that may be potentially exposed to atrazine include freshwater fish, invertebrates and aquatic plants. For freshwater vertebrate and invertebrate species, including the assessed mussels and their host fish, the major routes of exposure are considered to be via the respiratory surface (gills) or the integument. Direct uptake and adsorption are the major routes of exposure for aquatic plants. Direct effects to freshwater invertebrates and aquatic plants resulting from exposure to atrazine may indirectly affect the assessed mussels via reduction and/or alteration in food and habitat (i.e., substrate, water quality including oxygen content) availability necessary for normal behavior, growth, and viability of all life stages. The available data indicate that atrazine is not likely to bioconcentrate in aquatic food items, with fish bioconcentration factors (BCFs) ranging from 2 to 8.5 (U.S. EPA, 2003c). Therefore, bioconcentration of atrazine in mussels or in host fish via the diet was not considered as a route of exposure.

In addition to aquatic receptors, terrestrial plants may also be exposed to spray drift and runoff from atrazine use in the vicinity of the rivers and streams that comprise the mussel species' current range. Detrimental changes in the riparian vegetation adjacent to the mussel's current habitat may cause adverse effects to water quality (i.e., temperature and turbidity), stream bank stability, substrate composition, sediment loading, and spawning habitat for host fish. Specifically, changes in the riparian vegetation adjacent to the habitat of the assessed mussels may adversely affect mussel feeding and respiratory efficiency, growth rates, and burrowing activity, and cause increased substrate instability and potential physical smothering via increased sedimentation (Ellis, 1936; Stansbery, 1971; Markings and Bills, 1979; Kat, 1982; Vannote and Minshall, 1982; Aldridge et al., 1987; and Waters, 1995).

The source and mechanism of release of atrazine into surface water are ground application via foliar spray and coated fertilizer granules for agricultural (i.e., corn, sorghum, and fallow/idle land) and non-agricultural uses (i.e., golf courses, residential lawns, rights-of-way, and forestry). Surface water runoff from the areas of atrazine application is assumed to follow topography, resulting in direct runoff to the rivers and streams within the action area. Spray drift and runoff of atrazine may also affect the foliage and seedlings of terrestrial plants that comprise the riparian habitat that may be adjacent to the mussel's habitat. Additional release mechanisms include spray drift and atmospheric transport via volatilization, which may potentially transport site-related contaminants to the surrounding air. Atmospheric transport is not considered as a route of exposure for this assessment because the magnitude of documented exposures in rainfall are at or below available surface water and monitoring data, as well as modeled estimates of exposure.

2.9 Analysis Plan

The purpose of this assessment is to make an "effects determination" for three listed species of freshwater mussels including the fat pocketbook, PCPP mussel, and the northern riffleshell by evaluating the potential direct and indirect effects of the herbicide atrazine on the survival, growth, and reproduction of these Federally endangered species. This assessment was completed in accordance with the procedures outlined in the Agency's Overview Document (U.S. EPA, 2004) and the Services' Evaluation Memorandum (USFWS/NMFS, 2004b).

Atrazine is used throughout the United States on a number of agricultural crops (primarily corn, sorghum, and sugarcane) and on non-agricultural sites (including residential uses, forestry, and turf). Although the action area is likely to encompass a large area of the United States, given its use, the scope of this assessment limits consideration of the overall action area to those portions that are applicable to the protection of the three listed mussels. Specifically, the action area for the three listed mussels includes a wide geographic range from Louisiana, north along the Mississippi River Valley, to the lower Missouri River Valley, northwest into Iowa, and east along the Ohio River Valley extending into Pennsylvania and West Virginia. In general, the three listed species are found streams and rivers within the Mississippi, Missouri, and Ohio River watersheds.

Screening-level estimates of aquatic exposure are based on PRZM/EXAMS modeling, which assumes a static non-flowing water body. Terrestrial plant exposure concentrations were estimated using OPP's TerrPlant model (U.S. EPA, 2007d; Version 1.2.2), considering use conditions likely to occur in the watersheds where the listed mussels occur. Screening-level EECs were modeled for agricultural (corn, sorghum, fallow/idle land) and non-agricultural (forestry, turf, residential) uses in accordance with the label. The non-flowing nature of the standard water body provides a reasonable estimation of peak exposures for many smaller headwater streams found in agricultural areas; however, it appears to overestimate exposures for longer time periods and for flowing water bodies. Given that exposure concentrations based on the standard ecological body are likely to overestimate exposure for the listed mussels (because these species require flowing water), additional flow-adjusted modeling was used together with available monitoring data to refine atrazine exposures in flowing waters.

A robust set of surface water monitoring data, which is described in further detail in Section 3.2.6, is available for atrazine. Based on an analysis of site-specific flow data for occupied streams and locations sampled as part of the targeted monitoring data, targeted atrazine monitoring data from the AEMP were used to refine exposure for populations of two of the three listed mussels (fat pocketbook and northern riffleshell) that occur in low flow vulnerable watersheds (defined as watersheds most vulnerable to atrazine runoff because they are located in high atrazine use areas) of the action area. Available non-targeted monitoring data (i.e., data in which the study design was not specifically targeted to detect atrazine in high use areas) and flow-adjusted modeling were used to refine exposure for the PCPP mussel and populations of fat pocketbook and northern riffleshell mussels that occur outside the boundary of vulnerable watersheds. In addition, the non-targeted monitoring data and flow-adjusted modeling were also used to refine exposures for populations of fat pocketbook and northern riffleshell mussels that occupy large, fast-flowing rivers within the boundary of vulnerable watersheds, such as the Mississippi, Cumberland, and Ohio Rivers. Therefore, separate effects determinations were derived for direct/indirect endpoints by considering the flow regime of the occupied streams as well as the location of the assessed mussels within highly vulnerable and less vulnerable watersheds of the action area.

The assessment endpoints for the listed mussels include direct toxic effects on the survival, reproduction, and growth of individual mussels, as well as indirect effects, such as reduction of the prey base, perturbation of host fish, and/or modification of its habitat. Direct effects to the listed mussels are based on available toxicity information for freshwater mussels and invertebrates. Given that the mussel's prey items, host fish, and habitat requirements are dependant on the availability of freshwater fish and aquatic invertebrates, aquatic plants, and terrestrial plants (i.e., riparian habitat), toxicity information for these taxonomic groups is also discussed. In addition to the registrant-submitted and open literature toxicity information, indirect effects to the listed mussels, via impacts to aquatic plant community structure and function, are also evaluated based on time-weighted threshold concentrations that correspond to potential aquatic plant community-level effects.

Degradates of atrazine include hydroxyatrazine (HA), deethylatrazine (DEA), deisopropylatrazine (DIA), and diaminochloroatrazine (DACT). Comparison of available toxicity information for the degradates of atrazine indicates lesser aquatic toxicity than the parent for freshwater and estuarine/marine fish, aquatic invertebrates, and aquatic plants. Although degradate toxicity data are not available for terrestrial plants, lesser or equivalent toxicity is assumed, given the available ecotoxicological information for other taxonomic groups including aquatic plants and the likelihood that the degradates of atrazine may lose efficacy as an herbicide. Because degradates are not of greater toxicological concern than atrazine, concentrations of the atrazine degradates are not assessed further, and the focus of this assessment is parent atrazine. An analysis of registered products that contain multiple active ingredients, including atrazine, is also included as part of this assessment.

Risk quotients (RQs) are derived as quantitative estimates of potential high-end risk. Acute and chronic RQs are compared to the Agency's levels of concern (LOCs) to identify instances where atrazine use within the action area has the potential to adversely affect the listed mussels via direct toxicity or indirectly based on direct effects to their host fish, food supply (i.e., phytoplankton and zooplankton) or habitat (i.e., aquatic plants and terrestrial riparian vegetation). When RQs for a particular type of effect are below LOCs, the pesticide is considered to have "no effect" on the species. Where RQs exceed LOCs, a potential to cause adverse effects is identified, leading to a conclusion of "may affect". If a determination is made that use of atrazine within the action area "may affect" the listed mussels, additional information is considered to refine the potential for exposure and effects, and the best available information is used to distinguish those actions that "may affect, but are not likely to adversely affect" from those actions that are "likely to adversely affect" the listed mussels.

3. Exposure Assessment

3.1 Label Application Rates and Intervals

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

In the January and October 2003 IREDs (U.S. EPA, 2003a and b), EPA stipulated numerous changes to the use of atrazine including label restrictions and other mitigation measures designed to reduce risk to human health and the environment. Specifically pertinent to this assessment are provisions of a Memorandum of Agreement (MOA) between the Agency and atrazine registrants. In the MOA, the Agency stipulated that certain label changes must be implemented on all manufacturing-use product labels for atrazine and on all end-use product labels for atrazine prior to the 2005 growing season. These label changes included cancellation of certain uses, reduction in application rates, and requirements for harmonization across labels including setbacks from waterways. Specifically, the label changes prohibit atrazine use within 50 feet of sinkholes, 66 feet of intermittent and perennial streams, and 200 feet of lakes and reservoirs.

While these setbacks were required to reduce atrazine deposition to water bodies as a result of spray drift, it is expected that they will also result in a reduction in loading due to runoff across the setback zone; however, current models do not address this reduction quantitatively. Therefore, these restrictions are not quantitatively evaluated in this assessment. A qualitative discussion of the potential impact of these setbacks on estimated environmental concentrations of atrazine for the assessed mussels is discussed further in Section 3.2.3. Table 3.1 provides a summary of label application rates for atrazine uses evaluated in this assessment.

Currently registered non-agricultural uses of atrazine within the action area include residential areas such as playgrounds and home lawns, turf (golf courses and recreational fields), rights-of-way, and forestry. Agricultural uses within the action area include corn, sorghum, and fallow/idle land[5]. Other agricultural uses (macadamia nut, guava, and sugarcane) are not present in the action area.

Atrazine is formulated as liquid, wettable powder, dry flowable, and granular formulations. Application methods for the agricultural uses include ground application (the most common application method), aerial application, band treatment, and incorporated treatment; and application using various sprayers (low-volume, hand held, directed) for liquids, and spreaders for granulars. Risks from ground boom and aerial applications are considered in this assessment because they are expected to result in the highest off-target levels of atrazine due to generally higher spray drift levels. Ground boom and aerial modes of application tend to use lower volumes applied in finer sprays than applications coincident with sprayers and spreaders, and thus have a higher potential for off-target movement via spray drift.

Table 3.1 Atrazine Label Application Information for the Three Listed Mussels Assessment[a]

Scenario	Maximum Application Rate (lbs/acre)	Maximum Number of Annual Applications	Formulation	Method of Application	Interval Between Applications
Forestry	4.0	1	Liquid	Aerial and Ground	NA
Residential	2.0	2	Granular	Ground	30 days
Residential	1.0	2	Liquid	Ground	30 days
Rights-of-Way	1.0	1	Liquid	Ground	NA

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

Table 3.1 Atrazine Label Application Information for the Three Listed Mussels Assessment[a]					
Scenario	**Maximum Application Rate (lbs/acre)**	**Maximum Number of Annual Applications**	**Formulation**	**Method of Application**	**Interval Between Applications**
Fallow/ Idle land	2.25	1	Liquid	Ground and Aerial	NA
Corn	2.5	2	Liquid	Ground and Aerial	NA
Sorghum	2.0	1	Liquid	Ground and Aerial	NA
Turf	2.0	2	Granular	Ground	30 days
Turf	1.0	2	Liquid	Ground	30 days

[a] Based on 2003 IRED and Label Change Summary Table memorandum dated June 12, 2006 (U.S. EPA, 2006b).

3.2 Aquatic Exposure Assessment

As discussed in Section 2.5 and Appendix C, the three listed mussels principally reside in watersheds with a wide variety of streams orders and variable flow rates within in the Mississippi and lower Missouri River Valleys (fat pocketbook) and Ohio River Basin (northern riffleshell and PCPP mussel). The action area includes the entire watershed of rivers and streams in the areas defined above and is presented graphically in Figure 2.8. In general, the three listed mussels reside in streams that are typically classified as 2nd through 5th order using the Strahler system, although the fat pocketbook mussel also resides in smaller streams and chutes classified as 1st order streams, as well as larger rivers (i.e., 6th and 7th order) such as the Mississippi, Columbia, and Ohio Rivers. The general range of flow conditions required for the three listed freshwater mussels is discussed further in Section 3.2.7.

3.2.1 Introduction

The assessment of exposure within the action area is dependent upon a combination of modeling and monitoring data. In accordance with the Overview Document (U.S. EPA, 2004), screening-level exposures were based on modeling which assumes a static water body. Available monitoring data for atrazine, as well as refined flow-adjusted modeling (adjusted based on the low end of flow data from rivers and streams where the three listed mussels are likely to occur), were used to refine the screening-level modeled exposures.

Screening-level EECs based on the PRZM/EXAMS static water body are used in the risk estimation to derive initial RQs and distinguish between "no effect" and "may affect" determinations. Refined EECs are used to characterize exposure in the risk description for three listed mussels based on a combination of flow-adjusted EECs and available monitoring data. These refined exposure estimates are used to distinguish whether the three listed mussels are likely or not likely to be adversely affected by atrazine use within the action area. Selection of refined EECs is based on the site-specific flow data from the occupied streams and the location of the assessed mussels within highly vulnerable and less vulnerable watersheds of the action area. Further detail on the standard modeling, refined modeling, monitoring data evaluation, and characterization of exposure is presented in the following sections.

For this assessment, screening-level modeling using a static water body indicates long-term (e.g. 30-day average) exposure concentrations that are higher than concentrations seen in most monitoring data. Refined modeling based on flowing water suggests that concentrations in flowing water are lower than screening-level EECs, particularly for longer durations of exposure (e.g. 30-day rolling average). However, AEMP monitoring targeted to the upper 20th percentile vulnerable watersheds (based on WARP modeling[6]) indicates that, under certain conditions, long-term atrazine concentrations can be higher than those estimated by flow-adjusted modeling. Therefore, concentrations in some low flow portions of the most vulnerable areas (based on atrazine runoff) are likely to be less than the screening-level EECs using the static waterbody, but greater than the flow-adjusted EECs.

Based on the analysis described in Section 3.2.6.1, all three mussels inhabit watersheds that are at least partially located within the boundary of the 1,172 vulnerable watersheds (defined as those watersheds most prone to atrazine runoff). In the case of the northern riffleshell and PCPP mussel, there are distinct locations where occupied stream miles (determined based on information provided by USFWS but not graphically depicted to protect USFWS concerns regarding the exact location of the species) appear to be located outside the 1,172 vulnerable watershed boundary. For example, the northern riffleshell is reportedly present in locations in Pennsylvania, Michigan, and West Virginia outside the range of the 1,172 watersheds, while the PCPP mussel is reportedly present in locations in Ohio and Tennessee also outside the vulnerable watershed boundary. There are also distinct populations in Kentucky for the PCPP mussel and in Indiana, Ohio, and Kentucky for the northern riffleshell that may be within the boundary of 1,172 vulnerable watersheds. The location information for the fat pocketbook mussel is less certain and appears to cover a much broader range than the other two assessed mussels; therefore, a more spatially explicit comparison of species' location with vulnerable watersheds is uncertain. It is expected that a large portion of the fat pocketbook range (as defined by the HUC8 watersheds) is within the range of the 1,172 vulnerable watersheds, but that a portion of the range in Kentucky, Indiana, Tennessee, Missouri, Arkansas Mississippi, and Louisiana is outside the boundary of the 1,172 vulnerable watersheds. A summary of general species' locations relative to the 1,172 watersheds is provided in Table 3.2.

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[6] Watershed Regression of Pesticides model (USGS 2005) at http://pubs.usgs.gov/circ/2005/1291/

Table 3.2 Summary of General Location of Listed Mussels Relative to 1,172 Vulnerable Watersheds		
Mussel Species	**Location of Populations Within Vulnerable Watersheds**	**Location of Populations Outside of Vulnerable Watersheds**
Fat pocketbook	SE Iowa, NE Missouri, W and SE Illinois, W and N central Indiana, W Kentucky, E Arkansas, NE Louisiana, and W Mississippi	S central Indiana, S Kentucky, N Tennessee, and SE Missouri
PCPP mussel	SW Kentucky and central Ohio	Central Kentucky, E Ohio, and N Tennessee
Northern riffleshell	W Kentucky, S central Ohio, NE Indiana, and NW Pennsylvania	S central Kentucky, West Virginia, W Pennsylvania, and Michigan

As previously discussed, selection of refined EECs was based on a comparison of site-specific flow data from the watersheds occupied by the three listed mussels with flow data from the locations sampled as part of the targeted AEMP. Based on this analysis, which is described in further detail in Section 3.2.7, the AEMP data represent only a subset of occupied streams within the boundary of vulnerable watersheds that is limited to those watersheds with flow below the 15th percentile of flow from occupied streams (< 200 ft^3/sec). Therefore, the targeted AEMP data are used to refine exposures only for the fat pocketbook and northern riffleshell in occupied streams within the vulnerable watershed boundary that have flow rates < 200 ft^3/sec or for which no flow rate information is available. Further analysis of the potential extrapolation of AEMP monitoring results from the 40 sampled sites to the full population of 1,172 watersheds is ongoing. Once this analysis is complete, it will be possible to more definitively determine whether AEMP monitoring data from the 40 sampled sites is representative of exposure in other watersheds within the vulnerable watershed boundary where the fat pocketbook and northern riffleshell mussels may occur. The PCPP mussel currently resides in a small number of streams, both within and outside the boundary of vulnerable watersheds; however, stream flow data, which is available for all occupied streams, suggests that this species requires a higher flow rate than those represented by the available targeted monitoring data. Therefore, refined flow-adjusted EECs and non-targeted monitoring data are used to refine estimated exposure concentrations for the PCPP mussel. Flow-adjusted modeling and non-targeted monitoring data are also used to refine exposure for those populations of fat pocketbook and northern riffleshell mussels that occur outside of vulnerable watershed boundary, as well as larger streams/rivers with flow rates > 200 ft^3/sec that occur within the boundary of vulnerable watersheds. The methods used to derive screening-level and refined estimated environmental concentrations (EECs) for use in this endangered species risk assessment are summarized in Table 3.3.

Table 3.3 Methodology for EEC Derivation and Use in Risk Assessment			
Assessed Mussel	Screening-Level EEC[1]	Refined EEC[2]	
		Occupied Streams Within the Boundary of Vulnerable Watersheds	Occupied Streams Outside the Boundary of Vulnerable Watersheds
Fat pocketbook and Northern Riffleshell	PRZM/EXAMS Static Water Body EECs[3]	Targeted AEMP Monitoring Data[4]: For occupied streams with flow rates < 200 ft^3/sec or for which no flow rate information is available Flow-adjusted EECs[5] and Non-targeted Monitoring Data[6]: For large streams/rivers with flow > 200 ft^3/sec	Flow-adjusted EECs[5] and Non-targeted Monitoring Data[6]: For all occupied streams outside the boundary of vulnerable watersheds
PCPP Mussel		Flow-adjusted EECs[5] and Non-targeted Monitoring Data[6]: For all occupied streams within vulnerable watersheds because site-specific flow data for the PCCP mussel indicate that the species requires higher flow rates than those represented by the available targeted monitoring data.	

[1] Used in the risk estimation to calculate screening-level RQs and distinguish "no effect" from "may affect" determinations.
[2] Used in the risk description to refine RQs and distinguish "LAA" from "NLAA" determinations.
[3] PRZM/EXAMS Static Water Body EECs are described further in Sections 3.2.2 through 3.2.4.
[4] Targeted refers to data from the AEMP in which the study design was specifically targeted to detect atrazine in vulnerable watersheds near high-use areas (see Section 3.2.6.1).
[5] Flow-adjusted EECs are described further in Section 3.2.5.1.
[6] Non-targeted refers to monitoring data in which the study design was not specifically targeted to detect atrazine in high-use areas. However, some non-targeted study sites are located in highly vulnerable watersheds and correlated with high atrazine use (see Sections 3.2.6.2 and 3.2.6.3).

Further detail on the standard modeling, refined modeling, monitoring data evaluation, and characterization of exposure is presented in the following sections.

3.2.2 Modeling Approach

Screening-level risk quotients (RQs) were initially based on EECs derived using the Pesticide Root Zone Model/Exposure Analysis Modeling System (PRZM/EXAMS) standard ecological pond scenario, according to the methodology specified in the Overview Document (U.S. EPA, 2004). While peak concentrations predicted with the static water body are generally consistent with monitored values, longer-term EECs predicted by modeling with the static water body likely overestimate exposure as compared to monitoring. Further, the three listed mussels reside in low order streams with moderate to strong flow relative to the no-flow condition assumed for the PRZM/EXAMS screening-level EECs. Therefore, additional modeling (adjusted for flow) (Section 3.2.5.1) together with available monitoring data (Section 3.2.6) is used to characterize and refine potential exposures for the three listed mussels. The targeted monitoring data also add a spatial component to the assessment by focusing on those areas most vulnerable to atrazine runoff. Where LOCs for direct/indirect effects are exceeded based on the modeled EECs using the static water body (i.e., "may affect"), the refined modeling and available monitoring data are used to differentiate "may affect, but not likely to adversely affect or NLAA" from "likely to adversely affect or LAA" determinations for the assessed species.

The general conceptual model of exposure for this assessment is that the highest exposures are expected to occur in headwater streams adjacent to agricultural fields and other non-agricultural use sites (residential, right-of-way, turf, and forestry). Many of the streams and rivers within the action area defined for this assessment are in close proximity to both agricultural and non-agricultural uses sites (Figures 2.2 and 2.3). The action area was divided into five representative regions and modeling scenarios were selected to represent each area. These regions, which are described in more detail in Section 3.2.3 and depicted in Figure 3.1, represent the western (Arkansas, Missouri, and eastern Nebraska), southern (Mississippi, Alabama, Georgia, and western Tennessee), northern (Kentucky, Ohio, western Pennsylvania, Illinois, and Indiana), eastern (West Virginia, Virginia, and North Carolina), and the upper Great Plains (Iowa, Nebraska, South Dakota, North Dakota, and Montana) portions of the United States which overlap with the action area.

None of the three listed mussels span the entire range of the five regions assessed for exposure. The fat pocketbook mussel ranges from the lower Mississippi River Valley to the lower Missouri and Ohio River Valley and is best represented by EECs derived from the PRZM/EXAMS scenarios representing the south, north, west, and upper Great Plains regions (Figure 3.2). The northern riffleshell mussel ranges from the lower Ohio River Valley in Kentucky to western Pennsylvania and includes areas draining the Great Lakes. This species is best represented by EECs from PRZM/EXAMS scenarios representing the north and east regions (Figure 3.3). Finally, the PCPP mussel, which is found in isolated pockets in Ohio, Kentucky, and Tennessee near the border with Kentucky, is best represented by EECs derived from PRZM/EXAMS scenarios representing the northern region (Figure 3.4). A summary of the distribution of the three assessed mussels within the five geographical regions of the action area is provided in Table 3.4.

Table 3.4 Regional Distribution of the Assessed Mussels

Assessed Mussel	Region				
	South	East	North	West	Upper Great Plains
Fat pocketbook	X	--	X	X	X
PCPP mussel	--	--	X	--	--
Northern riffleshell	--	X	X	--	--

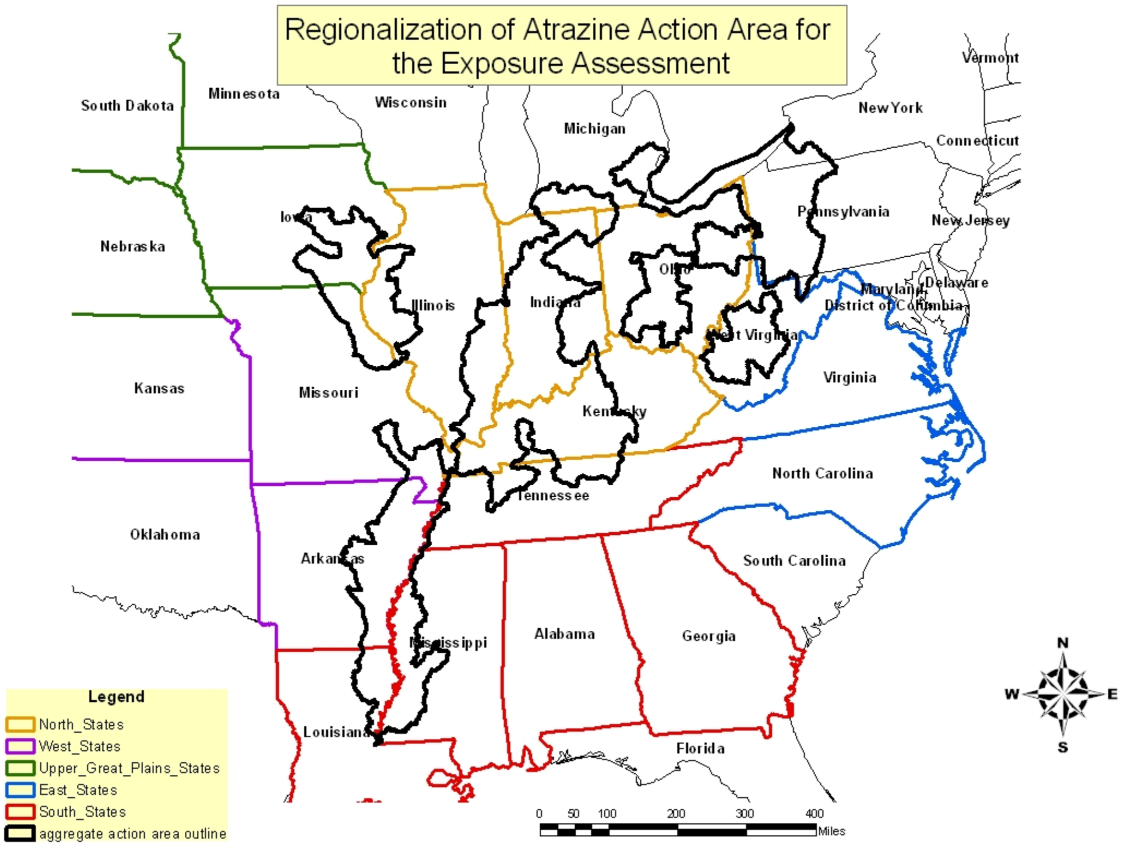

Figure 3.1 Regionalization of the Aggregated Action Area

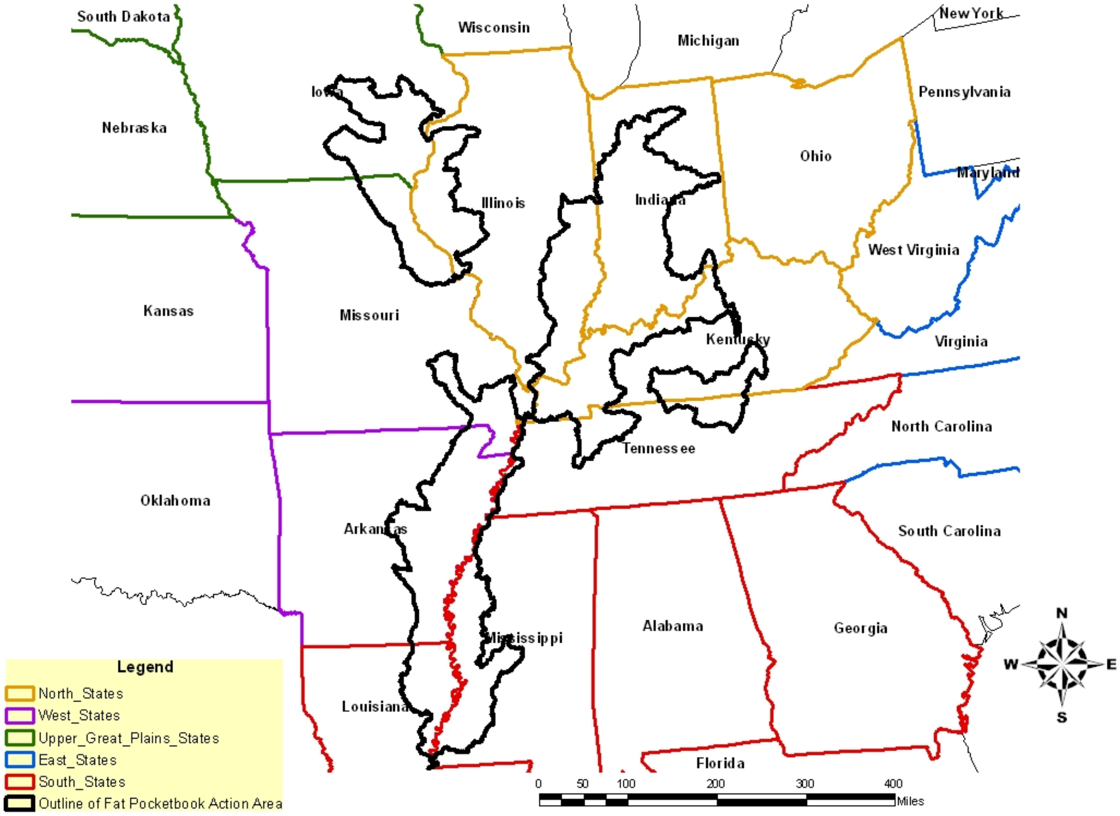

Figure 3.2 Regionalization of Fat Pocketbook Mussel Portion of the Action Area

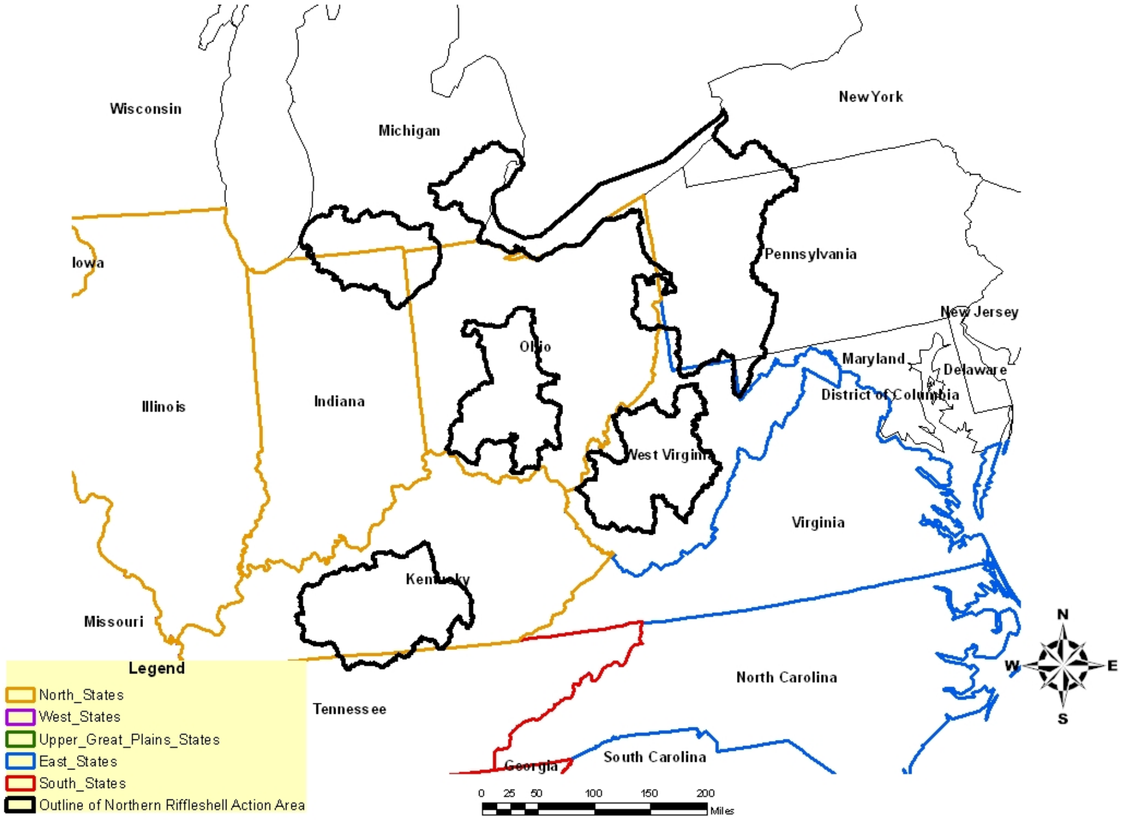

Figure 3.3 Regionalization of Northern Riffleshell Mussel Portion of the Action Area

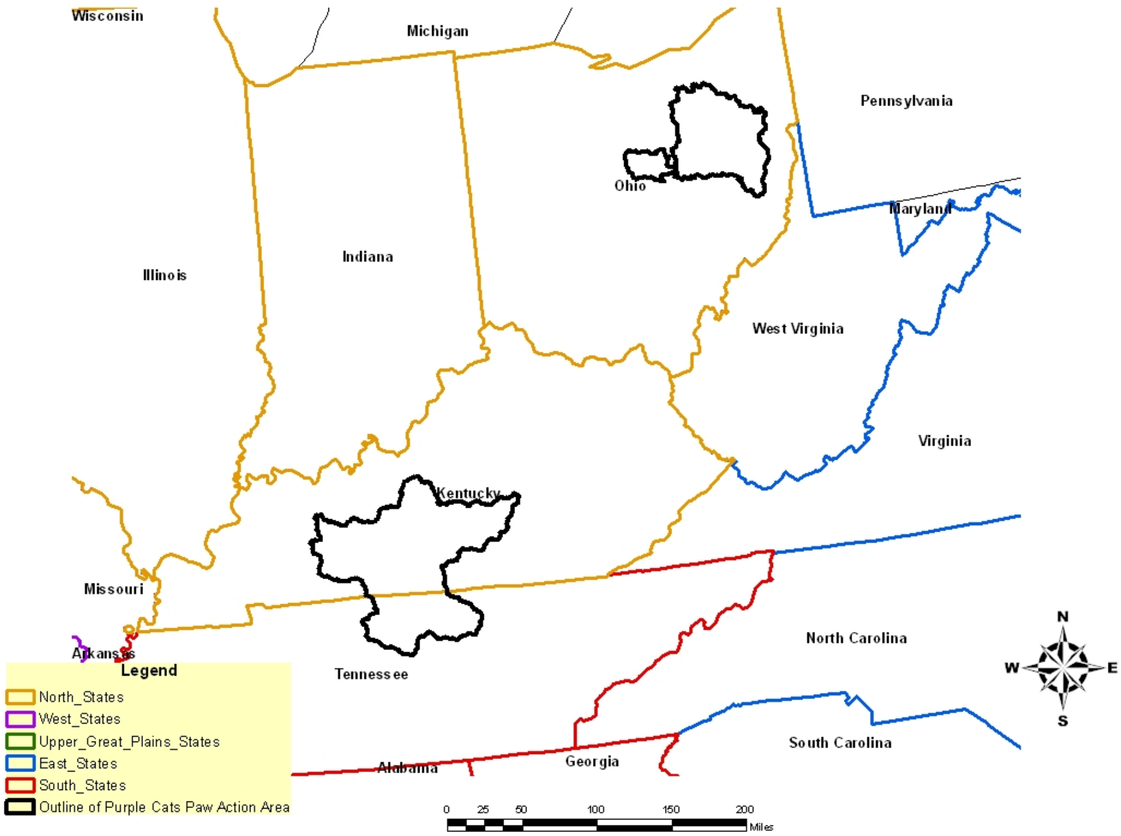

Figure 3.4 Regionalization of Purple Cats Paw Mussel Portion of the Action Area

Available usage data (Kaul, et al., 2005) suggest that the heaviest usage of atrazine relative to the action area is likely to be in a band stretching from eastern Iowa across Illinois and Indiana to Ohio with decreasing intensity south and east of this area. As noted above, the action area was segmented into regions to allow for modeling that covers the expected range of runoff vulnerability. All existing PRZM scenarios were evaluated, and a subset was selected for use in this assessment. The scenarios were selected to provide a spatial context to predicted exposures.

Currently a suite of 63 PRZM standard scenarios and 7 Barton Springs scenarios (recently developed for use in the Barton Springs salamander endangered species risk assessment (U.S. EPA, 2006c), are available for use in ecological risk assessments representing predominantly agricultural uses. Each scenario is intended to represent a high-end exposure setting for a particular crop. Scenario locations are selected based on various factors including crop acreage, runoff and erosion potential, climate, and agronomic practices. Once a location is selected, a scenario is developed using locally specific soil, climatic, and agronomic data. Each PRZM scenario is assigned a specific climatic weather station providing 30 years of daily weather values.

Specific scenarios were selected for use in this assessment using two criteria. First, an evaluation of all available PRZM scenarios was conducted, and those scenarios that represent atrazine uses (e.g. Ohio corn) were selected for modeling. Weather information was assigned to these scenarios at development. Second, an additional suite of scenarios was identified to represent both agricultural and non-agricultural uses for which scenarios within the action area are not available (e.g. Barton Springs residential). These scenarios were used in the assessment as surrogates for atrazine uses without current scenarios (e.g. Oregon Christmas tree as surrogate for forestry) and to provide geographic coverage where no current scenario exists (e.g. Ohio corn scenario modeled using Springfield, Missouri weather data).

This approach is deemed protective for a variety of reasons. All PRZM/EXAMS scenarios have been developed to represent high end exposures for either a national or regional objective. For example, the Mississippi cotton scenario was originally developed to provide a national estimate for pesticide use on cotton, while subsequent cotton scenarios in California, Texas, and North Carolina were developed to provide a regional context to exposure from pesticide use on cotton. All PRZM/EXAMS scenarios are developed to represent high end conditions (climatic, soil, agronomic) that will yield high exposures for a given area. The goal of scenario development is to yield EECs that are representative of the highest exposures expected to occur nationally and regionally. Key parameters driving exposure predictions from PRZM/EXAMS are curve number, slope, and rainfall. Curve numbers for each scenario are selected to represent a reasonable worst case situation and are selected by soil hydrologic group (i.e. A, B, C, or D soils). In this context, a D soil will have a higher curve number than a C soil and yield a higher EEC. In general, all PRZM/EXAMS scenarios are developed with either C or D soils. Similarly, the slope for a given scenario is typically selected to represent a high end of slopes associated with the use site being modeled. The combination of curve number and slope with rainfall will yield relatively high EECs for the area where that rainfall occurs. When using a surrogate scenario, moving the high end scenario from a relatively dry region (such as the Oregon Christmas tree scenario with a curve number based on a C soil and a 4% slope) to a wetter region (such as the Midwest where soil types and slopes are similar but rainfall is greater) should provide a reasonably protective exposure estimate for the Midwest.

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

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

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

For this assessment, available PRZM weather stations were associated with watersheds highly vulnerable to atrazine runoff. As shown in Figure 3.5**,** weather stations associated with Sioux City, Iowa; Springfield, Missouri; Evansville/Indianapolis, Indiana; and Mobile, Alabama were selected to represent highly vulnerable locations for modeling surrogate scenarios (both agricultural and non-agricultural). As such, surrogate scenarios used to model this region were run using weather data from these locations to represent exposures within the entire region.

For this assessment, the following corn scenarios were modeled to represent all the various regions of the action area: North Dakota (using weather data from Fargo) representative of the upper Great Plains states; Illinois and Ohio (using weather data from Peoria and Dayton, respectively) representative of the northern tier states; Mississippi (using the weather data from Mobile, Alabama) representative of the southern tier states; and the Ohio (using the Springfield, Missouri weather data) representative of the western states. The Kansas sorghum scenario (the only existing sorghum scenario) was modeled with local weather stations including Topeka, Kansas (western states), Sioux City, Iowa (upper great plain states), and Mobile, Alabama (southern states).

Currently, the only non-agricultural scenarios available for use in aquatic exposure assessment are those developed specifically for the Barton Springs Salamander Endangered Species Risk Assessment (U.S. EPA, 2006c). For the Barton Springs assessment, a suite of non-agricultural scenarios was developed including a residential, impervious (to be used in tandem with the residential scenario), and rights-of-way scenarios. These scenarios were used in this assessment in a manner similar to the agricultural scenarios described above. Each scenario was modeled using a representative weather station for each region. For example, the residential scenario was modeled using the Mobile, Alabama weather data to represent exposures in the southern states, while the same scenario was modeled with the Sioux City, Iowa weather data to represent the upper Great Plains states, the Indianapolis weather data to represent the northern tier states, and the Springfield, Missouri weather data to represent the western states. A summary of all the modeled scenarios along with associated weather information is included in Table 3.5.

Both the agricultural and non-agricultural scenarios were used within the standard framework of PRZM/EXAMS modeling using the standard graphical user interface (GUI) shell, PE4v01.pl. The models and GUI used in this assessment may be found at the following website:

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

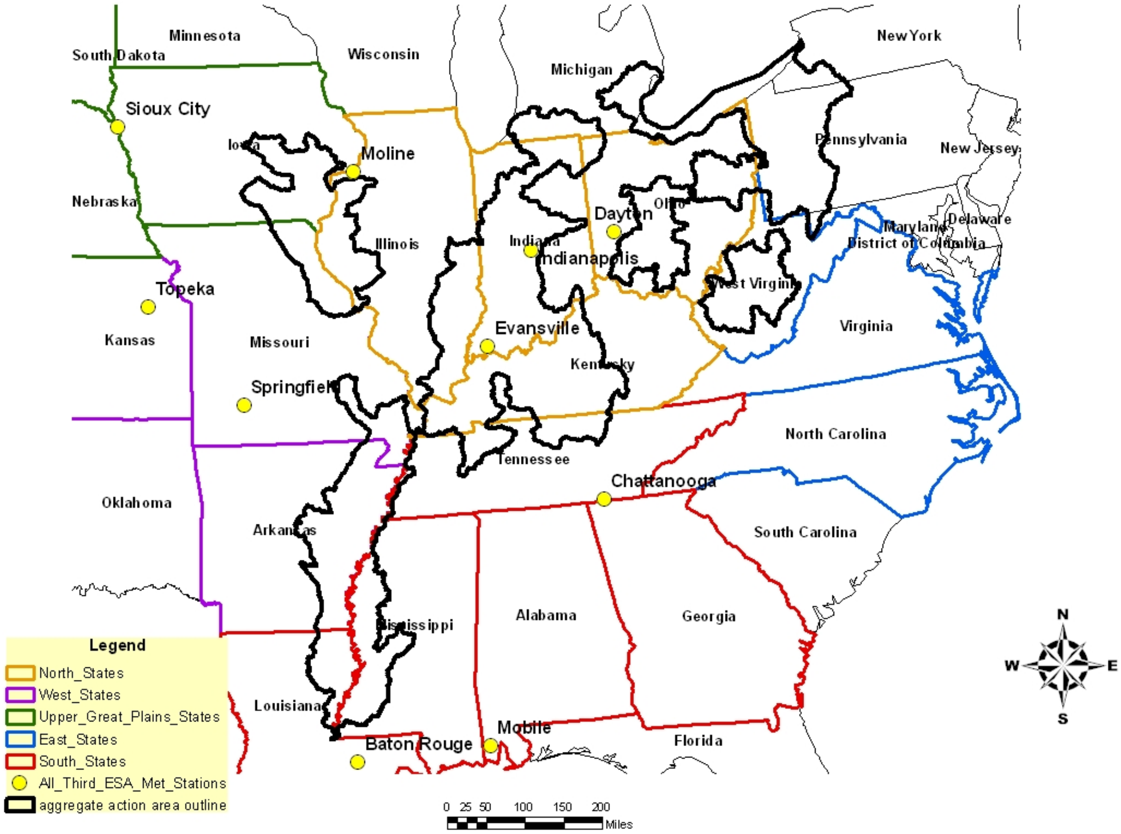

Figure 3.5 Location of Various Weather Stations Used to Model Agricultural and Non-agricultural Scenarios

Table 3.5 Summary of PRZM Scenarios				
Region	**Use**	**Scenario**	**First Application**	**Weather Station (WBAN #)**
South	Corn	MS corn	April 1	Mobile, AL (13894)
	Sorghum	KS sorghum	May 1	Mobile, AL (13894)
	Fallow	BSS meadow[a]	November 15	Mobile, AL (13894)
	Residential	BSS residential	April 1	Mobile, AL (13894)
	Right-of-way	BSS row	June 1	Mobile, AL (13894)
	Forestry	OR xmastree	June 1	Mobile, AL (13894)
	Turf	BSS turf	April 1	Mobile, AL (13894)
North	Corn	OH corn IL corn	April 15	Dayton, OH (93815) Moline, IL (14923)
	Sorghum	KS sorghum	May 1	Evansville, IN (93817)
	Fallow	BSS meadow	October 15	Evansville, IN (93817)
	Residential	BSS residential	May 1	Indianapolis, IN (93819)
	Right-of-way	BSS row	June 1	Indianapolis, IN (93819)
	Forestry	OR xmastree	June 1	Evansville, IN (93819)
	Turf	BSS turf	May 1	Indianapolis, IN (93819)
West	Corn	IL corn	April 15	Springfield, MO (13995)
	Sorghum	KS sorghum	May 1	Topeka, KS (13996)
	Fallow	BSS meadow	November 1	Springfield, MO (13995)
	Residential	BSS residential	April 15	Springfield, MO (13995)
	Right-of-way	BSS row	June 1	Springfield, MO (13995)
	Forestry	OR xmastree	June 1	Springfield, MO (13995)
	Turf	BSS turf	April 15	Springfield, MO (13995)
Upper Great Plains	Corn	ND corn	April 1	Fargo, ND (14914)
	Sorghum	KS sorghum	May 1	Sioux City, SD (14943)
	Fallow	BSS meadow	November 1	Sioux City, SD (14943)

Table 3.5 Summary of PRZM Scenarios				
Region	**Use**	**Scenario**	**First Application**	**Weather Station (WBAN #)**
	Residential	BSS residential	May 1	Sioux City, SD (14943)
	Right-of-way	BSS row	June 1	Sioux City, SD (14943)
	Forestry	OR xmastree	June 1	Sioux City, SD (14943)
	Turf	BSS turf	May 1	Sioux City, SD (14943)

[a] BSS scenarios developed for Barton Springs Salamander (BSS) Endangered Species Risk Assessment (U.S. EPA, 2006c).

Peak concentrations, as well as rolling time-weighted averages of 14 days, 21 days, 30 days, 60 days, and 90 days were derived for comparison with the appropriate ecotoxicity endpoints (including the community-level threshold concentrations) for atrazine.
The 30-year time series output file was used to recalculate the peak, 14-day, 21-day, 30-day, 60-day, and 90-day rolling averages at the 90th percentile. All model outputs were post-processed manually using Microsoft Excel to provide the equivalent of the standard one in ten year return frequency exposures, as predicted by PRZM/EXAMS. A sample of how this post-processing was conducted may be found in the previous atrazine assessments for the Chesapeake Bay and Alabama Sturgeon (EPA, 2007a and 2007b).

Additional information on the modeling approach for the non-agricultural residential, rights-of-way, and forestry use scenarios may also be found in the previous atrazine endangered species risk assessments (U.S. EPA, 2006c, 2007a, 2007b, and 2007c).

3.2.3 Model Inputs

The estimated concentrations from surface water sources were calculated using Tier II PRZM (Pesticide Root Zone Model) and EXAMS (Exposure Analysis Modeling System). PRZM is used to simulate pesticide transport as a result of runoff and erosion from a standardized watershed, and EXAMS estimates environmental fate and transport of pesticides in surface waters. The linkage program shell (PE4v01.pl) that incorporates the site-specific scenarios was used to run these models.

Scenarios used in this assessment consist of agricultural uses for corn and sorghum developed previously. Other scenarios representing areas outside the action area were modeled using weather stations specific to the action area in order to represent atrazine uses where no scenario existed within the action area including one agricultural use (fallow/idle land) and several non-agricultural uses (residential, turf, forestry, and rights-or-way). All scenarios were modeled using local weather data selected to represent the highest rainfall potential in a region as described above. Linked use site-specific scenarios and meteorological data were used to estimate exposure as a result of specific use for each modeling scenario. The PRZM/EXAMS model was used to calculate concentrations using the standard ecological water body scenario in EXAMS. Weather and agricultural practices were simulated over 30 years so that the 1 in 10 year exceedance probability at the site was estimated for the standard ecological water body.

One outcome of the 2003 IRED process was a modification to all existing atrazine labels that requires setback distances around intermittent/perennial streams and lakes/reservoirs. The label changes specify setback distances of 66 feet and 200 feet for atrazine applications surrounding intermittent/perennial streams and lakes/reservoirs, respectively. The Agency incorporated these distances into this assessment and has modified the standard spray drift assumptions accordingly using AgDrift to estimate the impact of a setback distance of 66 feet on the fraction of drift reaching a surface water body. The revised spray drift percentages, which are incorporated into the PRZM/EXAMS modeling, are 0.6% for ground applications and 6.5% for aerial applications.

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

The date of first application was developed based on several sources of information including data provided by BEAD and Crop Profiles maintained by the USDA. In general, the date of first application was selected to represent the most vulnerable window of exposure. Typical use patterns for atrazine as a pre-emergent herbicide show that the majority of first applications occur during the spring planting/emergence season (an exception to this is the treatment for fallow land, which is typically applied post-emergence and expected to occur in the fall). More detail on the crop profiles and the previous assessments may be found at:

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

The appropriate PRZM input parameters were selected from the environmental fate data submitted by the registrant and in accordance with US EPA-OPP EFED water model parameter selection guidelines, Guidance for Selecting Input Parameters in Modeling the Environmental Fate and Transport of Pesticides, Version 2.3, February 28, 2002. These parameters are consistent with those used in both the 2003 IRED (U.S. EPA, 2003a) and the cumulative triazine risk assessment (U.S. EPA, 2006a) and are summarized in Table 3.6. More detail on these assessments may be found at:

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

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

Table 3.6 Summary of PRZM/EZAMS Environmental Fate Data Used for Aquatic Exposure Inputs for Atrazine Three Listed Mussels Assessment		
Fate Property	**Value**	**MRID[a] (or source)**
Molecular Weight	215.7	MRID 41379803
Henry's constant	2.58 x10 -9	MRID 41379803
Vapor Pressure	3 x 10 -7	MRID 41379803
Solubility in Water	33 mg/l	MRID 41379803
Photolysis in Water	335 days	MRID 42089904
Aerobic Soil Metabolism Half-lives	152 days	MRID 40431301 MRID 40629303 MRID 42089906
Hydrolysis	stable	MRID 40431319
Aerobic Aquatic Metabolism (water column)	304 days	2x aerobic soil metabolism rate constant
Anaerobic Aquatic Metabolism (benthic)	608 days	MRID 40431323
Koc	88.78 ml/g	MRID 40431324 MRID 41257901 MRID 41257902 MRID 41257904 MRID 41257905 MRID 41257906
Application Efficiency	95 % for aerial 99 % for ground	Default value[b]
Spray Drift Fraction	6.5 % for aerial 0.6 % for ground	AgDrift adjusted values based on label restrictions

[a] Master Record Identification (MRID) is record tracking system used within OPP to manage data submissions to the Agency. Each data submission if given a unique MRID number for tracking purposes.
[b] Inputs determined in accordance with EFED "*Guidance for Chemistry and Management Practice Input Parameters for Use in Modeling the Environmental Fate and Transport of Pesticides*" dated February 28, 2002.

3.2.4 Results

As noted above, a total of seven scenarios were evaluated in this assessment. Of these, four were developed as part of the Barton Springs salamander endangered species risk assessment (U.S. EPA, 2006c). Two of the Barton Springs scenarios (residential and rights-of-way) were used in tandem with an impervious scenario, while two (fallow/idle land and turf) are standard PRZM/EXAMS scenarios. The remaining three scenarios (corn, sorghum, and Christmas trees as surrogate for forestry) were taken from existing scenarios developed for other regions of the United States and modeled using local weather data. No new scenarios were developed specifically for this assessment. The results of the modeling are summarized in Table 3.7. An example of the modeling approach and the model input files may be found in Appendix D of the previous endangered species risk assessments for atrazine (EPA, 2006c, 2007a, b, and c).

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

As previously discussed in Section 3.2.2 and summarized in Table 3.4, none of the three listed mussels span the entire range of the areas being assessed for exposure. The fat pocketbook mussel occurs in the south, north, west, and upper Great Plains regions; the northern riffleshell occurs in the north and east regions; and the PCPP mussel is found in the northern region.

Table 3.7 Summary of PRZM/EXAMS Output Screening-Level EECs for all Modeled Scenarios (Using the Standard Water Body)									
Region	Use Site	Application Rate (lbs/acre)	No. of Applications	90th Percentile of 30 Years of Output					
				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)
South	Corn	2.0	2 (not to exceed 2.5 lbs/year)	109.1	107.8	107.0	106.3	103.9	101.4
South	Sorghum	2.0	1	63.6	62.9	62.4	61.7	59.6	57.4
South	Fallow	2.25	1	58.8	58.2	58.0	57.6	56.6	55.6
South	Residential[a] Granular	2.0	2 (not to exceed 4.0 lbs/year)	19.9	19.6	19.4	19.2	18.6	17.9
South	Residential[a] Liquid	1.0	2 (not to exceed 2.0 lbs/year)	14.6	14.4	14.2	14.1	13.7	13.4
South	Rights-of-way[b]	1.0	1	2.4	2.4	2.4	2.4	2.3	2.2
South	Forestry	4.0	1	46.1	45.2	44.7	44.1	42.2	40.8
South	Turf Granular	2.0	2 (not to exceed 4.0 lbs/year)	17.9	17.7	17.7	17.7	17.6	17.1
South	Turf Liquid	1.0	2 (not to exceed 2.0 lbs/year)	14.8	14.6	14.4	14.3	13.7	13.1
East	Corn	2.0	2	83.3	82.1	81.8	81.6	79.7	77.8
East	Sorghum	2.0	1	69.2	68.3	68.1	67.6	65.9	63.8
East	Fallow	2.25	1	54.7	54.2	54.0	54.0	53.8	53.7

Table 3.7 Summary of PRZM/EXAMS Output Screening-Level EECs for all Modeled Scenarios (Using the Standard Water Body)									
Region	Use Site	Application Rate (lbs/acre)	No. of Applications	90th Percentile of 30 Years of Output					
				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)
East	Residential[a] Granular	2.0	2	13.3	13.3	13.3	13.3	13.3	13.3
East	Residential[a] Liquid	1.0	2	9.6	9.5	9.4	9.3	9.0	8.8
East	Rights-of-way[b]	1.0	1	2.4	2.4	2.3	2.3	2.3	2.2
East	Forestry	4.0	1	44.2	43.5	43.1	42.7	41.2	40.2
East	Turf Granular	2.0	2	13.2	13.2	13.2	13.2	13.2	13.2
East	Turf Liquid	1.0	2	9.1	9.1	9.1	9.1	9.1	9.1
North	Corn[c]	2.0	2 (not to exceed 2.5 lbs/year)	100.8	100.3	99.9	99.3	97.5	96.2
North	Sorghum	2.0	1	58.4	57.7	57.4	56.9	54.9	52.8
North	Fallow	2.25	1	51.6	51.5	51.5	51.5	51.0	50.4
North	Residential[a] Granular	2.0	2 (not to exceed 4.0 lbs/year)	9.9	9.9	9.9	9.9	9.9	9.9
North	Residential[a] Liquid	1.0	2 (not to exceed 2.0 lbs/year)	7.6	7.5	7.5	7.5	7.5	7.4
North	Rights-of-way[b]	1.0	1	2.7	2.7	2.7	2.6	2.6	2.5
North	Forestry	4.0	1	48.5	47.7	47.2	46.7	44.9	43.3

Table 3.7 Summary of PRZM/EXAMS Output Screening-Level EECs for all Modeled Scenarios (Using the Standard Water Body)									
Region	**Use Site**	**Application Rate (lbs/acre)**	**No. of Applications**	**90th Percentile of 30 Years of Output**					
				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)**
North	Turf Granular	2.0	2 (not to exceed 4.0 lbs/year)	7.1	7.1	7.1	7.1	7.1	7.1
North	Turf Liquid	1.0	2 (not to exceed 2.0 lbs/year)	6.6	6.6	6.6	6.6	6.5	6.5
West	Corn	2.0	2 (not to exceed 2.5 lbs/year)	92.8	91.7	91.4	90.7	88.0	85.4
West	Sorghum	2.0	1	60.1	59.4	58.9	58.4	57.3	56.3
West	Fallow	2.25	1	103.4	103.1	103.1	103.1	103.0	103.0
West	Residential[a] Granular	2.0	2 (not to exceed 4.0 lbs/year)	11.9	11.8	11.7	11.6	11.3	11.0
West	Residential[a] Liquid	1.0	2 (not to exceed 2.0 lbs/year)	9.9	9.7	9.7	9.6	9.3	9.1
West	Rights-of-way[b]	1.0	1	3.8	3.8	3.8	3.8	3.6	3.5
West	Forestry	4.0	1	27.4	26.9	26.8	26.5	25.6	24.8
West	Turf Granular	2.0	2 (not to exceed 4.0 lbs/year)	7.2	7.1	7.0	7.0	6.7	6.5
West	Turf Liquid	1.0	2 (not to exceed 2.0 lbs/year)	7.6	7.5	7.5	7.5	7.4	7.2

Table 3.7 Summary of PRZM/EXAMS Output Screening-Level EECs for all Modeled Scenarios (Using the Standard Water Body)									
Region	**Use Site**	**Application Rate (lbs/acre)**	**No. of Applications**	**90th Percentile of 30 Years of Output**					
				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)**
Upper Great Plains	Corn	2.0	2 (not to exceed 2.5 lbs/year)	84.8	84.0	83.6	83.5	82.3	80.8
Upper Great Plains	Sorghum	2.0	1	57.2	56.6	56.3	55.8	54.4	52.8
Upper Great Plains	Fallow	2.25	1	49.2	49.1	49.1	49.1	49.1	48.8
Upper Great Plains	Residential[a] Granular	2.0	2 (not to exceed 4.0 lbs/year)	10.9	10.9	10.9	10.8	10.8	10.8
Upper Great Plains	Residential[a] Liquid	1.0	2 (not to exceed 2.0 lbs/year)	8.2	8.1	8.1	8.0	7.8	7.6
Upper Great Plains	Rights-of-way[b]	1.0	1	3.3	3.2	3.2	3.2	3.1	3.0
Upper Great Plains	Forestry	4.0	1	64.5	61.0	60.7	60.2	58.3	56.5
Upper Great Plains	Turf Granular	2.0	2 (not to exceed 4.0 lbs/year)	10.1	10.1	10.1	10.1	10.0	9.9
Upper Great Plains	Turf Liquid	1.0	2 (not to exceed 2.0 lbs/year)	8.2	8.1	8.1	8.0	8.0	7.9

a Assumes 1% overspray of atrazine to the impervious surfaces.
b Assumes that10% of the watershed is in rights-of-way. Rationale for selection of 10% treated area was documented in previous assessment (EPA, 2006c)
c A second corn scenario for Ohio was modeled, but is not presented because it yielded a lower EEC.

3.2.5 Additional Modeling Exercises Used to Characterize Potential Exposures

Additional characterization of the screening-level modeling results has been completed, including a characterization of the importance of flowing water, a detailed analysis of monitoring data, and alternative modeling assumptions. These refined predictions are used to characterize the exposures used in risk estimation and are not directly used to calculate RQs. These analyses are described in the sections that follow.

3.2.5.1 Impact of Flowing Water on Modeled EECs

The Agency's standard ecological assessment for aquatic organisms relies on estimates of exposure derived from PRZM/EXAMS using the standard water body. The standard water body is a 1-hectare pond that is 2 meters deep with a total volume of 20,000,000 liters and is modeled without flow. The standard water body was developed in order to provide an approximation of high end exposures expected in ponds, lakes, and perennial/intermittent streams adjacent to treated agricultural fields. Typically, this has been interpreted as a stream with little, or low flow. For pesticides with low to moderate persistence, the standard water body provides a reasonably high end estimate of exposure in headwater streams and other low flow water bodies for both acute and longer-term exposures. For more persistent compounds, the non-flowing nature of the standard water body provides a reasonable high end estimate of peak exposure for many streams found in agricultural areas; however, it appears to over-estimate exposure for longer time periods in all but the most static water bodies.

In order to further characterize the impact of larger water bodies with flow, each selected scenario was also modeled using the Index Reservoir as the receiving water body. The Index Reservoir represents a 5.3-hectare water body draining a 172-hectare watershed. In the case of the Index Reservoir, the standard approach is to allow EXAMS to estimate total runoff accumulated from the 172-hectare watershed and route that volume of water as flow through the reservoir while assuming no change in reservoir volume. The estimated flow rate within EXAMS was over-written using the lowest USGS value specific for the listed mussel occurring within a particular region, and the impact of flow on peak and long-term EEC was evaluated. Unlike a standard drinking water assessment, these values were not adjusted for percent cropped area (PCA). Therefore, dilution is not factored into the EEC, which leads to a conservative estimate of exposure. More information on the Index Reservoir and USGS flow data may be found at:

http://www.epa.gov/oppfead1/trac/science/reservoir.pdf

http://waterdata.usgs.gov/nwis/sw

The three listed mussels reside in 2^{nd}, 3^{rd}, and higher order streams in the lower Mississippi River basin, the lower Missouri River basin, and the Ohio River basin with moderate to strong flow rates. The hydrologic landscape of the three listed mussel's action area is diverse and has been broken into five regions (east, north, south, west, and upper Great Plains), as shown in Figure 3.1. Four of these regions (east, north, west, and south) were previously assessed for the Alabama sturgeon (U.S. EPA, 2007b) and eight listed mussels (U.S. EPA 2007c). The fifth region (upper Great Plains) is occupied by the fat pocketbook in only two locations in the far southeastern corner of Iowa. Given the tangential nature of the co-occurrence of species locations within the regionalized exposure approach, the fact that these occurrences are documented in the Nature Serve database based on the presence of "dead shells", and that exposures in the upper Great Plains region are generally lower than the other four regions, refined flow-adjusted modeling was not conducted for this area. It is expected that any occurrence of the fat pocketbook mussel in southeastern Iowa will be conservatively represented by exposures from the western and northern regions.

A comparison of the site-specific flow rate information for the three listed mussels included in this assessment with the eight listed mussels included in the previous atrazine endangered species assessment (U.S. EPA, 2007c) indicates that the fat pocketbook, PCPP mussel, and northern riffleshell mussels occupy streams with similar and higher flow rates. Site-specific flow rates for these three listed mussels (provided in Table 3.13) range from 28 to 4,728 ft^3/sec as compared with flow rates ranging from 22 to 105 ft^3/sec for the eight listed mussels evaluated in the previous U.S. EPA 2007c assessment. Given that the three listed mussels occupy streams with similar or higher flow than those mussels previously assessed, flow-adjusted EECs for the four regions (east, north, west, and south) previously evaluated are used again in this assessment. In addition, there has been no change in atrazine use rates since completion of the February 2007 atrazine mussel assessment.

An analysis of the impact of flowing water on modeled EECs was completed to characterize the representativeness of the static water body EEC to the habitat of the assessed species. In general, the analysis from the previous assessment (U.S. EPA, 2007c) showed that long-term screening-level EECs (e.g. 30-day average) were reduced to concentrations below levels of concern by adjusting the EECs to account for low to moderate flow rates (between 22 and 105 ft^3/sec) consistent with those where the three listed mussels reside. Although these mussels also occupy streams with higher flow rates, use of low to moderate flow rates used in the previous assessment is assumed to be representative of lower flow conditions (and potentially higher atrazine concentrations) where the three listed mussels occur. Use of low to moderate flow rates is assumed to be protective of the three listed mussels because higher flow rates, which are specific for the streams in which the listed mussels occur, would yield lower atrazine concentrations. The results along with the flow rates used in this evaluation are presented in Table 3.8. As expected, the flow-adjusted EECs are lower than EECs from the standard static ecological water body. Impact of flow on the EECs is greater as flow rate and exposure duration increases.

Table 3.8 Comparison of Alternative PRZM Modeling (assuming flow) with EECs Generated Using the Static Water Body							
Scenario	**Flow (ft^3/sec)**	**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)**
South Region							
South corn with static water body[a]	0	109.1	107.8	107.0	106.3	103.9	101.4
South corn (IR) with mean seasonal flow from USGS stream data[c]	105	113	14	10	7	3	2
Percent decrease in EEC using USGS mean seasonal flow data		na	87	91	93	97	98
East Region							
East corn with static water body[a]	0	83.3	82.1	81.8	81.6	79.7	77.8
East corn (IR) with mean seasonal flow from USGS stream data[c]	110	64	9	6	4	2	2
Percent decrease in EEC using USGS mean seasonal flow data		23	89	93	95	97	97
North Region							
North corn with static water body[a]	0	100.8	100.3	99.9	99.3	97.5	96.2
North corn (IR) with mean seasonal flow from USGS stream data[c]	22	65	16	12	8	4	3
Percent decrease in EEC using USGS mean seasonal flow data		36	84	88	92	96	97
West Region							
West fallow with static water body[a]	0	103.4	103.1	103.1	103.1	103.0	103.0
West fallow (IR) with mean seasonal flow from USGS stream data[c]	90	74	7	5	4	2	1

Table 3.8 Comparison of Alternative PRZM Modeling (assuming flow) with EECs Generated Using the Static Water Body							
Scenario	**Flow (ft^3/sec)**	**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)**
Percent decrease in EEC using USGS mean seasonal flow data		29	93	95	97	98	99

[a] EECs generated using PE4v01.pl in this table are slightly different from those presented in Table 3.7 due to different duration of exposure and slight differences in the manual estimation technique used in Table 3.6.
[b] Index Reservoir scenarios EEC are typically reported using percent cropped area (PCA) of 46% for corn and 87% for fallow. In this characterization no PCA is applied to the modeled output.
[c] USGS flow data reported as annual mean or annual seasonal (April to September) mean values.

3.2.6 Existing Monitoring Data

The second step in the process of characterizing EECs used for risk description is to compare the modeling results with available surface water monitoring data. A fairly robust set of surface water monitoring data exists for atrazine from a variety of targeted and non-targeted studies. Targeted studies are those studies whose design is specifically tailored to the use pattern for a specific compound. Sample location, number of samples, frequency of sampling, and sample collection timing are specifically designed to capture exposures for the target compound. Non-targeted monitoring is typically more general in nature and is not designed for a specific compound. The study design for non-targeted studies are typically broad with the intent of capturing as many compounds as possible, but not necessarily focused on the main exposure period for a single compound.

Atrazine data from the USGS NAWQA program (http://water.usgs.gov.nawqa), Watershed Regression for Pesticides (WARP), Heidelberg College, Community Water System (CWS) data from drinking water sources, published USGS studies, and data recently submitted by the atrazine registrants (AEMP: Atrazine Ecological Monitoring Program) are included in this assessment. These monitoring data are characterized in terms of general statistics including number of samples, frequency of detection, maximum concentration, and mean from all detections. In general, the targeted monitoring data are relevant to the fat pocketbook and northern riffleshell mussels that occur in lower flow vulnerable watersheds because the data were collected from the most vulnerable watersheds. The majority of the other data, though non-targeted to atrazine, are also relevant because the sample locations generally co-occur with the species' location, although sample frequency and timing may not be specifically designed to match atrazine use patterns.

3.2.6.1 Atrazine Ecological Monitoring Program (AEMP) Data

The 2003 IRED required the atrazine registrants to conduct watershed monitoring for atrazine as a condition of re-registration. One component of the monitoring program is focused on flowing water bodies, and provides two to three years of monitoring data, accrued over a three-year period (2004-2006), in the most vulnerable watersheds associated with corn and sorghum production. These data are targeted specifically to atrazine use and are designed to represent exposure in the watersheds most prone to atrazine runoff. In this case, vulnerability was defined using the USGS WARP model. The principal factors influencing WARP predictions of exposure and hence the vulnerability ranking are:

- Atrazine use,
- Rainfall intensity,
- Soil erodibility,
- Watershed area, and
- Dunne overland flow

3.2.6.1.1 AEMP Study Design

Surface water data included in this study were collected using a targeted methodology that relied on WARP to identify the upper 20th percentile of vulnerable watersheds and a statistical design to select a subset of 40 watersheds that may be representative of 1,172 vulnerable watersheds. The atrazine use input was derived by calculating the mean annual atrazine concentration (at the 95th percent confidence limit) across all watersheds in the United States where atrazine is used. Given the statistical nature of the sampling design of this study, it is not possible to extrapolate the monitoring data from the 40 watersheds beyond the upper 20th percentile of watersheds (i.e., the 1,172 vulnerable watersheds).

Samples were collected from 20 locations within the designated watersheds every four days during the peak use period for atrazine (April to August) during the 2004-2005 growing season, and a second set of 20 watersheds were sampled during the 2005-2006 growing season (several watersheds from the 2004-2005 sample period were carried over for a third year of monitoring). The strength of this data set is the targeted nature of site selection to areas of high atrazine use, the frequency of the sampling (every four days during peak use season), and the collection of multiple samples on selected days from a number of sites that allows for a statistical description of the variability surrounding the time series data. More detail on the approach, methodology and objectives of the surface water AEMP for atrazine may be found at:

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

3.2.6.1.2 AEMP Results

A preliminary analysis of the AEMP data from 2004 to 2006 has been completed. The data have been statistically evaluated for each site/year combination, including number of non-detections, frequency of detection, maximum concentration, mean concentration, median concentration, and number of scheduled samples that ultimately did not occur or samples that were not subsequently analyzed. These statistics provide a general picture of the level of exposures seen in these data relative to the other data sets described in this assessment. The AEMP data, including site-specific flow information for the sampled locations, is provided in Appendix D.

Overall, the data suggest a similar pattern of atrazine exposure in surface water as in the other data sets evaluated as part of this assessment. Atrazine was detected in a total of 2,979 out of 3,601 samples for an overall frequency of detection of 79%. The frequency of detection ranged across all watersheds and years from a maximum of 100% to a minimum of 11%. The maximum concentration detected from all watersheds was 208.8 μg/L from the Indiana 11 site in 2005. The mean annual concentrations ranged from a maximum of 9.5 μg/L from the Missouri 01 site in 2004 to a low of 0.1 μg/L for the Nebraska 06 site in 2006, while the median values ranged from 4.2 μg/L for the Missouri 02 site in 2004 to 0.1 μg/L for the Ohio 03 site in 2004. It should be noted that a number of watersheds, particularly in Nebraska, experienced dry periods where scheduled sampling did not take place; therefore, the statistics for those watersheds may not represent actual conditions expected in normal or wetter years.

This data set is currently releasable only upon completion and submission of an Affirmation of Non-multinational Status form under section 10(g) of FIFRA. Information on how to submit a request to obtain a copy of the data may be obtained from the following website:

http://www.epa.gov/espp/atrazine_ewm_data.htm

In addition, the site selection process was focused on watersheds deemed to be highly vulnerable to atrazine runoff based on use, soil, and climatic conditions and were selected to be statistically representative of the 1,172 watersheds from the highly vulnerable area. As seen in Figure 3.6, a sub-set of the 1,172 watersheds from which the 40 watersheds were selected are within the aggregated action area. A similar analysis shows limited overlap between the 1,172 watersheds and individual species locations. The overlap for the fat pocketbook, northern riffleshell, and PCPP mussel is shown in Figures 3.7 through 3.9, respectively.

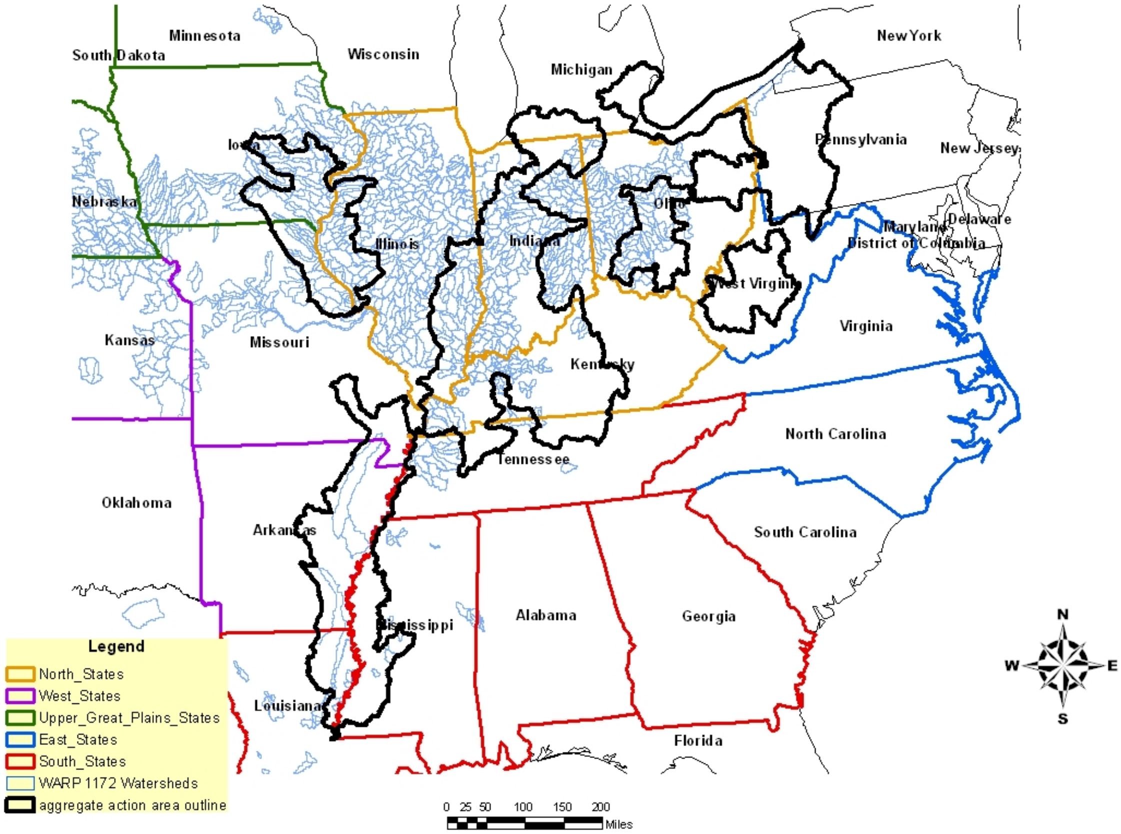

Figure 3.6 Relationship of WARP Vulnerable Watersheds Relative to Aggregated Action Area

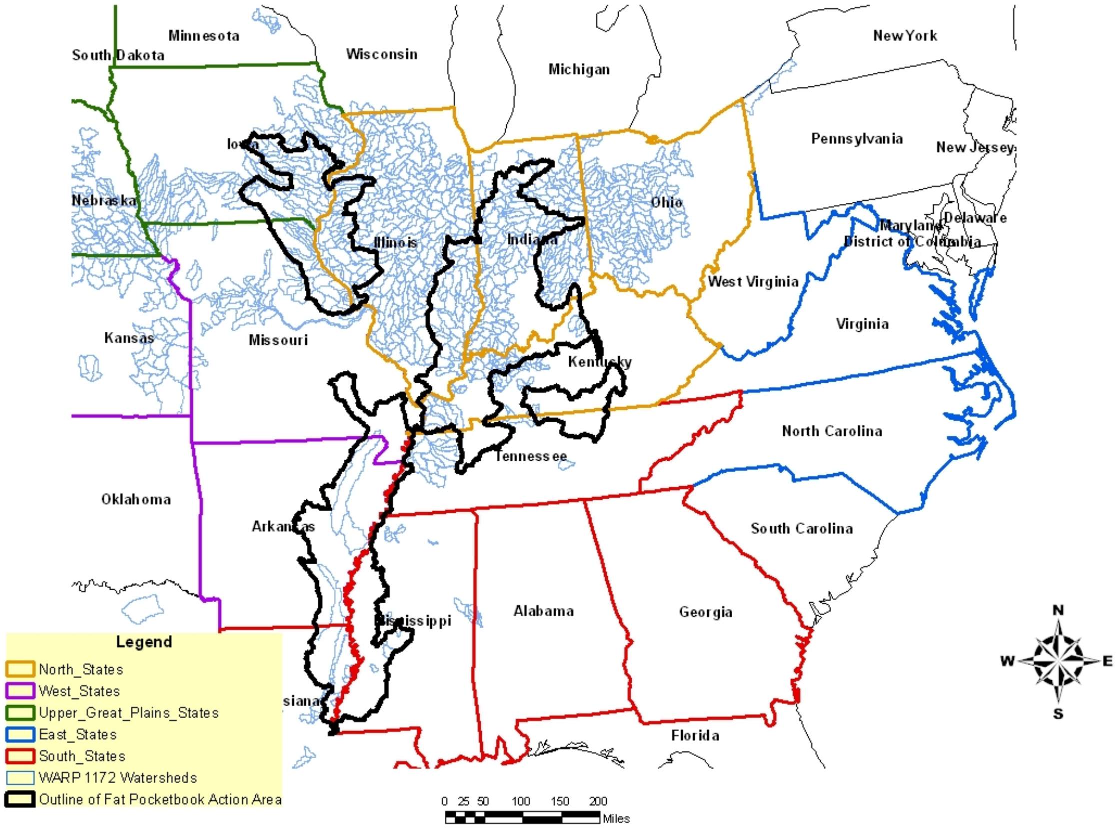

Figure 3.7 Relationship of WARP Vulnerable Watersheds Relative to the Fat Pocketbook Mussel Portion of the Action Area

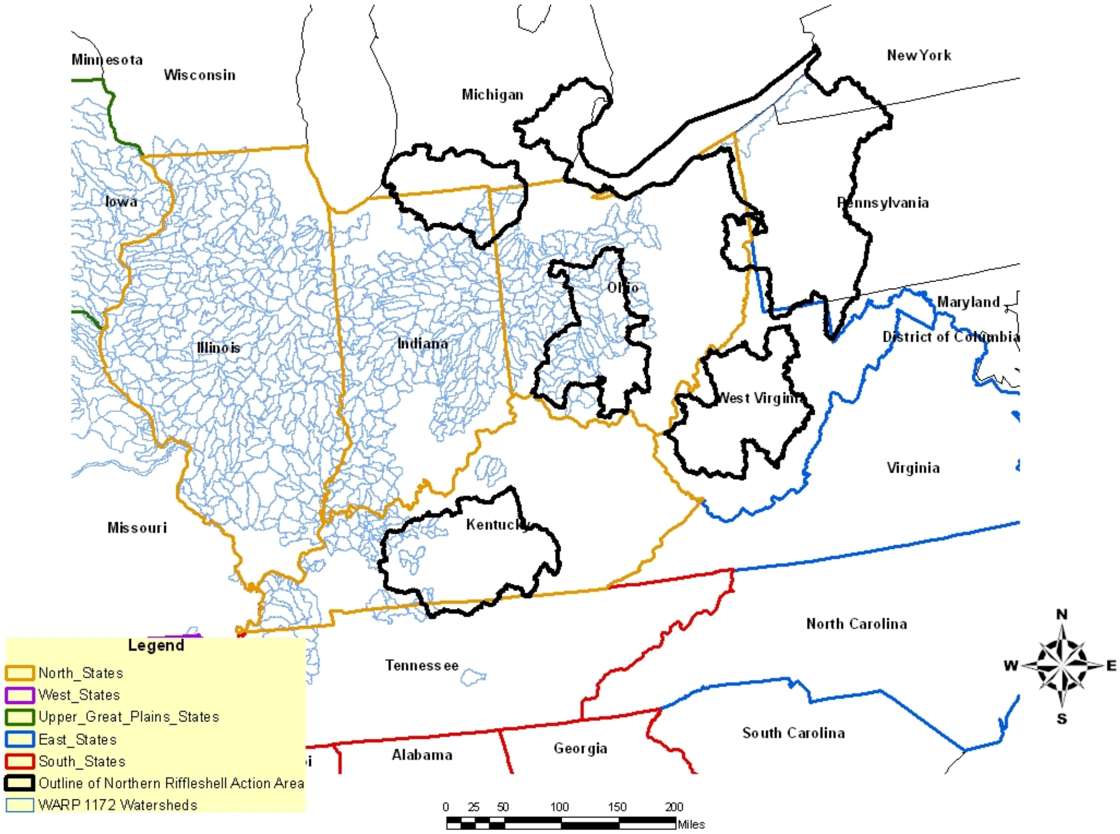

Figure 3.8 Relationship of WARP Vulnerable Watersheds Relative to the Northern Riffleshell Mussel Portion of the Action Area

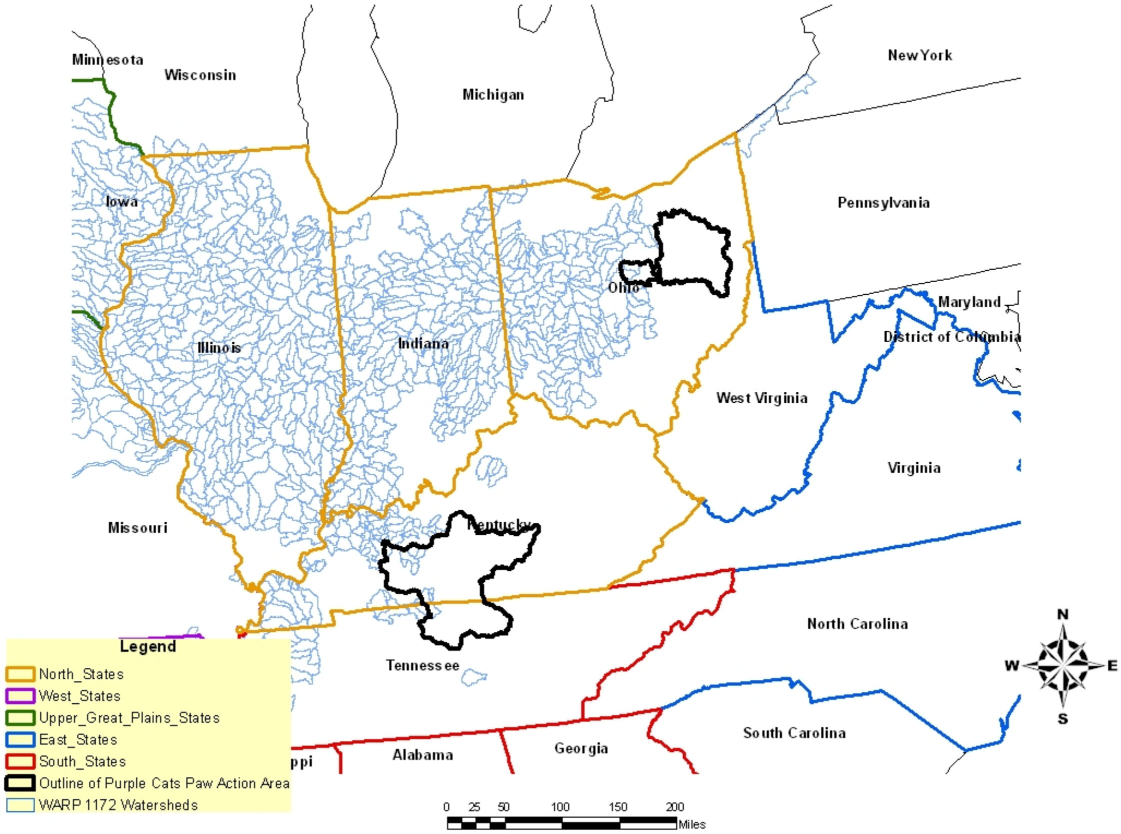

Figure 3.9 Relationship of WARP Vulnerable Watersheds Relative to the Purple Cat's Paw Mussel Portion of the Action Area

The statistical nature of the study design is critical in the selection of the 40 watersheds to sample. Watersheds were selected using a generalized random tessellation stratified (GRTS) method to identify spatially representative locations that can be linked back to the entire population of 1,172 watersheds. In general, most of the sites within watersheds selected for monitoring are second and third order streams in high atrazine use areas deemed vulnerable to runoff (a few of these sites are first and fourth order streams). The sampling locations were selected from a set of 1,172 watersheds using a statistical design, and thus are representative of some proportion of the total 1,172 watersheds. Comparison of the site locations from the ecological monitoring data with the action area for the listed mussels indicates that 18 of the 40 sites are within the action area (Figure 3.10).

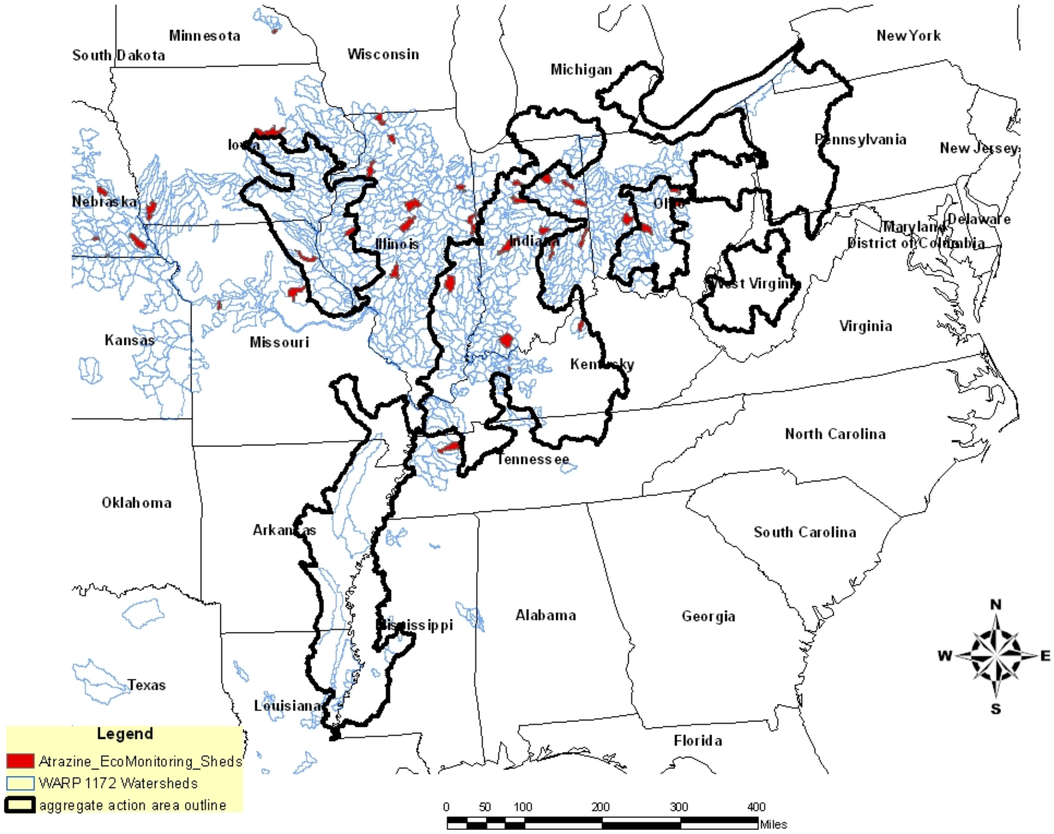

Figure 3.10 AEMP Sites Relative to Action Area

The following analysis represents a preliminary evaluation of the raw data and does not include the statistical analysis required to describe how the conditions in individual watersheds represent the larger population of 1,172 vulnerable watersheds. That analysis is not currently available and will be subject of a Scientific Advisory Panel (SAP) meeting in November 2007. In order to complete this preliminary analysis, each site/year of data was analyzed separately. Each data set was expanded to a 365-day time series and data interpolation was conducted. Preliminary data interpolation used a linear step method where the three un-sampled days after each sampled day were considered to have the same analytical result as the sampled day. Samples prior to the first sample date were considered to have the same result as the first sample date from that year, and a similar approach was taken for the un-sampled dates after the last sampling event. In addition, sample results from each date that were reported as non-detects were conservatively assigned an assumed value of the detection limit. Finally, dates where no sample was collected or analyzed were assumed to be equal to the nearest previous sample with a result. This final assumption results in uncertainty for a selected number of sites, particularly in Nebraska, where dry conditions resulted in fewer samples being collected.

Once the time series profile was created, a distribution of 14-day, 30-day, 60-day, and 90-day rolling average concentrations were calculated across the 365-day time series. In addition, an annual average concentration was calculated for comparison with screening-level EECs derived by PRZM modeling. Overall, a total of 84 individual site years of data have been collected from the 40 watersheds. Two of the watersheds (NE 04 in 2005 and NE 07 in 2005) represent years when multiple samples were not collected reportedly due to low flow conditions (NE 04 in 2005 with 15 missed samples and NE 07 in 2005 with 8 missed samples). Therefore, the rolling averages for these sites are questionable given the large amount of interpolation needed to infill data gaps. For all 40 watersheds, the exposures cover a range of concentrations for each duration with peak concentrations of 0.13 μg/L to 208.76 μg/L, 14-day concentrations ranging from 0.11 μg/L to 79.98 μg/L, 30-day concentrations from 0.10 μg/L to 45.17 μg/L, 60-day concentrations ranging from 0.1 μg/L to 25.74 μg/L, and 90-day concentrations ranging from 0.10 μg/L to 17.85 μg/L.

3.2.6.1.3 Comparison of AEMP Results with Modeling

Comparison of the calculated duration-magnitude concentrations from the monitoring data with flow-adjusted modeled EECs indicates that 5 of the 40 watersheds (13%) exceed the highest peak flow-adjusted EECs, 11 (28%) watersheds exceed the highest 14-day flow-adjusted EECs, 12 (30%) watersheds exceed the highest 30-day flow-adjusted EECs, 17 (43%) watersheds exceed the highest 60-day flow-adjusted EECs, and 17 (43%) watersheds exceed the highest 90-day flow-adjusted EECs. However, the magnitude of under-prediction by the flow-adjusted EEC is brought into context when considering that of these, only 2 watersheds are higher than two times the peak flow adjusted concentration, only 5 watersheds are greater than two times above the 14-day and 30-day average concentrations, and only 7 watersheds are greater than two times above the 60-day and 90-day average concentrations. In general, flow rates for the monitored sites yielding exposures higher than the flow adjusted modeling are low flow streams suggesting that flow is an important consideration, particularly when considering longer-term durations of exposure.

Although the ecological monitoring data set was targeted specifically to high atrazine use areas, only about half the watersheds are within the aggregated action area. It is difficult without specific site information to determine if species' locations are within these vulnerable watersheds; however, it does appear, from the limited information, that some potentially occupied fat pocketbook, northern riffleshell, and PCPP locations may be within the 1,172 watersheds. No critical habitat has been designated for the three listed mussels; therefore, no analysis of co-occurrence of critical habitat with the 1,172 watershed has been completed.

The 40 watersheds sampled in this study were selected using a statistical design intended to allow for extrapolation of monitoring results to the entire 1,172 watersheds including those present in the action area. However, the analysis to allow for such extrapolation is not currently available; therefore, it is not possible to determine the representative nature of these locations to the original 1,172 vulnerable watersheds, including those specific locations where listed mussels may occur.

An additional analysis of flow in occupied streams relative to flow in the monitored watersheds was completed. Based on this analysis, which is described in further detail in Section 3.2.7, the AEMP data represent a subset of occupied streams with flow below the 15^{th} percentile of flow from occupied streams (< 200 ft^3/sec). Therefore, these targeted monitoring data are used quantitatively to assess exposure and potential risk to populations of fat pocketbook and northern riffleshell mussels that may be found in low flow streams (i.e., < 200 ft^3/sec), as well as occupied streams for which no flow data is available, within the total 1,172 vulnerable watersheds. Until such time as the extrapolation from the subset of 40 sampled watersheds that may exceed thresholds of concern for atrazine to the entire 1,172 watershed has been completed, it cannot be precluded that some portion of the occupied habitat for the fat pocketbook and northern riffleshell is represented by results from the AEMP.

3.2.6.1.4 Conclusions Based on AEMP Results

Given the analysis above, it appears that some portion of occupied streams for each of the three listed species may be within the 1,172 watersheds. However, as further described in Section 3.2.7, the PCPP mussel occupies in streams with a higher flow rate than those represented by the available targeted monitoring data. Therefore, refined flow-adjusted EECs and non-targeted monitoring data are used to refine estimated exposure concentrations for the PCPP mussel. In addition, the range of the fat pocketbook and northern riffleshell outside the 1,172 vulnerable watershed boundary and in larger streams/rivers (with flow > 200 ft^3/sec) within the vulnerable watershed boundary (i.e., Mississippi, Columbia, and Ohio Rivers) and are best represented by flow-adjusted EECs and non-targeted monitoring data. The ancillary non-targeted monitoring data are described further in Sections 3.2.6.2 and 3.2.6.3.

3.2.6.2 USGS NAWQA Data

An analysis of the entire USGS NAWQA data set for atrazine was completed. A data download was conducted from the USGS data warehouse (http://water.usgs.gov/nawqa). Overall, a total of 20,812 samples were analyzed for atrazine. Of these, 16,742 samples had positive detections (including estimated values) yielding a frequency of detection of roughly 80%. The maximum detection from all samples was 201 µg/L from the Bogue Chitto Creek in Alabama near Memphis in 1999; however this sampling site is located outside of the action area boundary for this assessment. The maximum atrazine detection from samples collected within the action area for the three assessed mussels was 129 µg/L from Sugar Creek in New Palestine (near Indianapolis), Indiana in 1997. Overall, the average concentration detected was 0.26 µg/L when considering only detections and 0.21 µg/L when considering all detections and non-detections (using the detection limit as the value for estimation). The location of all NAWQA surface water sites relative to the action area and the targeted monitoring data is shown in Figure 3.11.

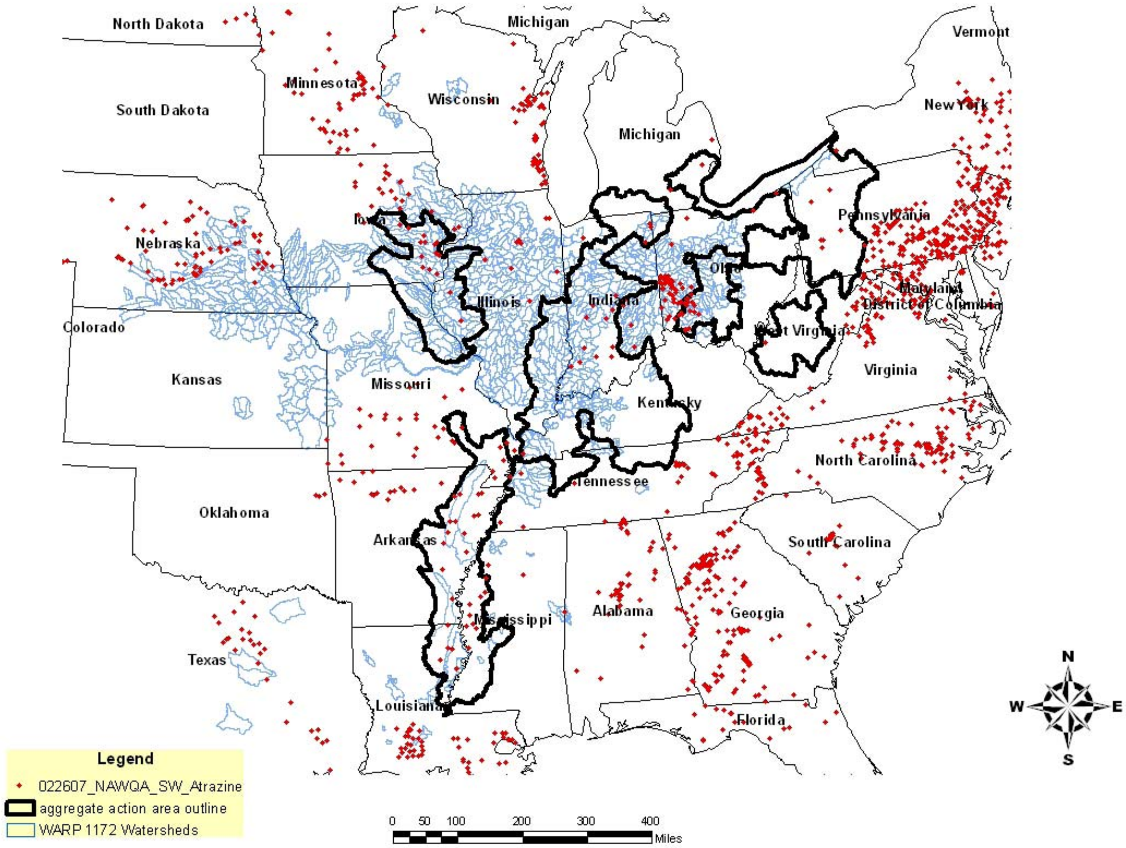

Figure 3.11 All USGS NAWQA Sites Relative to Action Area

The top sites with the highest atrazine concentrations from the national NAWQA data were selected for refined analysis of the detections. All of these sites are located within the 1,172 vulnerable watersheds, although some sites are located outside of the action area for this assessment. All values from the national data set were ranked and the top sites were selected based on maximum concentration. Each location was analyzed separately by year, and the annual maximum and annual time weighted mean (TWM) concentrations were calculated. The TWM estimates are intended to provide context to the screening-level and refined EECs by allowing for a comparison of the annual average concentrations from modeling with actual monitored data. The minimum criterion for calculating time-weighted means for each sampling station was at least 4 samples in a single year. The equation used for calculating the time weighted annual mean is as follows:

$$[((T_{0+1}-T_0) + ((T_{0+2}-T_{0+1})/2))*C\ t_{0+1})] + (((T_{i+1}-T_{i-1})/2)*C_i) + [((T_{end}-T_{end-1}) + ((T_{end-1}-T_{end-2})/2)*C_{Tend-1})]/365$$

where: Ci = Concentration of pesticide at sampling time (Ti)
Ti = Julian time of sample with concentration Ci

T_0 = Julian time at start of year = 0
T_{end} = Julian time at end of year = 365

Generally, the maximum (peak) concentrations from the USGS NAWQA data are consistent with peak concentrations observed from the targeted monitoring data, and roughly two times the values predicted using both the static water body and the flow adjusted approach. The TWM values from this analysis are roughly an order of magnitude below the static water body model predictions, two times above those estimated in the refined flow-adjusted EECs, and consistent with the targeted monitoring data. This analysis is somewhat biased because the selected USGS NAWQA data represent those sites with the highest concentrations and the majority of the sampling locations are within the same geographic extent as the targeted data – the 1,172 vulnerable watersheds. In reality, there are many more NAWQA sites within and outside the action area (Figure 3.11) with atrazine detections, and these sites would be expected to have lower concentrations (peak and annual average) than those reported for the top ten sites. Also of note is that there appears to be a general downward trend in atrazine exposures over time in these data (e.g. Bogue Chitto Creek), although some exceptions are noted (e.g. Sugar Creek, IL). Downward trends in exposure over time are expected given the label changes that have reduced application rates and implemented setbacks in the 1990's. Comparison of these data with modeled predictions for the intermediate duration exposures (14-day, 30-day, etc.) was not conducted because the NAWQA data generally do not have the frequency needed to conduct a meaningful interpolation between data points. Table 3.9 presents a summary of the annual time weighted mean concentrations, and Table 3.10 presents a summary of the annual maximum concentrations.

Table 3.9 Annualized Time Weighted Mean (TWM) Concentration (µg/L) for the Top Ten NAWQA Surface Water Sites (Ranked by Maximum Concentration Detected)										
	Station Name (ID)									
Year	Bogue Chitto Creek, near Memphis, TN (02444490)	Tributary to S Fork Dry Creek, near Schuyler, NE (06799750)	Sugar Creek, New Palestine, IN (394340085524601)	Kessinger Ditch, near Monroe City, IN (03360895)	LaMoine River @ Colmar, IL (05584500)	Sugar Creek @ Milford, IL (05525500)	Tensas River @ Tendal, LA (07369500)	Maple Creek near Nickerson, NE (06800000)	Auglaize River near Ft Jennings, OH (04186500)
1992			0.98					1.32	
1993			0.77	3.80				1.43	
1994			0.87	2.56					
1995			2.28	0.74					
1996			1.30				4.32		2.18
1997			5.36		3.45		5.55	1.03	2.82
1998			0.82		1.79		2.94	1.21	1.88
1999	9.62		0.28				2.50	0.68	
2000	6.49		0.56			1.26		0.15	
2001	1.20		0.83			0.78		0.22	1.28
2002	2.88		0.51			2.22		1.26	0.80
2003	2.14	4.46	0.70			7.83		2.23	1.42
2004	1.77	68.78[a]	0.67			1.24		3.31	1.93

[a] TWM concentration likely biased because the first sample on May 8 is the peak sample from this year.

Table 3.10 Maximum Concentration (µg/L) for the Top Ten NAWQA Surface Water Sites (Ranked by Maximum Concentration Detected)										
	Station Name (ID)									
Year	Bogue Chitto Creek, near Memphis, TN (02444490)	Tributary to S Fork Dry Creek, near Schuyler, NE (06799750)	Sugar Creek, New Palestine, IN (394340085524601)	Kessinger Ditch, near Monroe City, IN (03360895)	LaMoine River @ Colmar, IL (05584500)	Sugar Creek @ Milford, IL (05525500)	Tensas River @ Tendal, LA (07369500)	Maple Creek near Nickerson, NE (06800000)	Auglaize River near Ft Jennings, OH (04186500)
1992			14					25	
1993			8.5	120				11.2	
1994			11	24					
1995			27	2.6					
1996			14.2				30		18
1997			129		108		92.3	10.3	85.2
1998			7.88		27.7		19.3	30	9.96
1999	201		2.39				13.9	10.7	
2000	136		3.84			23		0.87	
2001	4.5		14.4			6.96		1.21	10.4
2002	24.8		4.01			21.3		16.4	2.58
2003	18.8	21.3	10.5			108		34.8	13.4
2004	14.6	191	28.3			10.9		91.9	18.7

3.2.6.3 Heidelberg College Data

Data from Heidelberg College, which consists of two intensively sampled watersheds (Maumee and Sandusky) in Ohio, were also analyzed. These sample sites are on the extreme northern edge of the action area and are also included in this analysis to provide context to the modeled exposures. It appears that the Sandusky watershed is within the boundary of the vulnerable watersheds included in the targeted monitoring study, while the Maumee watershed is outside this boundary. More information on the water quality monitoring program at Heidelberg College may be found at the following website:

http://wql-data.heidelberg.edu/

The Heidelberg data were collected more frequently than other data included in this assessment. The study design was specifically established to capture peak and longer-term trends in pesticide exposures. Data were collected between 1983 and 1999 and consist of an average of roughly 100 samples per year with several days of multiple sampling.

For the Sandusky watershed, a total of 1,597 samples were collected with 1,444 detections of atrazine (90.4% frequency of detection). The maximum concentration detected in the Sandusky watershed was 52.2 µg/L, and the overall average concentration was 4.5 µg/L. For the Maumee watershed, a total of 1,437 samples were collected with 1,305 detections of atrazine (90.8% frequency of detection). The maximum concentration detected in the Maumee watershed was 38.7 µg/L with an overall average concentration of 3.7 µg/L.

This analysis was further refined by deriving the annual TWM and maximum concentrations by sampled watershed by year. The results of this analysis are presented in Table 3.11. The results show a consistent pattern with that seen in other data collected from high atrazine use areas with general TWM concentrations between 1 and 3 µg/L. In addition, these data are generally two times lower than the peak refined flow-adjusted EECs and are generally consistent with the longer-term flow-adjusted average concentrations.

Table 3.11 Annual Time Weighted Mean and Maximum Concentrations (µg/L) for Atrazine in Two Ohio Watersheds from the Heidelberg College Data

Year	Sandusky Watershed		Maumee Watershed	
	TWM	Max	TWM	Max
1983	1.34	7.97	0.98	5.42
1984	1.08	8.73	1.27	11.71

Table 3.11 Annual Time Weighted Mean and Maximum Concentrations (µg/L) for Atrazine in Two Ohio Watersheds from the Heidelberg College Data				
Year	**Sandusky Watershed**		**Maumee Watershed**	
	TWM	**Max**	**TWM**	**Max**
1985	1.83	19.46	1.00	6.21
1986	3.32	24.61	1.64	10.01
1987	1.76	16.45	1.80	9.92
1988	0.41	1.53	0.43	2.15
1989	1.30	15.71	1.07	8.49
1990	1.96	19.31	1.69	14.78
1991	1.49	20.59	2.044	21.45
1992	0.39	40.53	0.51	7.35
1993	1.27	26.34	1.21	22.66
1994	0.86	10.10	0.82	4.02
1995	1.39	15.46	1.30	14.06
1996	1.56	23.40	1.19	16.19
1997[a]	**2.16**	**53.21**	2.09	38.74
1998	1.49	40.03	1.41	27.62
1999	1.57	17.11	1.88	19.37

[a] Sample year 1997 from Sandusky selected for data infilling by interpolation in order to calculate CASM duration exposure values.

Unlike the NAWQA data set, this data set had a sampling frequency adequate to interpolate between data points to estimate 14-day, 30-day, 60-day, and 90-day average concentrations. A final analysis of the data was completed by selecting one year's worth of data from the Heidelberg data. The 1997 sampling year was selected because it was one of the more recent data sets and because the maximum and TWM concentrations were higher than most other year's data. To process these data, it was necessary to "fill in the gaps". A total of 126 samples were collected during 1997 with 50 days with multiple samples yielding a time series of roughly 75 days. A step-wise approach was used to estimate daily concentrations between sampling dates that consisted of simply extending an analytical result from the date of analysis to the next date. For example, on January 6, 1997, atrazine was detected at a concentration of 0.475 µg/L. On the next sample date of January 20, 1997, no atrazine was detected (0 µg/L). In the step-wise interpolation, all dates between January 6 and January 20 were assigned the concentration of 0.475 µg/L. Also, because January 6 was the first sample date of the year, all previous days were also assigned a value of 0.475 µg/L. This process was repeated throughout the year to fill in the time series and yield 365 days worth of data. In addition, where multiple samples were analyzed on any given day, the highest of the values on that day was assigned. There is uncertainty with this type of interpolation because there is no information to suggest whether the interpolated value represents actual exposure. For example, where a gap in time exists between two samples, it is unlikely that a continuous concentration exists. It is more likely that there are upward and downward fluctuations in exposure, with a greater likelihood that higher exposures are missed between sample times with larger gaps in data points.

Table 3.12 presents the results of this analysis. The analysis suggests that, for the Sandusky watershed, in 1997, the estimated longer-term exposures are similar to those seen in the targeted data at roughly the 90th percentile of the distribution of 14-day, 30-day, 60-day, and 90-day rolling averages. Although the Sandusky watershed is located within the vulnerable watershed boundary defined by WARP, the rolling averages provided in Table 3.12 are used to characterize the potential upper bound of the refined flow-adjusted EECs for listed mussels that occur in less vulnerable watersheds and larger streams/rivers (> 200 ft^3/sec) within the boundary of vulnerable watersheds. These data are used to provide context to the flow-adjusted EECs because they were derived from non-targeted data with sufficient sampling frequency to derive 14 though 90 day rolling average exposure concentrations, and are considered as conservative estimates of exposure.

Table 3.12 Magnitude and Duration Estimates (µg/L) from the 1997 Data from Sandusky Watershed Using Stepwise Interpolation Between Samples

	14 day	21 day	30 day	60 day	90 day
Maximum	28.26	21.11	18.30	12.38	8.89

3.2.6.4 Summary of Open Literature Sources of Monitoring Data for Atrazine

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

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

The analysis in the IRED also documents the occurrence of atrazine in the atmosphere. The data indicate that atrazine can enter the atmosphere via volatilization and spray drift. The data also suggest that atrazine is frequently found in rain samples and tends to be seasonal, related to application timing. Finally, the data suggest that although frequently detected, atrazine concentrations detected in rain samples are less than those seen in the monitoring data and modeling conducted as part of this assessment and support the contention that runoff and spray drift are the principal routes of exposure. More details on these data can be found in the 2003 IRED (U.S. EPA, 2003a).

3.2.6.5 Miscellaneous Drinking Water Monitoring Data Derived from Surface Water

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

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

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

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

3.2.7 Comparison of Modeling and Monitoring Data

Modeling with the static water body provides screening-level EECs for use in risk estimation (Section 5.1). These screening-level EECs are refined and used in the risk description to characterize the relevance of predicted screening-level modeled exposures to the lower flow watersheds that are occupied by the three listed mussels. In this case, the listed species reside in 1st through 7th order streams with a wide range of flow rates. As previously discussed, lower to moderate flow rates (i.e. 22 ft^3/s to 110 ft^3/s) were assumed for refined modeling, based on a previous endangered species assessment for eight listed mussels (U.S. EPA, 2007c), because they are considered as representative of the low to moderate range of possible flow rates in streams where the fat pocketbook, PCPP mussel, and northern riffleshell occur. Flow rates from the low end of the range were selected because EECs derived from the refined modeling approach decrease with increasing flow; therefore, predicted EECs using flow rates from the low end of the range are expected to yield higher, more protective refined estimates of exposure. Additional characterization of the modeled static water body screening-level EECs used for risk estimation was completed to determine its relevance (and hence the RQs) to the species' habitat. In order to complete this characterization, additional refinement of the screening-level EECs is completed based on evaluation of modeled flow-adjusted EECs and available atrazine monitoring data.

Available monitoring data consists of both targeted and non-targeted data, as described above. Targeted monitoring data (i.e. AEMP; discussed in Section 3.2.6.1) is designed specifically to capture atrazine concentrations in watersheds with high atrazine use and exposure patterns in the most runoff prone settings; these data are representative of low order headwater streams (2nd and 3rd order generally) and are useful for direct comparison with effects data where the three listed mussels reside in similar low order, low flow, vulnerable streams. Non-targeted data (e.g. USGS NAWQA and Heidelberg College data; discussed in Sections 3.2.6.2 and 3.2.6.3) are typically designed to capture the general pattern of pollutants in the environment and are not designed specifically for any one chemical.

3.2.7.1 Relevance of AEMP Data to Listed Species Habitat

In this assessment, data from the AEMP provide a robust data set targeted to the most vulnerable watersheds in areas of atrazine use. These data are deemed directly comparable to the listed species that occur within the boundary of the most vulnerable watersheds (i.e. 1,172 WARP sites) where those species reside in similar stream types (many documented occurrences of the listed mussels are in higher order streams). It should be noted that because of the statistical nature of the study design, the results cannot be quantitatively comparable to less vulnerable watersheds outside the study design area. As noted above, although some portion of each species' range may overlap with the 1,172 watersheds, there are many potentially occupied locations outside the range of the 1,172 watersheds. For those occupied locations outside the 1,172 watersheds, exposures are best represented by the screening-level EEC with characterization from refined (flow-adjusted) modeling and the non-targeted monitoring data that occurs outside the range of the most vulnerable areas.

3.2.7.2 Direct Comparison of AEMP Data and Refined Model Estimates

In general, the targeted monitoring and refined flow-adjusted modeling provide a reasonable consistent picture of overall exposure. Of the 40 watersheds sampled, between 60% and 75% (depending on the duration of exposure) of the sampled sites are similar to, or less than the flow-adjusted model EEC. Of the targeted watersheds that exceed the refined flow-adjusted EECs, all but 10% to 15% of these exposures are within 2 times the refined modeling. Given that the targeted monitoring data represent the most vulnerable watersheds for the entire country and that the conditions modeled (low flow rates) are generally at or above those seen in the targeted monitoring data, it is not unexpected that there are a few excursions above the modeling. For example, 40% of sites from the upper 20th percentile of vulnerable watersheds that have higher atrazine concentrations than refined modeling sites represents approximately 8% of all atrazine watersheds nationally. In other words, 8% of all atrazine watersheds nationally are expected to be higher than the flow-adjusted EECs (assuming lower exposures in the lower vulnerability areas). If it is assumed that only 10% of sites are higher than 2 times the refined modeling (which is considered to be within the normal uncertainty of a model run), only 2% of all atrazine watersheds nationally would be expected to be higher than the flow-adjusted modeling. This suggests that, relative to the targeted monitoring, the refined flow-adjusted EECs, though exceeded occasionally, represent reasonably high end exposures for all watersheds nationally where atrazine is used.

3.2.7.3 Direct Comparison of Non-targeted Monitoring Data and Refined Model Estimates

A similar comparison of non-targeted monitoring data with refined flow-adjusted modeling yields similar conclusions. Non-targeted monitoring also provides a sense of how well the screening and refined modeling predict exposures in portions of the action area not directly represented by the targeted data. Comparison with modeling suggests that under certain conditions (low flow rates) concentrations can be higher in the non-targeted monitoring data than those predicted by the refined flow-adjusted modeling, however, it appears that most of these sites are located within the same watersheds as the targeted monitoring (i.e. WARP 1,172 highly vulnerable watersheds). However, much of the non-targeted monitoring data considered in this assessment are from the same general geographic area as the targeted data described above (Figure 3.11), although these non-targeted data have differing study objectives and are generally less robust.

In general, the trends in the non-targeted data are similar to those seen in the targeted data. Peak concentrations (though generally more than 10 years old) are twice as high as those predicted in screening and refined modeling. Given the less robust nature of these data, a direct comparison of various rolling averages with refined flow-adjusted rolling averages is not possible for the NAWQA data. However, rolling averages were considered for the Heidelberg data and like some of the targeted data is approximately 2 times higher than the refined modeling. For the NAWQA data, the annual mean concentrations can be compared and generally show the same pattern as the targeted data. The ranked percentiles (99th, 95th, 90th, 75th, 50th, 25th, 10th, and 5th) from the non-targeted data are comparable to those seen in the targeted data. This information is further summarized in Table D-7 of Appendix D.

3.2.7.4 Relationship Between Flow Rates from Monitored Sites and Flow Rates Used in Modeling

An important consideration when comparing the monitoring results to modeling is stream type and flow rate relative to each other. Several lines of evidence were evaluated to determine whether trends in the targeted and non-targeted data could be determined which would provide context to the overall exposure assessment. The range of flow rates in the targeted data was compared to the flow rates used in the refined modeling (i.e., those flows specific to streams and rivers where the listed mussels occur). In general, the species reside in watersheds with variable stream order (i.e., 1st through 7th order), while the targeted monitoring data are generally from 2nd and 3rd order streams. Flow rates used in refined modeling were between 20 ft^3/s and 110 ft^3/s, while flow rates for the targeted monitoring ranged from roughly < 10 ft^3/s to 180 ft^3/s. This suggests that the flow rates used in modeling were a reasonable approximation of flow in the targeted monitoring study.

Data on flow rates from occupied streams was captured from the USGS National Water Information System (NWIS) on July 16, 2007 (http://waterdata.usgs.gov/nwis/sw/). The flow data for selected occupied watersheds is compared directly with flow rates used in the refined modeling and is presented in Table 3.13. The data represent those occupied stream and rivers where USGS maintained a stream gage over an extended period of time (typically more than 20 years worth of data). In addition, the site locations were selected to best represent the occupied locations using a geospatial analysis of occupied stream locations as described by USFWS information and a comparison of the descriptive location information with actual gage sites. In many cases, the selected flow information was derived from a gage located on the stream or river near where the species lives, but not necessarily on the exact reach where reported occurrences have been observed. The information indicates that the flow rates used in the refined modeling are generally lower than those found in many of the streams where the species reside. It is important to note that the flow information below does not include occupied reaches in major rivers such as the Mississippi and Ohio Rivers, where higher flow rates are expected. This accounts for the fact that the range of flow rates reported previously in Table 2.3 show a much broader range than the values reported in Table 3.13.

Table 3.13 Summary of Listed Mussel Flow Rates Relative to Refined Modeling Flow Rates

Site	Mean Seasonal Flow (ft^3/s)
South Region – Refined Modeling	105
North Region – Refined Modeling	22
West Region – Refined Modeling	90
East Region – Refined Modeling	110
Purple Cats Paw	
Walhonding River @ Nellie OH	474
Cumberland River @ Carthage TN	4728
Green River @ Campbellsville KY	770
Green River @ Greensburg KY	691
Killibuck Creek @ Killibuck OH	390
Green River @ Lock 6 KY	3380
Northern Riffleshell	
French Creek @ Meadeville PA	293
French Creek @ Union City PA	303
Allegheny River @ Warren PA	3193
Elk River @ Sutton WV	801
Fish Creek @ Arctic IN	75
Fish Creek @ Hamilton IN	28
Fat Pocketbook	
White River @ Fayetteville AR	443
St Francis River @ St Francis AR	1906

In order to provide context of the relevance of the AEMP data to occupied locations, a comparison of flow rates from the targeted monitoring data with occupied streams was completed. The analysis (Table 3.14) shows that there is little overlap between the flow rates from the targeted monitoring data and the occupied streams, with higher flow rates occurring in occupied streams. The maximum value from the targeted data (177 ft^3/s) represents the 11th percentile of the occupied streams. It should be noted that data for occupied watersheds with high (e.g., Mississippi River) and low flow (e.g., Gilliam Chute) were not captured and/or available; therefore, there is uncertainty associated with the comparison of stream flow data from occupied watersheds with the Ecological Monitoring Data. The analysis suggests that the AEMP data are representative of only those streams with low flow such as Fish Creek in Indiana, and not sites with higher flow rates such as the Green River in Kentucky.

Table 3.14 Comparison of Ranked Percentile of Flow Rates (ft^3/s) from Occupied Streams versus Ecological (Targeted) Steams Sites

Occupied Sites[a]		Ecological Steam Monitoring Sites	
Max Value	4728	Max Value	177
99th Percentile	4553	99th Percentile	177
95th Percentile	3852	95th Percentile	141
90th Percentile	3324	90th Percentile	105
75th Percentile	1630	75th Percentile	67
50th Percentile	583	50th Percentile	30
25th Percentile	325	25th Percentile	18
10th Percentile	140	10th Percentile	7
5th Percentile	59	5th Percentile	4

[a] Available flow rate information for occupied streams from Table 3.13.

Given the analysis above, it is clear that the AEMP data represent a subset of occupied streams. The analysis suggests that this subset is limited to those sites with flow below the 15th percentile of flow from occupied streams (or approximately < 200 ft^3/s). Where flow information is unavailable for smaller occupied streams and rivers within the boundary of 1,172 vulnerable watersheds, it is presumed that exposure may be represented by the targeted monitoring data. For larger rivers, targeted monitoring data is not considered as representative because the flow rates in larger rivers are typically much higher than those presented above for the targeted monitoring data. For occupied sites outside the range of the 1,172, or within the 1,172 but not well represented by the flowing conditions in the targeted monitoring data, it is expected that a combination of the refined modeling and non-targeted data should be used for estimating exposure to the species in those areas. For example, the PCPP mussel occupies a limited range of streams both within and outside the boundary of vulnerable watersheds; however, stream flow data, which is available for all occupied streams, suggests that this species occupies streams with a higher flow rate than those represented by the available targeted monitoring data. Therefore, refined flow-adjusted EECs and non-targeted monitoring data are used to refine estimated exposure concentrations for the PCPP mussel. In summary, the targeted monitoring data are used to refine exposures only for the fat pocketbook and northern riffleshell in occupied streams within the vulnerable watershed boundary that have flow rates < 200 ft^3/s or for which no flow rate information is available. Flow-adjusted modeling and non-targeted data are used to refine exposure the PCPP mussel, and for those populations of fat pocketbook and northern riffleshell mussels that occur outside of the vulnerable watershed boundary and in larger streams/rivers (with flow rates > 200 ft^3/s) within the boundary of vulnerable watersheds.

Additional characterization comparing atrazine detections from all NAWQA surface water sites with all detections from the Ecological Stream Monitoring data was completed. In this analysis, all samples, regardless of site location or year, were ranked for both data sets**.** Table 3.15 presents the results of this analysis. Direct comparison indicates that peak values are roughly equivalent for both data sets; however, the distribution across the entire spectrum of atrazine detections is dramatically different. As the percentile decreases, the Ecological Stream Monitoring data becomes increasingly higher in concentration relative to the NAWQA data, with a two-fold difference at the 99.9th%, an order of magnitude difference at the 50th%, and nearly two orders of magnitude difference at the 10th%. A simple comparison of the two distributions of Ecological Stream Monitoring and NAWQA data was conducted using the t-test (two samples assuming unequal variances) in Microsoft Excel for both raw data and log-normalized data. In both cases the p values were less than 0.05 indicating that the distributions are significantly different. This analysis confirms that there are significant differences between the Ecological Stream Monitoring data and the entire NAWQA data set, which are likely due to differences in the sampling design (i.e., the Ecological Monitoring data are focused on the upper 20th% of vulnerable watersheds while the NAWQA data cover the entire range of atrazine use areas).

Table 3.15 Comparison of all NAWQA Atrazine Surface Water Data with the Ecological Stream Monitoring Data

Percentile	All NAWQA Surface Water Data (µg/L)	Ecological Stream Monitoring Data (µg/L)	Difference (µg/L)	Percent Difference
Max Value	201.00	237.50	36.50	18%
99.9th Percentile	61.25	137.21	75.96	124%
99.5th Percentile	20.09	59.51	39.41	196%
99th Percentile	11.70	33.37	21.67	185%
95th Percentile	1.96	10.70	8.74	446%
90th Percentile	0.63	4.97	4.34	685%

Table 3.15 Comparison of all NAWQA Atrazine Surface Water Data with the Ecological Stream Monitoring Data				
Percentile	**All NAWQA Surface Water Data (µg/L)**	**Ecological Stream Monitoring Data (µg/L)**	**Difference (µg/L)**	**Percent Difference**
75th Percentile	0.13	1.12	0.99	762%
50th Percentile	0.02	0.32	0.30	1233%
25th Percentile	0.01	0.11	0.10	1471%
10th Percentile	0.00	0.10	0.10	9900%
5th Percentile	0.00	0.10	0.10	9900%

3.2.8 Impact of Typical Usage Information on Exposure Estimates

A final piece of the exposure characterization includes an evaluation of usage information. Label application information was provided by EPA's Biological and Economic Analysis Division and summarized in Table 2.2. This information suggests that atrazine use on corn and sorghum (non-agricultural usage data is not available as part of this analysis) is typically 1.2 lbs/acre and 1.3 lbs/acre in the states considered within the action area of this assessment. This suggests that if typical application rates were used, atrazine exposures would be reduced below those modeled with the label maximum application rate by 40% for corn and 35% for sorghum. Typically usage information is not incorporated into these assessments, but does provide context to the exposures predicted. Caution is used when evaluating "typical" application rate information because this represents the average of all reported applications and thus roughly 50% of the time higher application rates are being applied.

3.3 Terrestrial Plant Exposure Assessment

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

Concentrations of atrazine on the riparian vegetation were estimated using OPP's TerrPlant model (U.S. EPA, 2007d; Version 1.2.2), considering use conditions likely to occur in the watersheds associated with the listed mussel's action area. The TerrPlant model evaluates exposure to plants via runoff and spray drift and is EFED's standard tool for estimating exposure to non-target plants. The runoff loading of TerrPlant is estimated based on the solubility of the chemical and assumptions about the drainage and receiving areas. As previously discussed in Section 3.2.3 (model inputs), the standard spray drift assumptions were modified using AgDrift to estimate the impact of a setback distance of 66 feet on the fraction of drift reaching a surface water body. These revised spray drift percentages were also incorporated into the TerrPlant model, assuming that non-target terrestrial plants adjacent to atrazine use sites would receive the same percentage of spray drift as an adjacent surface water body. The revised spray drift percentages are 0.6% for ground applications and 6.5% for aerial applications.

Although TerrPlant calculates exposure values for terrestrial plants inhabiting two environments (i.e., dry adjacent areas and semi-aquatic areas), only the exposure values from the dry adjacent areas are used in this assessment. The 'dry, adjacent area' is considered to be representative of a slightly sloped area that receives relatively high runoff and spray drift levels from upgradient treated fields. In this assessment, the 'dry, adjacent area' scenario is used to estimate screening-level exposure values for terrestrial plants in riparian areas. The 'semi-aquatic area' is considered to be representative of depressed areas that are ephemerally flooded, such as marshes, and, therefore, is not used to estimate exposure values for terrestrial riparian vegetation.

The following input values were used to estimate terrestrial plant exposure to atrazine from all uses: solubility = 33 ppm; minimum incorporation depth = 1 (TerrPlant default for incorporation depths ≤ 1 inch; from product labels); application methods: ground boom, aerial, and granular (from product labels). The following agricultural and non-agricultural scenarios were modeled: ground/aerial application to fallow/idle land at 2.25 lbs ai/A, corn/sorghum at 2.0 lb ai/A, and forestry at 4.0 lbs ai/A, and granular application to residential lawns at 2 lbs ai/A.

Terrestrial plant EECs for non-granular and granular formulations is summarized in **Table 3.16**. EECs resulting from spray drift are derived for non-granular applications only.

Table 3.16 Screening-Level Exposure Estimates for Terrestrial Plants to Atrazine

Use/ App. Rate (lbs/acre)	Application Method	Total Loading to Dry Adjacent Areas (lbs/acre)	Drift EEC (lbs/acre)
Forestry / 4.0	Aerial	0.34	0.26
	Ground	0.10	0.02
Fallow/idle land / 2.25	Aerial	0.19	0.15
	Ground	0.06	0.01
Corn and Sorghum / 2.0	Aerial	0.17	0.13
	Ground	0.05	0.01
Residential / 2.0	Granular	0.04	NA

For non-granular applications of atrazine, the highest off-target loadings of atrazine predicted by TerrPlant are approximately 8.5% of the application rate for dry adjacent areas. As expected, resulting exposure estimates for terrestrial plants are higher for aerial than ground boom applications. Granular applications associated with residential use of atrazine result in estimated exposures, as a percentage of the associated application rate, of 2% for adjacent areas.

4. Effects Assessment

This assessment evaluates the potential for atrazine to directly or indirectly affect the listed assessed mussels. As previously discussed in Section 2.7, assessment endpoints for the listed mussels include direct toxic effects on the survival, reproduction, and growth of the assessed mussels, as well as indirect effects, such as reduction of the prey base, perturbation of host fish, and/or modification of its habitat. Toxicity data used to evaluate direct and indirect effects are summarized in Table 4.1.

Table 4.1 Summary of Toxicity Data Used to Assess Direct and Indirect Effects

Toxicity Data	Assessment Endpoint	Comment
Acute and chronic studies in freshwater aquatic invertebrates	- Direct effects to listed mussels - Indirect effects to listed mussels via reduction in food supply	Preference given to tested species closest in taxonomy to assessed species and appropriate dietary items of assessed mussels
Acute and chronic studies in freshwater fish	- Indirect effects to listed mussel species via effects to host fish	Most sensitive studies used for assessment
Acute studies in vascular and non-vascular aquatic plants	- Indirect effects to listed mussels via reduction in food supply, habitat, and primary productivity	Most sensitive vascular and non-vascular aquatic plant studies initially used for screening-level RQ calculations; refinements include use of threshold concentrations to predict community-level effects
Terrestrial plant toxicity data	- Indirect effects to listed mussels via potential effects to habitat and water quality	Distribution of seedling emergence and vegetative vigor terrestrial plant data used in combination with toxicity data for woody vegetation, and riparian habitat characteristics

Acute (short-term) and chronic (long-term) effects toxicity information is characterized based on registrant-submitted studies and a comprehensive review of the open literature on atrazine, consistent with the Overview Document (U.S. EPA, 2004). In addition to registrant-submitted and open literature toxicity information, indirect effects to the listed mussels, via impacts to aquatic plant community structure and function are also evaluated based on community-level threshold concentrations. Other sources of information, including use of the acute probit dose response relationship to establish the probability of an individual effect and reviews of the Ecological Incident Information System (EIIS), are conducted to further refine the characterization of potential ecological effects associated with exposure to atrazine. A summary of the available freshwater and terrestrial plant ecotoxicity information, the community-level endpoints, use of the probit dose response relationship, and the incident information for atrazine are provided in Sections 4.1 through 4.4, respectively.

With respect to atrazine degradates, including hydroxyatrazine (HA), deethylatrazine (DEA), deisopropylatrazine (DIA), and diaminochloroatrazine (DACT), it is assumed that each of the degradates are less toxic than the parent compound. As shown in Table 4.2, comparison of available toxicity information for HA, DIA, and DACT indicates lesser aquatic toxicity than the parent for freshwater fish, invertebrates, and aquatic plants.

Table 4.2 Comparison of Acute Freshwater Toxicity Values for Atrazine and Degradates			
Substance Tested	**Fish LC_{50} (µg/L)**	**Daphnid EC_{50} (µg/L)**	**Aquatic Plant EC_{50} (µg/L)**
Atrazine	5,300	3,500	1
HA	>3,000 (no effects at saturation)	>4,100 (no effects at saturation)	>10,000
DACT	>100,000	>100,000	No data
DIA	17,000	126,000 (NOAEC: 10,000)	2,500
DEA	No data	No data	1,000

Although degradate toxicity data are not available for terrestrial plants, lesser or equivalent toxicity is assumed, given the available ecotoxicological information for other taxonomic groups including aquatic plants and the likelihood that the atrazine degradates are expected to lose efficacy as an herbicide.

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

As previously discussed in the problem formulation, the available toxicity data show that other pesticides may combine with atrazine to produce synergistic, additive, and/or antagonistic toxic interactions. The results of available toxicity data for mixtures of atrazine with other pesticides are presented in Section A.7 of Appendix A. Based on the results of the available data, study authors claim that synergistic effects with atrazine may occur for a number of organophosphate insecticides including diazinon, chlorpyrifos, and methyl parathion, as well as herbicides including alachlor. If chemicals that show synergistic effects with atrazine are present in the environment in combination with atrazine, the toxicity of the atrazine mixture may be increased relative to the toxicity of each individual chemical, offset by other environmental factors, or even reduced by the presence of antagonistic contaminants if they are also present in the mixture. The variety of chemical interactions presented in the available data set suggest that the toxic effect of atrazine, in combination with other pesticides used in the environment, can be a function of many factors including but not necessarily limited to (1) the exposed species, (2) the co-contaminants in the mixture, (3) the ratio of atrazine and co-contaminant concentrations, (4) differences in the pattern and duration of exposure among contaminants, and (5) the differential effects of other physical/chemical characteristics of the receiving waters (e.g. organic matter present in sediment and suspended water). Quantitatively predicting the combined effects of all these variables on mixture toxicity to any given taxa with confidence is beyond the capabilities of the available data. However, a qualitative discussion of implications of the available pesticide mixture effects data involving atrazine on the confidence of risk assessment conclusions for the freshwater mussels is addressed as part of the uncertainty analysis for this effects determination.

4.1 Evaluation of Aquatic Ecotoxicity Studies

Toxicity endpoints are established based on data generated from guideline studies submitted by the registrant, and from open literature studies that meet the criteria for inclusion into the ECOTOX database maintained by EPA/Office of Research and Development (ORD) (U.S. EPA, 2004). Open literature data presented in this assessment were obtained from the 2003 atrazine IRED as well as ECOTOX information obtained on May 31, 2007. The May 2007 ECOTOX search included all open literature data for atrazine (i.e., pre- and post-IRED). In order to be included in the ECOTOX database, papers must meet the following minimum criteria:

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

Meeting the minimum criteria for inclusion in ECOTOX does not necessarily mean that the data are suitable for use in risk estimation. Data that pass the ECOTOX screen are evaluated along with the registrant-submitted data, and may be incorporated qualitatively or quantitatively into this endangered species risk assessment. In general, effects data in the open literature that are more conservative than the registrant-submitted data are considered. Based on the results of the 2003 IRED for atrazine, potential adverse effects on sensitive aquatic plants and non-target aquatic organisms including their populations and communities, are likely to be greatest when atrazine concentrations in water equal or exceed approximately 10 to 20 µg/L on a recurrent basis or over a prolonged period of time (U.S. EPA, 2003a). Given the large amount of microcosm/mesocosm and field study data for atrazine, only effects data that are less than or more conservative than the 10 µg/L aquatic-community effect level identified in the 2003 atrazine IRED were 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 listed mussel survival, reproduction, and growth) identified in the problem formulation. For example, endpoints such as behavior modifications are likely to be qualitatively evaluated unless it is possible to quantitatively link these endpoints with reduction in species survival, reproduction, and/or growth (e.g., the magnitude of effect on the behavioral endpoint needed to result in effects on survival, growth, or reproduction is known).

Citations of all open literature not considered as part of this assessment because it was either rejected by the ECOTOX screen or accepted by ECOTOX but not used (e.g., the endpoint is less sensitive and/or not appropriate for use in this assessment) are included in Appendix G. Appendix G also includes a rationale for rejection of those studies that did not pass the ECOTOX screen and those that were not evaluated as part of this endangered species risk assessment.

As described in the Agency's Overview Document (U.S. EPA, 2004), the most sensitive endpoint for each taxa is evaluated. For this assessment, evaluated taxa include freshwater fish, freshwater aquatic invertebrates, freshwater aquatic plants, and terrestrial plants. Table 4.3 summarizes the most sensitive ecological toxicity endpoints for the three listed mussels, 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 three listed mussels is presented below. Additional information is provided in Appendix A. It should be noted that Appendix A also includes ecotoxicity data for taxonomic groups that are not relevant to this assessment (i.e., birds, estuarine/marine fish and invertebrates) because the Agency is completing endangered species risk assessments for other species concurrently with this assessment.

Table 4.3 Freshwater Aquatic and Terrestrial Plant Toxicity Profile for Atrazine

Assessment Endpoint	Species	Toxicity Value Used in Risk Assessment	Citation MRID # (Author & Date)	Comment
Acute Direct Toxicity to Listed Mussels	Freshwater mussel (*Anodonta imbecillis*)	24- and 48-hour LC_{50} = >36 mg/L Probit slope unavailable	ECOTOX #50679 (Johnson et al., 1993)	Open literature study
Chronic Direct Toxicity to Listed Mussels and Indirect Toxicity to Listed Mussels via Chronic Toxicity to Zooplankton (i.e., food items)	Scud	NOAEC = 60 μg/L LOAEC = 120 μg/L	000243-77 (Macek et al., 1976)	Acceptable: 25 % reduction in development of F_1 to seventh instar at the LOAEC
Indirect Effect to Mussel Glochidia via Direct Acute Toxicity to Host Fish	Rainbow trout	96-hour LC_{50} = 5,300 μg/L Probit slope = 2.72	000247-16 (Beliles and Scott, 1965)	Acceptable
Indirect Effect to Mussel Glochidia via Direct Chronic Toxicity to Host Fish	Brook trout	NOAEC = 65 μg/L LOAEC = 120 μg/L	000243-77 (Macek et al., 1976)	Acceptable full life-cycle study: 7.2% reduction in length; 16% reduction in weight occurred at the LOAEC
Indirect Effect to Listed Mussels via Acute Toxicity to Zooplankton (i.e., food items)	Midge	48-hour LC_{50} = 720 μg/L	000243-77 (Macek et al.,	Supplemental: raw data unavailable

Table 4.3 Freshwater Aquatic and Terrestrial Plant Toxicity Profile for Atrazine				
Assessment Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	**Citation MRID # (Author & Date)**	**Comment**
		Probit slope unavailable	1976)	
Indirect Effect to Listed Mussels via Acute Toxicity to Non-vascular Aquatic Plants	4 species of freshwater algae	1-week EC_{50} = 1 μg/L	000235-44 (Torres & O'Flaherty, 1976)	Supplemental study
Indirect Effect to Listed Mussels via Acute Toxicity to Vascular Aquatic Plants	Duckweed	14-day EC_{50} = 37 μg/L	430748-04 (Hoberg, 1993)	Supplemental study: NOAEC not determined
Indirect Effect to Listed Mussels via Acute Toxicity to Terrestrial Monocot Plants	Oat (monocot)	Tier II Seedling Emergence EC_{25} = 0.004 lb ai/A	420414-03 (Chetram, 1989)	Acceptable: EC_{50} based on reduction in dry weight
Indirect Effect to Listed Mussels via Acute Toxicity to Terrestrial Dicot Plants	Carrot (dicot)	Tier II Seedling Emergence EC_{25} = 0.003 lb ai/A	420414-03 (Chetram, 1989)	Acceptable: EC_{50} based on reduction in dry weight

Toxicity to aquatic fish and invertebrates is categorized using the system shown in Table 4.4 (U.S. EPA, 2004). Toxicity categories for aquatic plants have not been defined.

Table 4.4 Categories of Acute Toxicity for Aquatic Organisms	
LC/EC_{50} (mg/L)	**Toxicity Category**
< 0.1	Very highly toxic
> 0.1 - 1	Highly toxic
> 1 - 10	Moderately toxic
> 10 - 100	Slightly toxic
> 100	Practically nontoxic

4.1.1 Toxicity to Freshwater Mussels

Available freshwater mussel toxicity data were used to assess potential direct acute effects of atrazine to the assessed mussel species. A summary of acute and chronic freshwater mollusk and bivalve toxicity data is provided below in Sections 4.1.1.1 and 4.1.1.2. No freshwater mussel studies were submitted; therefore, all freshwater mussel studies were located in the open literature.

4.1.1.1 Freshwater Mussels: Acute Exposure Studies

The results of two acute toxicity tests using juvenile (*i.e.*, glochidial) and mature freshwater mussels suggest that two species of unionid mussels, *Anodonta imbecillis* and *Utterbackia imbecillis*, are less sensitive to atrazine on an acute exposure basis than other freshwater invertebrates commonly used in aquatic toxicity tests (e.g., cladocerans and amphipods) (Johnson et al., 1993; Conners and Black, 2004). The results of the freshwater mussel studies obtained from the open literature are summarized in Table A-

21 of Appendix A. Johnson *et al*. (1993) exposed juvenile mussels (20/concentration) to atrazine under static conditions at nominal concentrations up to 36 mg/L and evaluated survival of exposed individuals for 48 hours. Glochidia (1 to 2 days old and 7 to 10 days old) were exposed in a separate experiment for 24 hours under similar environmental conditions and exposure concentrations and evaluated for survival. The study reported LC_{50} values that were >60 mg/L for all life stages; therefore, it appears that the relative sensitivity of both the glochidial and mature mussel life stages to atrazine is similar. No acute toxicity was observed at any concentration tested. However, the methods did not report that 60 mg/L was tested either in a definitive or range-finding study. Therefore, the LC_{50} for this study is assumed to be >36 mg/L (corresponding NOAEC = 36 mg/L, the highest concentration reportedly tested). Using methods similar to the Johnson et al. (1993) study, Conners and Black (2004) report a 24-hr LC_{50} value of 214 mg/L for *U. imbecillis* glochidia for a formulated product (Atrazine 4L, 40.8% a.i.).

Guideline acute toxicity data for atrazine are also available for the Eastern oyster (*Crassostrea virginica*); however, this species inhabits estuarine/marine habitats. The results of Eastern oyster acute shell deposition studies report EC_{50} values ranging from >1,000 to >1,700 µg/L, with no effects reported at the highest atrazine test concentrations (MRIDs 466482-01 and 466482-01).

Given that the unionid mussel toxicity data from the open literature is more representative of the freshwater adult and juvenile mussel species being assessed as part of this effects determination than other tested species, and the available guideline data on estuarine/marine Eastern oysters shows no effects at the highest test concentrations of atrazine, the LC_{50} endpoint for *A. imbecillis* of >36 mg/L is used to calculate risk quotients for direct acute effects to the assessed mussels.

4.1.1.2 Freshwater Mussels: Chronic Exposure Studies

Chronic atrazine toxicity data for bivalves that are suitable for quantitative use in this risk assessment are not available from submitted studies or the open literature. However, several mollusk chronic exposure studies were located, with study durations ranging from approximately 6 to 12 weeks and endpoints including survival, fecundity, growth, and behavior. Baturo et al. (1995) did not observe any effects to marsh snail, *Lymnaea palustris*, in a 12-week mesocosm study at concentrations up to 125 µg/L (the highest concentration tested). Streit and Peter (1978) evaluated effects to the river limpet and to leeches from a 40-day exposure duration at atrazine concentrations of 1,000 to 16,000 µg/L. Effects at the LOAEC of 1,000 µg/L included increased mortality (although statistical significance was not indicated), increased ingestion, and reduced egg development. Although these studies were not considered appropriate for use in RQ calculations due to limitations in the study design and the lack of definitive NOAEC values (see Table A-21b of Appendix A), collectively, they suggest that effects to freshwater mollusks may occur at chronic exposure concentrations between 125 µg/L (NOAEC from Baturo et al., 1995) and 1,000 µg/L (LOAEC from Streit and Peter, 1978).

In the absence of appropriate chronic toxicity data for freshwater animals of similar taxa as mussels, the most sensitive endpoint across all freshwater aquatic invertebrate data was used to derive risk quotients. Uncertainties in using the most sensitive value across all species tested are discussed in Section 5.2. The most sensitive chronic endpoint for freshwater invertebrates is based on a 30-day flow-through study on the scud (*Gammarus fasciatus*), which showed a 25% reduction in the development of F_1 to the seventh instar at atrazine concentrations of 140 µg/L; the corresponding NOAEC is 60 µg/L (MRID # 000243-77).

4.1.2 Toxicity to Freshwater Fish

Freshwater fish toxicity data were used to assess potential indirect effects to the assessed mussels because the presence of suitable host fish is considered an essential elemental in the glochidial stage of the mussel's life cycle. Specific host fish species for the assessed mussels include freshwater drum, rock bass, brown trout, and various species of darters and sculpins (Table 2.3). Given the variability in host fish, the most sensitive acute and chronic freshwater fish data are used in the effects determination. A summary of acute and chronic freshwater fish atrazine toxicity data, in addition to data from the open literature on sublethal effects, is provided below in Sections 4.1.2.1 through 4.1.2.3.

4.1.2.1 Freshwater Fish: Acute Exposure (Mortality) Studies

Freshwater fish acute toxicity studies were used to assess potential indirect effects to the glochidial stage of the assessed mussels because all assessed mussels occur within freshwater rivers and/or streams and all identified fish hosts for the assessed mussels are presumably freshwater species (see Table 2.3). Atrazine toxicity has been evaluated in numerous freshwater fish species, including rainbow trout, brook trout, bluegill sunfish, fathead minnow, tilapia, zebrafish, goldfish, and carp, and the results of these studies demonstrate a wide range of sensitivity. The range of acute freshwater fish LC_{50} values for atrazine spans one order of magnitude, from 5,300 to 60,000 µg/L; therefore, atrazine is categorized as moderately (>1,000 to 10,000 µg/L) to slightly (>10,000 to 100,000 µg/L) toxic to freshwater fish on an acute basis. The freshwater fish acute LC_{50} value of 5,300 µg/L is based on a static 96-hour toxicity test using rainbow trout (*Oncorhynchus mykiss*) (MRID # 000247-16). No sublethal effects were reported as part of this study. A complete list of all the acute freshwater fish toxicity data for atrazine is provided in Table A-8 of Appendix A.

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

Chronic freshwater fish toxicity studies were used to assess potential indirect effects to mussel glochidia via growth and reproduction to mussel's host fish. Freshwater fish life-cycle studies for atrazine are available and summarized in Table A-12 of Appendix A. Following 44 weeks of exposure to atrazine in a flow-through system, statistically significant reductions in brook trout mean length (7.2%) and body weight (16%) were observed at a concentration of 120 µg/L, as compared to the control (MRID # 000243-77). The corresponding NOAEC for this study is 65 µg/L. Although the acute toxicity data for atrazine show that rainbow trout are the most sensitive freshwater fish, available chronic rainbow trout toxicity data indicate that it is less sensitive to atrazine, on a chronic exposure basis than the brook trout with respective LOAEC and NOAEC values of 1,100 µg/L and 410 µg/L. Further information on chronic freshwater fish toxicity data for atrazine is provided in Section A.2.2 of Appendix A.

4.1.2.3 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. Although these studies report potentially sensitive endpoints, effects on survival, growth, or reproduction were not observed in the four available life-cycle studies at concentrations that induced the reported sublethal effects described below and in Appendix A.

Reported sublethal effects in adult largemouth bass show increased plasma vitellogenin levels in both female and male fish at 50 µg/L and decreased plasma testosterone levels in male fish at atrazine concentrations greater than 35 µg/L (Wieser and Gross, 2002 [MRID 456223-04]). Vitellogenin (Vtg) is an egg yolk precursor protein expressed normally in female fish and dormant in male fish. The presence of Vtg in male fish is used as a molecular marker of exposure to estrogenic chemicals. It should be noted, however, that there is a high degree of variability with the Vtg effects in these studies, which confounds the ability to resolve the effects of atrazine on plasma steroids and vitellogenesis.

Effects of atrazine on freshwater fish behavior, including a preference for the dark part of the aquarium following one week of exposure (Steinberg et al., 1995 [MRID 452049-10]) and a reduction in grouping behavior following 24-hours of exposure (Saglio and Trijase, 1998 [MRID 452029-14]), have been observed at atrazine concentrations of 5 µg/L. In addition, alterations in rainbow trout kidney histology have also been observed at atrazine concentrations of 5 µg/L and higher (Fischer-Scherl et al., 1991 [MRID 452029-07]).

In salmon, atrazine effects on gill physiology and endocrine-mediated olfactory functions have been studied. Data from Waring and Moore (2004; ECOTOX #72625) suggest that salmon smolt gill physiology, represented by changes in Na-K-ATPase activity and increased sodium and potassium levels, was altered at 1 µg/L atrazine and higher. It should be noted, however, that a non-recommended solvent (methylated industrial spirits) was used in this study. Also, since the assessed mussels are freshwater species, seawater survival is not a relevant endpoint for potential host fish. Moore and Lower (2001; ECOTOX #67727) reported that endocrine-mediated functions of male salmon parr were affected at 1 µg/L atrazine. The reproductive priming effect of the female pheromone prostaglandin $F_{2\alpha}$ on the levels of expressible milt in males was reduced after exposure to atrazine at 1 µg/L. Although the hypothesis was not tested, the study authors suggest that exposure of smolts to atrazine during the freshwater stage may potentially affect olfactory imprinting to the natal river and subsequent homing of adults. However, no quantitative relationship is established between reduced olfactory response of male epithelial tissue 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). A negative control was not included as part of the study design; therefore, potential solvent effect cannot be evaluated. Furthermore, the study did not determine whether the decreased response of olfactory epithelium to specific chemical stimuli would likely impair similar responses in intact fish.

Tierney et al. (2007) studied the effect of 30 minute exposure to atrazine on behavioral and neurophysiological responses of juvenile rainbow trout to an amino acid odorant (L-histidine at 10^{-7} M). L-histidine was chosen because it has been shown to elicit an avoidance response in salmonids; however, control fish exposed to L-histidine at 10^{-7} M showed a slight preference (1.2 response ratio). Although the study authors conclude that L-histidine preference behavior was altered by atrazine at exposures $\geq$ 1 ug/L, no statistically significant decreases in preference behavior were observed at 1 ug/L. Furthermore, no dose response relationship was observed in the behavioral response following pesticide exposure. At 1 and 100 ug/L, non-significant decreases in L-histidine preference were observed; however a statistically significant avoidance of L-histidine was observed at 10 ug/L, but not 100 ug/L. Hyperactivity (measured as the number of times fish crossed the centerline of the tank) was observed in trout exposed to 1 and 10 ug/L atrazine. In the study measuring neurophysiological responses following atrazine exposure, electro-olfactogram (EOG) response was significantly reduced (EOG measures changes in nasal epithelial voltage due to response of olfactory sensory neurons). Although this study produced a more sensitive effects endpoint for freshwater fish, the data were not used quantitatively in the risk assessment because of the following reasons: 1) A negative control was not used; therefore, potential solvent effects cannot be evaluated; 2) The study did not determine whether the decreased response of olfactory epithelium to specific chemical stimuli would likely impair similar responses in intact fish; and 3) A quantitative relationship between the magnitude of reduced olfactory response to an amino acid odorant in the laboratory and reduction in trout imprinting and homing, alarm response, and reproduction (i.e., the ability of trout to detect, respond to, and mate with ovulating females) in the wild is not established.

Although these studies raise questions about the effects of atrazine on plasma steroid levels, behavior modifications, gill physiology, neurophysiological responses, and endocrine-mediated functions in freshwater and anadromous fish, it is not possible to quantitatively link these sublethal effects to the selected assessment endpoints for the listed mussels (i.e., survival, growth, and reproduction of individuals). Also, effects on survival, growth, or reproduction were not observed in the four available life-cycle studies at concentrations that induced these reported sublethal effects. Therefore, potential sublethal effects on fish are evaluated qualitatively in Section 5.2 and not used as part of the quantitative risk characterization. Further detail on sublethal effects to fish is provided in Sections A.2.4a and A.2.4b of Appendix A.

4.1.3 Toxicity to Freshwater Invertebrates

Although the primary component of the listed mussel's diet is phytoplankton, they have also been observed to filter zooplankton. Direct effects to zooplankton resulting from exposure to atrazine could indirectly affect the listed mussels via reduction in available food. As previously discussed, freshwater mussels are capable of filter-feeding only smaller sized zooplankton (i.e., $\leq$ 250 μm); however, toxicity data on the relative sensitivity of various sizes of freshwater invertebrates to atrazine are not available. Therefore, toxicity data for the most sensitive freshwater invertebrate are used to assess potential indirect effects of atrazine to the listed mussels via reduction in available zooplankton as food.

A summary of acute and chronic freshwater invertebrate data is provided below in Sections 4.1.3.1 and 4.1.3.2, respectively. All available open literature data for freshwater aquatic invertebrates that may be consumed by the listed mussels are less sensitive than the submitted atrazine toxicity data.

4.1.3.1 Freshwater Invertebrates: Acute Exposure Studies

Atrazine is classified as highly toxic to slightly toxic to aquatic invertebrates. There are a wide range of EC_{50}/LC_{50} values for freshwater invertebrates ranging from 720 to >33,000 μg/L. The lowest freshwater LC_{50} value of 720 μg/L is based on an acute 48-hour static toxicity test for the midge, *Chironomus tentans* (MRID # 000243-77). Further evaluation of the available acute toxicity data for the midge shows high variability with the LC_{50} values, ranging from 720 to >33,000 μg/L. With the exception of the midge, reported acute toxicity values for the other five freshwater invertebrates tested (including the water flea, scud, stonefly, leech, and snail) are 3,500 μg/L and higher. Because the listed mussels are likely to consume smaller, pelagic invertebrates, such as the water flea, the lowest water flea LC_{50} value of 3,500 μg/L (MRID # 450874-13) is used to characterize and refine the potential acute toxicity of atrazine to zooplankton. Further evaluation of the available acute toxicity data for the water flea also shows high variability similar to other freshwater invertebrates with LC_{50} values ranging from 3,500 to >30,000 μg/L. All of the available acute toxicity data for freshwater invertebrates are provided in Section A.2.5 and Table A-18 of Appendix A. The LC_{50}/EC_{50} distribution for freshwater invertebrates is graphically represented in Figure 4.1. The columns represent the lowest reported value for each species, and the positive y error bar represents the maximum reported value. Values in parentheses represent the number of studies included in the analyses.

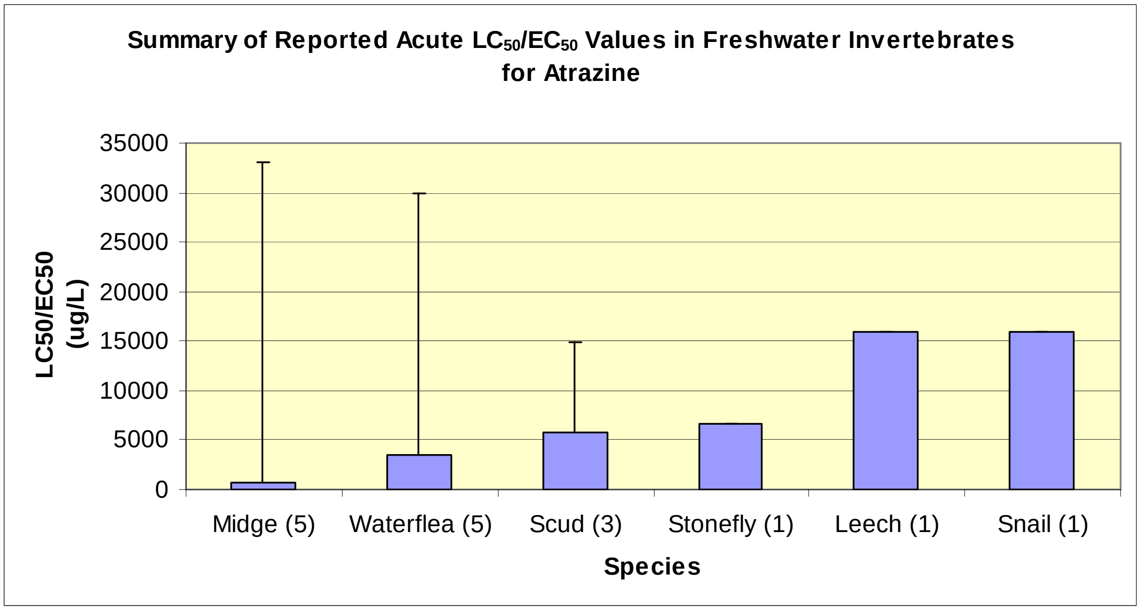

Figure 4.1 Summary of Reported Acute LC_{50}/EC_{50} Values in Freshwater Invertebrates for Atrazine

4.1.3.2 Freshwater Invertebrates: Chronic Exposure Studies

The most sensitive chronic endpoint for freshwater invertebrates is based on a 30-day flow-through study on the scud (*Gammarus fasciatus*), with respective NOAEC and LOAEC values of 60 and 140 µg/L, based on a 25% reduction in the development of F_1 to the seventh instar (MRID # 000243-77) (see Section 4.1.1.2). Although the acute toxicity data for atrazine show that the midge (*Chironomus tentans*) is the most sensitive freshwater invertebrate, available chronic midge toxicity data indicate that it is less sensitive to atrazine, on a chronic exposure basis, than the scud, with respective LOAEC and NOAEC values of 230 µg/L and 110 µg/L. The most sensitive chronic endpoint for zooplankton is based on a 21-day flow-through study on the water flea (*Daphnia magna*), which showed a 54% reduction in survival of F_0 young/female at atrazine concentrations of 250 µg/L; the corresponding NOAEC is 140 µg/L (MRID # 000243-77). Additional information on the chronic toxicity of atrazine to freshwater invertebrates is provided in Section A.2.6 and Table A-20 of Appendix A.

4.1.4 Toxicity to Aquatic Plants

Aquatic plant toxicity studies were used as one of the measures of effect to evaluate whether atrazine may affect primary production. In addition, aquatic plants including phytoplankton are a primary food source of both the juvenile and adult life stages of the listed freshwater mussels. In the watersheds within the action area for the mussels, primary productivity is essential for supporting the growth and abundance of the listed mussels.

Two types of studies were used to evaluate the potential of atrazine to affect primary productivity. Laboratory studies were used to determine whether atrazine may cause direct effects to aquatic plants. In addition, the community-level effect threshold concentrations, described in Section 4.2, were used to further characterize potential community-level effects to the listed mussel species resulting from potential effects to aquatic plants. A summary of the laboratory data for aquatic plants is provided in Section 4.1.4.1. A description of the threshold concentrations used to evaluate community-level effects is included in Section 4.2.

4.1.4.1 Aquatic Plants: Laboratory Data

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

The Tier II results for freshwater aquatic plants produced EC_{50} values for four different species of freshwater algae at concentrations as low as 1 μg/L, based on data from a 7-day acute study (MRID # 000235-44). Vascular plants are less sensitive to atrazine than freshwater non-vascular plants with an EC_{50} value of 37 μg/L, based on reduction in duckweed growth (MRID # 430748-04).

Comparison of atrazine toxicity levels for three different endpoints in algae suggests that the endpoints in decreasing order of sensitivity are cell count, growth rate and oxygen production (Stratton, 1984). Walsh (1983) exposed *Skeletonema costatum* to atrazine and concluded that atrazine is only slightly algicidal at relatively high concentrations (i.e., 500 and 1,000 μg/L). Caux et al. (1996) compared the cell count IC_{50} and fluorescence LC_{50} and concluded that atrazine is algicidal at concentrations affecting cell counts. Abou-Waly et al. (1991) measured growth rates on days 3, 5, and 7 for two algal species. The pattern of atrazine effects on growth rates differs sharply between the two species. Atrazine had a strong early effect on *Anabaena flos-aquae* followed by rapid recovery in clean water (i.e., EC_{50} values for days 3, 5, and 7 are 58, 469, and 766 μg/L, respectively). The EC_{50} values for *Selenastrum capricornutum* continued to decline from day 3 through 7 (i.e., 283, 218, and 214 μg/L, respectively). Based on these results, it appears that the timing of peak effects for atrazine may differ depending on the test species.

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

4.1.5 Freshwater Field Studies

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

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

As described in the 2003 IRED for atrazine (U.S. EPA, 2003a), potential adverse effects on sensitive aquatic plants and non-target aquatic organisms including their populations and communities are likely to be greatest when atrazine concentrations in water equal or exceed approximately 10 to 20 µg/L on a recurrent basis or over a prolonged period of time. A summary of all the freshwater aquatic microcosm, mesocosm, and field studies that were reviewed as part of the 2003 IRED is included in Section A.2.8a and Tables A-22 through A-24 of Appendix A. Given the large amount of microcosm and mesocosm and field study data for atrazine, only effects data less than or more conservative than the 10 µg/L aquatic community effect level identified in the 2003 IRED were considered from the open literature search that was completed in May 2007. Based on the selection criteria for review of new open literature, all of the available studies show effects levels to freshwater fish, invertebrates, and aquatic plants at concentrations greater than 10 µg/L.

It should be noted that the 10 to 20 µg/L community effect level has been further refined, since completion of the 2003 IRED. The community-level effects thresholds for various durations of exposure from 14 to 90 days are described in further detail in Section 4.2. In summary, the potential for atrazine to induce community-level effects depends on both atrazine concentration and duration. As the exposure duration increases, atrazine concentrations that may produce community level effects decrease. For example, 14-day atrazine concentrations of 38 µg/L or lower are not considered likely to result in aquatic community level effects, whereas 90-day atrazine concentrations of 12 µg/L or lower are not expected to produce community level effects.

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

4.1.6 Toxicity to Terrestrial Plants

Terrestrial plant toxicity data are used to evaluate the potential for atrazine to affect riparian zone vegetation within the action area for the listed mussels. Riparian zone effects may result in increased sedimentation, which may impact the assessed mussel species by reducing feeding and respiratory efficiency from clogged gills, disrupting metabolic processes, reducing growth rates, increasing substrata instability, limiting burrowing activity, and physical smothering (Ellis, 1936; Stansbery, 1971; Markings and Bills, 1979; Kat, 1982; Vannote and Minshall, 1982; Aldridge et al., 1987; and Waters, 1995). As previously discussed in Section 2.5 and Appendix C, the listed mussels require stable substrates for maintenance of viable mussel beds.

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

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

Based on the results of the submitted terrestrial plant toxicity tests, it appears that seedlings are more sensitive to atrazine via soil/root uptake exposure than emerged plants via foliar routes of exposure. However, all tested plants, with the exception of corn in the seedling emergence and vegetative vigor tests and ryegrass in the vegetative vigor test, exhibited adverse effects following exposure to atrazine. Tables 4.5 and 4.6 summarize the respective seedling emergence and vegetative vigor terrestrial plant toxicity data used to derive risk quotients in this assessment.

In Tier II seedling emergence toxicity tests, the most sensitive monocot and dicot species are oats and carrots, respectively. EC_{25} values for carrots and oats, which are based on a reduction in dry weight, are 0.003 and 0.004 lb ai/A, respectively; NOAEC values for both species are 0.0025 lb ai/A. Dry weight was the most sensitive parameter evaluated; emergence was not significantly affected at any level tested.

For Tier II vegetative vigor studies, the most sensitive dicot and monocot species are the cucumber and onion, respectively. In general, dicots appear to be more sensitive than monocots via foliar routes of exposure with all tested dicot species showing a significant reduction in dry weight at EC_{25} values ranging from 0.008 to 0.72 lb ai/A. In contrast, two of the four tested monocots showed no effect to atrazine (corn and ryegrass), while EC_{25} values for onion and oats were 0.61 and 2.4 lb ai/A, respectively.

Table 4.5 Non-target Terrestrial Plant Seedling Emergence Toxicity (Tier II) Data

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

Table 4.6 Non-target Terrestrial Plant Vegetative Vigor Toxicity (Tier II) Data

Surrogate Species	% ai	EC_{25} / NOAEC (lbs ai/A)	Endpoint Affected	MRID No. Author/Year	Study Classification
Monocot - Corn	97.7	> 4.0 / > 4.0	No effect	420414-03 Chetram 1989	Acceptable
Monocot - Oat	97.7	2.4 / 2.0	red. in dry weight	420414-03 Chetram 1989	Acceptable
Monocot - Onion	97.7	**0.61 / 0.5**	red. in dry weight	420414-03 Chetram 1989	Acceptable

Table 4.6 Non-target Terrestrial Plant Vegetative Vigor Toxicity (Tier II) Data					
Surrogate Species	**% ai**	**EC_{25} / NOAEC (lbs ai/A)**	**Endpoint Affected**	**MRID No. Author/Year**	**Study Classification**
Monocot - Ryegrass	97.7	> 4.0 / > 4.0	No effect	420414-03 Chetram 1989	Acceptable
Dicot - Carrot	97.7	1.7 / 2.0	red. in plant height	420414-03 Chetram 1989	Acceptable
Dicot - Soybean	97.7	0.026 / 0.02	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Lettuce	97.7	0.33 / 0.25	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Cabbage	97.7	0.014 / 0.005	red. in dry weight	420414-03 Chetram 1989	Acceptable
Dicot - Tomato	97.7	0.72 / 0.5	red. in plant height	420414-03 Chetram 1989	Acceptable
Dicot - Cucumber	97.7	**0.008 / 0.005**	red. in dry weight	420414-03 Chetram 1989	Acceptable

In addition, a report on the toxicity of atrazine to woody plants (Wall et al., 2006; MRID 46870400-01) was reviewed by the Agency. A total of 35 species were tested at application rates ranging from 1.5 to 4.0 lbs ai/A. Twenty-eight species exhibited either no or negligible phytotoxicity. Seven of 35 species exhibited >10% phytotoxicity. However, further examination of the data indicate that atrazine application was clearly associated with severe phytotoxicity in only one species (Shrubby Althea). These data suggest that, although sensitive woody plants exist, atrazine exposure to most woody plant species at application rates of 1.5 to 4.0 lbs ai/A is not expected to cause adverse effects. A summary of the available woody plant data is provided in Table A-39b of Appendix A.

4.2 Community-Level Endpoints: Threshold Concentrations

In this assessment, direct and indirect effects to the listed mussels are evaluated in accordance with the screening-level methodology described in the Agency's Overview Document (U.S. EPA, 2004). If aquatic plant RQs exceed the Agency's non-listed species LOC (because the assessed mussels do not have an obligate relationship with any one particular plant species, but rather rely on multiple plant species), based on available EC_{50} data for vascular and non-vascular plants, risks to individual aquatic plants are assumed.

It should be noted, however, that the indirect effects analyses in this assessment are unique, in that the best available information for atrazine-related effects on aquatic communities is more extensive than for other pesticides. Hence, atrazine effects determinations can utilize more refined data than is generally available to the Agency. Specifically, a robust set of microcosm and mesocosm data and aquatic ecosystem models are available for atrazine that allowed EPA to refine the indirect effects analysis associated with potential aquatic community-level effects (via aquatic plant community structural change and subsequent habitat modification) to the listed mussels. Use of such information is consistent with the guidance provided in the Overview Document (U.S. EPA, 2004), which specifies that "the assessment process may, on a case-by-case basis, incorporate additional methods, models, and lines of evidence that EPA finds technically appropriate for risk management objectives" (Section V, page 31 of EPA, 2004). This information, which represents the best scientific data available, is described in further detail below and in Appendix B of the previous atrazine endangered species effects determination for eight listed mussels (U.S. EPA, 2007c). This information is also considered a refinement of the 10-20 µg/L range reported in the 2003 IRED (U.S. EPA, 2003a).

The Agency has selected an atrazine level of concern (LOC) in the 2003 IRED (U.S. EPA, 2003a and b) that is consistent with the approach described in the Office of Water's (OW) draft atrazine aquatic life criteria (U.S. EPA, 2003c). Through these previous analyses (U.S. EPA, 2003a, b, and c), which reflect the current best available information, predicted or monitored aqueous atrazine concentrations can be interpreted to determine if a water body is likely to be affected via indirect effects to the aquatic community. Potential impacts of atrazine to plant community structure and function that are likely to result in indirect effects to the rest of the aquatic community, including the listed mussels, are evaluated as described below.

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

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

- 14-day average = 38 µg/L

- 30-day average = 27 μg/L
- 60-day average = 18 μg/L
- 90-day average = 12 μg/L

Effects of atrazine on aquatic plant communities that have the potential to subsequently pose indirect effects to the listed mussels are best addressed using the robust set of micro- and mesocosm studies available for atrazine and the associated risk estimation techniques (U.S. EPA, 2003a, b, c, and e). The 14-, 30-, 60-, and 90-day threshold concentrations developed by EPA (2003e) are used to evaluate potential indirect effects to aquatic communities for the purposes of this endangered species risk assessment. Use of these threshold concentrations is considered appropriate because: (1) the CASM-based index meets the goals of the defined assessment endpoints for this assessment; (2) the threshold concentrations provide a reasonable surrogate for the CASM index; and (3) the additional conservatism built into the threshold concentration, relative to the CASM-based index, is appropriate for an endangered species risk assessment (i.e., the threshold concentrations were set to be conservative, producing a low level (1%) of false negatives relative to false positives). Therefore, these threshold concentrations are used to identify potential indirect effects (via aquatic plant community structural change) to the listed mussels. If modeled atrazine EECs exceed the 14-, 30-, 60- and 90-day threshold concentrations following refinements of potential atrazine concentrations with available monitoring data, the CASM model could be employed to further characterize the potential for indirect effects. A step-wise data evaluation scheme incorporating the use of the threshold concentrations is provided in Figure 4.2. Further information on threshold concentrations is provided in Appendix B of the previous endangered species effects determination for eight listed mussels (U.S. EPA, 2007c).

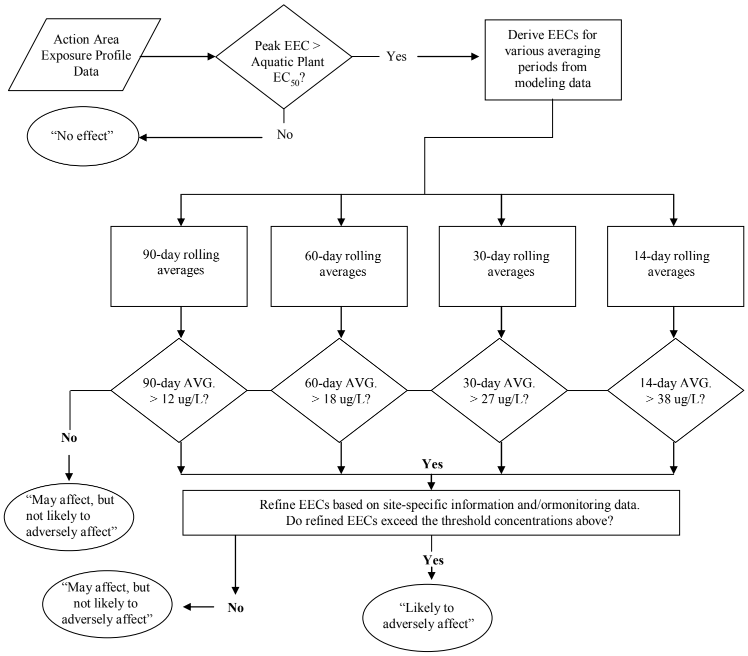

Figure 4.2 Use of Threshold Concentrations in Endangered Species Assessment

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

The Agency uses the probit dose response relationship as a tool for providing additional information on the potential for acute direct effects to individual listed species and aquatic animals that may indirectly affect the listed species of concern (U.S. EPA, 2004). As part of the risk characterization, an interpretation of acute RQ for listed species is discussed. This interpretation is presented in terms of the chance of an individual event (i.e., mortality or immobilization) should exposure at the EEC actually occur for a species with sensitivity to atrazine on par with the acute toxicity endpoint selected for RQ calculation. To accomplish this interpretation, the Agency uses the slope of the dose response relationship available from the toxicity study used to establish the acute toxicity measures of effect for each taxonomic group that is relevant to this assessment. The individual effects probability associated with the acute RQ is based on the mean estimate of the slope and an assumption of a probit dose response relationship. In addition to a single effects probability estimate based on the mean, upper and lower estimates of the effects probability are also provided to account for variance in the slope, if available. The upper and lower bounds of the effects probability are based on available information on the 95% confidence interval of the slope. A statement regarding the confidence in the estimated event probabilities is also included. Studies with good probit fit characteristics (i.e., statistically appropriate for the data set) are associated with a high degree of confidence. Conversely, a low degree of confidence is associated with data from studies that do not statistically support a probit dose response relationship. In addition, confidence in the data set may be reduced by high variance in the slope (i.e., large 95% confidence intervals), despite good probit fit characteristics. In the event that dose response information is not available to estimate a slope, a default slope assumption of 4.5 (lower and upper bounds of 2 to 9) (Urban and Cook, 1986) is used.

Individual effect probabilities are calculated using an Excel spreadsheet tool IECV1.1 (Individual Effect Chance Model Version 1.1) developed by the U.S. EPA, OPP, Environmental Fate and Effects Division (June 22, 2004). The model allows for such calculations by entering the mean slope estimate (and the 95% confidence bounds of that estimate) as the slope parameter for the spreadsheet. In addition, the acute RQ is entered as the desired threshold. Individual effect probabilities are discussed further as part of the Risk Description in Section 5.2.

4.4 Incident Database Review

A number of incidents have been reported in which atrazine has been associated with some type of environmental effect. Incidents are maintained and catalogued by EFED in the Ecological Incident Information System (EIIS). Each incident is assigned a level of certainty from 0 (unrelated) to 4 (highly probable) that atrazine was a causal factor in the incident. As of the writing of this assessment, 358 incidents are in EIIS for atrazine spanning the years 1970 to 2005. Most (309/358, 86%) of the incidents involved damage to terrestrial plants, and most of the terrestrial plant incidences involved damage to crops treated directly with atrazine. Of the remaining 49 incidents, 47 involved aquatic animals and 2 involved birds. Because the species included in this effects determination are aquatic species, incidents involving aquatic animals assigned a certainty index of 2 (possible) or higher (N=33) were re-evaluated. Results are summarized below, and additional details are provided in Appendix E. The 33 aquatic incidents were divided into three categories:

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

The presence of atrazine at levels thought to be sufficient to cause either direct or indirect effects was confirmed in 3 (9%) of the 33 aquatic incidents evaluated. Atrazine use was also correlated with 11 (33%) additional aquatic incidents where its presence in the affected water was not confirmed, but the timing of atrazine application was correlated with the incident. Therefore, a definitive causal relationship between atrazine use and the incident could not be established. The remaining 19 incidents (58%) were likely caused by some factor other than atrazine. Other causes primarily included the presence of other pesticides at levels known to be toxic to affected animals. Although atrazine use was likely associated with some of the reported incidents for aquatic animals, they are of limited utility to this assessment for the following reasons:

- No incidents in which atrazine is likely to have been a contributing factor have been reported after 1998. A number of label changes, including cancellation of certain uses, reduction in application rates, and harmonization across labels to require setbacks for applications near waterbodies, have occurred since that time. For example, several incidents occurred in ponds that are adjacent to treated fields. The current labels require a 66-foot buffer between application sites and water bodies.

- The habitat of the assessed species is not consistent with environments in which incidents have been reported. For example, no incidents in streams or rivers were reported.

Although the reported incidents suggest that high levels of atrazine may result in impacts to aquatic life in small ponds that are in close proximity to treated fields, the incidents are of limited utility to the current assessment. However, the lack of recently reported incidents in flowing waters does not indicate that effects have not occurred. Further information on the atrazine incidents and a summary of uncertainties associated with all reported incidents are provided in Appendix E.

5. Risk Characterization

Risk characterization is the integration of the exposure and effects characterizations to determine the potential ecological risk from varying atrazine use scenarios within the action area and likelihood of direct and indirect effects on the listed mussels. The risk characterization provides an estimation (Section 5.1) and a description (Section 5.2) of the likelihood of adverse effects; articulates risk assessment assumptions, limitations, and uncertainties; and synthesizes an overall conclusion regarding the likelihood of adverse effects to the listed mussels (i.e., “no effect,” “likely to adversely affect,” or “may affect, but not likely to adversely affect”). In accordance with the Agency’s Overview Document (U.S. EPA, 2004), RQs derived in the risk estimation are based on screening-level EECs using the PRZM-EXAMS static water body modeling. In the risk description, atrazine exposures are refined by considering the available targeted and non-targeted monitoring data and flow-adjusted EECs.

As previously discussed in the effects assessment (Section 4), the toxicity of the atrazine degradates has been shown to be less than the parent compound based on the available toxicity data for freshwater fish, invertebrates, and aquatic plants; therefore, the focus of the risk characterization is parent atrazine (i.e., RQ values were not derived for the degradates).

5.1 Risk Estimation

Risk was estimated by calculating the ratio of the screening-level estimated environmental concentration (EEC) (Table 3.7) and the appropriate toxicity endpoint (Table 4.3). 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 F). Screening-level RQs are based on the most sensitive endpoints and the following surface water concentration scenarios for atrazine:

- corn use @ 2.5 lbs ai/A; 2 applications
- sorghum use @ 2 lbs ai/A; 1 application
- fallow/idle land use @ 2.25 lb ai/A; 1 application
- forestry use @ 4.0 lb ai/A; 1 application
- residential granular use @ 2 lb ai/A; 2 applications with 30 days between applications
- residential liquid use @ 1 lb ai/A; 2 applications with 30 days between applications
- turf granular use @ 2 lb ai/A; 2 applications with 30 days between applications
- turf liquid use @ 1 lb ai/A; 2 applications with 30 days between applications
- rights-of-way liquid use @ 1 lb ai/A; 1 application

EECs are also derived for terrestrial plants, as discussed in Section 3.3, based on the highest application rates of atrazine use within the action area.

As previously discussed in Section 3.2, the action area for the listed mussels was divided into five regions representing the northern, eastern, southern, western, and upper Great Plains regions of the listed mussel's range. As shown in Table 3.2, all three of the assessed mussel species are known to occur in the northern region. In addition, the fat pocketbook also occurs in the southern, western, and upper Great Plains regions, and the northern riffleshell occurs in the eastern region. The highest screening-level EEC from the region where the species occurs was initially used to derive risk quotients. For the fat pocketbook, the highest screening-level EECs are based on atrazine uses in the southern region of the action area; for both the PCPP mussel and northern riffleshell, the highest screening-level EECs are based on atrazine use in the northern region of the action area. In cases where LOCs were not exceeded based on the highest EEC, additional RQs were not derived because it was assumed that RQs for lower EECs would also not exceed LOCs. However, if LOCs were exceeded based on the highest EEC, use/region-specific RQs were also derived.

In cases where the screening-level RQ exceeds one or more LOCs (i.e., "may affect"), additional factors, including the listed mussels life history characteristics, refinement of the screening-level EECs using site-specific information, available monitoring data, and consideration of community-level threshold concentrations are considered and used to characterize the potential for atrazine to adversely affect the listed mussels. Risk estimations of direct and indirect effects of atrazine to the three listed mussels are provided in Sections 5.1.1 and 5.1.2, respectively.

5.1.1 Direct Effects

Direct effects to the listed mussels associated with acute and chronic exposure to atrazine are based on the most sensitive toxicity data available for freshwater mussels and other surrogate aquatic invertebrates. Acute toxicity data specific for freshwater mussels are available; however, no chronic data for freshwater mussels exist. RQs used to estimate acute direct effects to the listed mussels are provided in Table 5.1 below. The peak screening-level EECs (109 and 101 µg/L) used to derive acute RQs for the assessed listed mussels are representative of the highest modeled EECs from corn use scenarios in the southern region (for the fat pocketbook) and the northern region (for the PCPP mussel and northern riffleshell). Based on the highest screening-level EECs modeled for atrazine use patterns within the five regions, acute RQs do not exceed the endangered species LOC of 0.05. Therefore, atrazine is not expected to result in acute direct effects to listed mussels within the action area. These RQs are further characterized in Section 5.2.1.1.

Table 5.1 Summary of Direct Effect Acute RQs for the Listed Mussels

Effect to Listed Mussels	Surrogate Species	Toxicity Value (µg/L)[a]	Peak EEC (µg/L)	RQ	Probability of Individual Effect[b]	LOC Exceedance and Risk Interpretation
Acute Direct Toxicity	Freshwater Mussel	LC_{50} = >36,000	South = 109[c]	<0.003	1 in 2.7E+29 (1 in 4.4E+06 to 1 in 5.1E+113)	No[d]
			North = 101[e]	<0.003	1 in 1.32E+30 (1 in 6.07E+06 to 1 in 2.44E+116)	No[d]

[a] Based on 48-hour LC_{50} value of >36,000 µg/L for freshwater mussels and glochidia (ECOTOX #50679).
[b] A probit slope value for the acute mussel toxicity test is not available; therefore the effect probability was calculated based on a default slope assumption of 4.5 with upper and lower 95% confidence intervals of 2 and 9 (Urban and Cook, 1986).
[c] Representative of the maximum screening-level acute exposure for the fat pocketbook mussel, based on peak southern corn screening-level EEC (Table 3.7).
[d] RQ < acute endangered species LOC of 0.05.
[e] Representative of the maximum screening-level acute exposure for the PCPP mussel and northern riffleshell, based on peak northern corn screening-level EEC (Table 3.7).

In the absence of chronic toxicity data to freshwater mussels, the most sensitive NOAEC value from the available freshwater invertebrate data was used as a surrogate to derive chronic risk quotients for freshwater mussels. RQs used to estimate chronic direct effects to the listed mussels are provided in Table 5.2. These RQs are also used to assess potential indirect effects to the listed mussels based on reduction in freshwater invertebrates (i.e., zooplankton) food items in Section 5.1.2.1. Chronic RQs exceed LOCs based on atrazine use on corn in all five regions of the action area, with RQs ranging from 1.37 to 1.78. In addition, chronic RQs based on atrazine use on sorghum in the south and east, fallow/idle land in the west, and forestry in the upper Great Plains regions of the action area also exceed LOCs. Chronic RQs based on non-agricultural atrazine use on residential, turf, and rights-of-way are less than LOCs; therefore, direct chronic effects to the listed mussels are not expected based on these use patterns. In summary, chronic RQs derived using screening-level EECs and the most sensitive aquatic invertebrate NOAEC exceeded LOCs for atrazine use on corn, sorghum, fallow/idle land, and forestry within the action area; however, atrazine use patterns related to residential, turf, and rights-of-way are less than the chronic LOC. Based on the distribution of the three assessed mussels within various regions of the action area, the screening-level effects determination for direct chronic effects is “may affect” for the fat pocketbook (for corn use in the south, north, west, and upper Great Plains; sorghum use in the south; fallow/idle land use in the west; and forestry use in the upper Great Plains), PCPP mussel (for corn use in the north), and northern riffleshell (for corn use in the north and sorghum use in the east). These RQs are further characterized in Section 5.2.1.1.

Table 5.2 Summary of Direct Effect Chronic RQs for the Listed Mussels

Effect to Listed Mussels	Use (appl. Method; rate; # appl.; interval between appl.)	Range of 21-day EECs (µg/L)[a]	Freshwater Invertebrate Chronic RQ (NOAEC= 60 µg/L[b])	LOC Exceedance (Species)
Chronic Direct Toxicity[c]	Corn (aerial liquid; 2.5 lb ai/A; 2 appl.)	82 - 107	**1.37 – 1.78**	**Yes[d] (all regions: FP, PCPP, and NR)**
	Sorghum (aerial liquid; 2 lb ai/A; 1 appl.)	South: 62 North: 57 West: 59 UGP: 56 East: 68	**South: 1.03** North: 0.95 West: 0.98 UGP: 0.93 **East: 1.13**	**South: Yes[d] (FP)** North: No[e] West: No[e] UGP: No[e] **East: Yes[d] (NR)**
	Fallow/Idle land (aerial liquid; 2.25 lb ai/A; 1 appl.)	South: 58 North: 52 West: 103 UGP: 49 East: 54	South: 0.97 North: 0.87 **West: 1.72** UGP: 0.82 East: 0.90	South: No[e] North: No[e] **West: Yes[d] (FP)** UGP: No[e] East: No[e]
	Forestry (aerial liquid; 4 lb ai/A; 1 appl.)	South: 45 North: 47 West: 27 UGP: 61 East: 43	South: 0.75 North: 0.78 West: 0.45 **UGP: 1.02** East: 0.72	South: No[e] North: No[e] West: No[e] **UGP: Yes[d] (FP)** East: No[e]
	All other non-agricultural uses	≤ 19	≤ 0.32	No[e]

[a] 21-day screening-level EECs include the range of modeled concentrations from all five regions of the action area (Table 3.7). 21-day screening-level EECs from each of the five regions are provided for the sorghum, fallow/idle land, and forestry scenarios in order to differentiate the specific region where chronic freshwater invertebrate RQs exceed LOCs. UGP = upper Great Plains.
[b] Based on 30-day NOAEC value of 60 µg/L for the scud (MRID # 000243-77).
[c] RQs associated with chronic direct toxicity to the listed mussels are also used to assess potential indirect effects to the listed mussels based on a reduction in freshwater invertebrates (i.e., zooplankton) as food items.
[d] RQ > chronic LOC of 1.0. Further evaluation of the RQs is necessary to determine if atrazine is likely to adversely affect the assessed species. FP = fat pocketbook; NR = northern riffleshell; PCPP = purple cat’s paw pearlymussel.
[e] RQ < chronic LOC of 1.0.

5.1.2 Indirect Effects

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

In conducting a screen for indirect effects, direct effects LOCs for each taxonomic group (i.e., freshwater fish, invertebrates, aquatic plants, and terrestrial plants) are employed to make inferences concerning the potential for indirect effects upon listed species that rely upon non-listed organisms in these taxonomic groups as resources critical to their life cycle (U.S. EPA, 2004). This approach used to evaluate indirect effects to listed species is endorsed by the Services (USFWS/NMFS, 2004b). If no direct effect listed species LOCs are exceeded for non-endangered organisms that are critical to the listed mussel's life cycle, indirect effects to the listed mussels are not expected to occur.

If LOCs are exceeded for freshwater invertebrates (i.e., zooplankton) or aquatic non-vascular plants (i.e., phytoplantkon) that are food items of the listed mussels, there is a potential for atrazine to indirectly affect the listed mussels by reducing available food supply. In addition, if LOCs are exceeded for freshwater fish that are host fish of the listed mussel glochidia, atrazine may indirectly affect the listed mussels by disrupting the parasitic glochidial life cycle stage of the mussel that is reliant on suitable host fish. In such cases, the dose response relationship from the toxicity study used for calculating the RQ of the surrogate prey item or host fish is analyzed to estimate the probability of acute effects associated with an exposure equivalent to the EEC. The greater the probability that exposures will produce effects on a taxa, the greater the concern for potential indirect effects for listed species dependant upon that taxa (U.S. EPA, 2004).

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

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

In summary, the potential for indirect effects to the listed mussels was evaluated using methods outlined in U.S. EPA (2004) and described below in Sections 5.1.2.1 through 5.1.2.4.

5.1.2.1 Evaluation of Potential Indirect Effects via Reduction in Food Items (Freshwater Zooplankton and Phytoplankton)

Freshwater mussels are filter-feeders, consuming primarily phytoplankton, but also detritus, zooplantkton, and other microorganisms (Ukeles, 1971; Coker et al., 1921; Churchill and Lewis, 1924; and Fuller, 1974). Data on the relative percentage of each type of food item in the mussel's diet are unavailable. Potential indirect effects from direct effects on plant and animal food items (i.e., phytoplankton and zooplankton) were evaluated by considering the diet of the assessed mussels and the effects data for the most sensitive food item in each taxonomic group (i.e., freshwater algae and midge). The acute RQs used to characterize potential indirect effects to the assessed mussels from direct acute effects on freshwater phytoplankton and zooplankton food sources are provided in Table 5.3. Acute RQs are presented for the atrazine use rates that correspond to agricultural and non-agricultural EECs across the five regions in order to provide a range of possible acute RQ values.

Indirect effects to the listed mussels based on direct acute effects to dietary items may occur for phytoplankton and zooplankton. As shown in Table 5.3, acute LOCs are exceeded for phytoplankton for all labeled uses of atrazine within the action area, with RQs ranging from 2.4 to 109. Acute RQs for zooplankton exceed LOCs for corn, sorghum, fallow/idle land, and forestry (in all geographic regions except the west) uses of atrazine, with values ranging from 0.06 to 0.15, based on the most sensitive freshwater invertebrate acute toxicity endpoint. However, acute RQs based on non-agricultural uses of atrazine on residential, turf, and rights-of-way, as well as forestry uses in the western region of the action area, are less than LOCs for aquatic invertebrates. Based on the distribution of the mussels within various regions of the action area, the screening-level effects determination for indirect effects via direct acute effects to dietary items is "may affect" for all three species. These risk quotients are further characterized in Section 5.2.1.2.

Table 5.3 Summary of Acute RQs Used to Estimate Indirect Effects to the Listed Mussels via Direct Effects on Dietary Food Items

Use (appl. method; rate; # appl.; interval between appl.)	Range of Peak EECs[a]	Direct Effects to Phytoplankton		Direct Effects to Zooplankton	
		Acute RQ (non-vascular plant EC_{50} = 1 µg/L)[b]	LOC Exceedance and Risk Interpretation	Acute RQ (midge LC_{50} = 720 µg/L)[c]	LOC Exceedance and Risk Interpretation
Corn (aerial liquid; 2.5 lb ai/A; 2 appl.)	83 – 109	**83 – 109**	**Yes[d]**	**0.12 – 0.15**	**Yes[d]**
Sorghum (aerial liquid; 2 lb ai/A; 1 appl.)	57 – 69	**57 – 64**	**Yes[d]**	**0.08 – 0.10**	**Yes[d]**

Fallow/Idle land (aerial liquid; 2.25 lb ai/A; 1 appl.)	49 – 103	**49 – 103**	**Yes[d]**	**0.07 – 0.14**	**Yes[d]**
Forestry (aerial liquid; 4 lb ai/A; 1 appl.)	South: 46 North: 49 West: 27 UGP: 65 East: 44	**South: 46** **North: 49** **West: 27** **UGP: 65** **East: 44**	**Yes[d]**	**South: 0.06** **North: 0.07** West: 0.04 **UGP: 0.09** **East: 0.06**	**South: Yes[d]** **North: Yes[d]** West: No[f] **UGP: Yes[d]** **East: Yes[d]**
All other non-agricultural uses	2.4 - 20	**2.4 - 20**	**Yes[d]**	$\leq$ 0.03	No[f]

[a] Peak screening-level EECs include the range of modeled concentrations from all five regions of the action area (Table 3.7). Peak screening-level EECs from each of the five regions are provided for the forestry scenario in order to differentiate the specific region where acute freshwater invertebrate RQs exceed LOCs. UGP = upper Great Plains.
[b] Based on 1-week EC_{50} value of 1 µg/L for four species of freshwater algae (MRID # 000235-44).
[c] Based on 48-hour LC_{50} value of 720 for the midge (MRID #000243-77). Slope information on the toxicity study that was used to derive the RQ for the midge is not available. Therefore, the probability of an individual effect was calculated using the probit slope of 4.4, which is the only technical grade atrazine value reported in the available freshwater invertebrate studies; 95% confidence intervals could not be calculated based on the available data (MRID # 452029-17; Table A-18).
[d] RQ > LOC (LOC = 1 for aquatic plants and 0.05 for invertebrates). Further evaluation of refined EECs (based on site-specific information and available monitoring data) relative to the threshold concentrations (for community-level effects) is necessary.
[e] Based on an assumed probit dose of 4.4, the range of individual effect probabilities for aquatic invertebrates at acute RQs that exceed LOCs is 1 in 6,930 (for RQ of 0.15) to 1 in 2.6E+07 (for RQ of 0.06).
[f] RQ < acute endangered species LOC of 0.05.

The screening-level methodology for aquatic plants (i.e., phytoplankton) and freshwater invertebrates (i.e., zooplankton) assumes risks to these taxonomic groups because the RQ values shown in Table 5.3 (based on the most sensitive toxicity data for non-vascular plants and freshwater invertebrates) exceed the Agency's LOCs. Although the listed species LOC is used for freshwater invertebrates, the non-listed species LOC is used for aquatic plants because the assessed mussels do not have an obligate relationship with any one particular type of phytoplankton as a food item. Further evaluation of the potential aquatic community-level effects that may result from atrazine exposure to phytoplankton and zooplankton as food sources for the listed mussels is provided as part of the risk description in Section 5.2.1.2.

The chronic RQs used to characterize potential indirect effects to the assessed mussels from direct acute effects on freshwater zooplankton food sources are provided in Table 5.2. Based on this analysis, LOCs were exceeded for chronic exposures to freshwater invertebrates for corn (all regions), sorghum (south and east regions), fallow/idle land (west region), and forestry (upper Great Plains region) uses of atrazine, with chronic RQ values ranging from 1.02 to 1.78. These exceedances are based on screening-level 21-day EECs and the toxicity data from the most sensitive freshwater invertebrate tested. The screening-level effects determination for indirect effects via direct chronic effects to freshwater invertebrate dietary items is "may affect" for all three species. Further analysis of potential chronic effects to aquatic invertebrates, as they relate specifically to zooplankton food items of the assessed mussels, is completed to determine if potential chronic risks to freshwater invertebrates are likely to adversely affect the assessed mussels in Section 5.2.1.2.

In summary, indirect effects based on direct impacts to the food supply "may affect" the three assessed mussels, because LOCs are exceeded for aquatic plants (i.e., phytoplankton) and freshwater invertebrates (i.e., zooplankton), which are food items of freshwater mussels. Additional analysis is needed to determine if the LOC exceedances, based on the most sensitive aquatic plant and freshwater invertebrate toxicity data and the screening-level EECs, are likely to adversely affect the assessed freshwater mussels.

5.1.2.2 Evaluation of Potential Indirect Effects via Reduction in Host Fish for Mussel Glochidia)

Freshwater mussels have a unique life cycle that involves a parasitic stage on host fish. Once mussel larvae (glochidia) fully develop, they are released into the water where they attach to the gills and fins of appropriate host fishes, which they parasitize for a short time until they develop into juvenile mussels. Glochidial hosts of the assessed mussel species are summarized in Table 2.3 and include freshwater drum, brown trout, different species of shiner and sculpin, and other warmwater fish. Potential indirect effects from direct effects on freshwater host fish were evaluated by considering the most sensitive freshwater fish effects data. The acute and chronic RQs used to characterize potential indirect effects to the assessed mussels from direct effects on freshwater host fish are provided in Tables 5.4 and 5.5, respectively. None of the acute LOCs were exceeded for freshwater fish based on the highest use pattern EECs; therefore, no indirect effects to the three listed mussels are expected based on direct acute effects to host fish.

Table 5.4 Summary of Acute RQs Used to Estimate Indirect Effects to the Listed Mussels via Direct Effects on Host Fish

Effect to Listed Mussels	Surrogate Species	Toxicity Value (µg/L)[a]	Peak EEC (µg/L)	RQ	Probability of Individual Effect[b]	LOC Exceedance and Risk Interpretation
Indirect effects to mussels via direct acute effects to host fish	Rainbow trout	LC_{50} = 5,300	South = 109[c]	0.021	1 in 3.98E+05 (1 in 226 to 1 in 2.97E+10)	No[d]
			North = 101[e]	0.019	1 in 7.03E+05 (1 in 276 to 1 in 9.31E+10)	No[d]

[a] Based on a 96-hour LC_{50} value of 5,300 µg/L for the rainbow trout (MRID# 000247-16).
[b] Based on a probit slope value of 2.72 for the rainbow trout with 95% confidence intervals of 1.56 and 3.89 (MRID #000247-16).
[c] Representative of the maximum screening-level acute exposure for the fat pocketbook mussel, based on peak southern corn screening-level EEC (Table 3.7).
[d] RQ < acute endangered species LOC of 0.05.
[e] Representative of the maximum screening-level acute exposure for the PCPP mussel and northern riffleshell, based on peak northern corn screening-level EEC (Table 3.7).

As shown in Table 5.5, the chronic LOC of 1.0 was exceeded for some uses based on the screening-level 60-day EECs and a freshwater fish NOAEC of 65 µg/L. Based on the distribution of the three assessed mussels within various regions of the action area, the screening-level effects determination for indirect effects via direct chronic effects to host fish is "may affect" for the fat pocketbook (for corn use in the south, north, west, and upper Great Plains and fallow/idle land use in the west), PCPP mussel (for corn use in the north), and northern riffleshell (for corn use in the north and sorghum use in the east). Therefore, indirect effects to the listed mussels via direct chronic effects to host fish may occur; however, acute toxicity to host fish is not expected. Further analysis of the potential effects to freshwater fish, as they relate to host availability for the assessed mussels, is completed to determine if potential chronic risks to fish are likely to adversely affect the assessed mussels in Section 5.2.1.3.

Table 5.5 Summary of Chronic RQs Used to Estimate Indirect Effects to the Listed Mussels via Direct Effects on Host Fish

Effect to Listed Mussels	Use (appl. Method; rate; # appl.; interval between appl.)	Range of 60-day EECs (µg/L)[a]	Freshwater Fish Chronic RQ (NOAEC= 65 µg/L)[b]	Chronic LOC Exceeded?
Indirect effects to mussels via direct chronic effects to host fish	Corn (aerial liquid; 2.5 lb ai/A; 2 appl.)	80 – 104	**1.23 – 1.60**	**Yes[c] (all regions: FP, PCPP, and NR)**
	Sorghum (aerial liquid; 2 lb ai/A; 1 appl.)	South: 60 North: 55 West: 57 UGP: 54 East: 66	South: 0.92 North: 0.85 West: 0.88 UGP: 0.83 **East: 1.02**	South: No[d] North: No[d] West: No[d] UGP: No[d] **East: Yes[c] (NR)**
	Fallow/Idle land (aerial liquid; 2.25 lb ai/A; 1 appl.)	South: 57 North: 51 West: 103 UGP: 49 East: 54	South: 0.88 North: 0.78 **West: 1.58** UGP: 0.75 East: 0.83	South: No[d] North: No[d] **West: Yes[c] (FP)** UGP: No[d] East: No[d]
	All other uses	≤ 58	≤ 0.89	No[d]

[a] 60-day screening-level EECs include the range of modeled concentrations from all five regions of the action area (Table 3.7). 60-day screening-level EECs from each of the five regions are provided for the sorghum and fallow/idle land scenarios in order to differentiate where chronic RQs exceed LOCs. UGP = upper Great Plains.
[b] Based on a 44-week NOAEC value of 65 µg/L for the brook trout (MRID #000243-77).
[c] RQ > chronic LOC of 1.0. Further evaluation of the RQs is necessary to determine if atrazine is likely to adversely affect the assessed species. FP = fat pocketbook; PCPP = purple cat's paw pearlymussel; NR = northern riffleshell
[d] RQ < chronic LOC of 1.0.

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

Potential indirect effects to the listed mussels via direct effects to habitat and/or primary production were assessed using RQs from freshwater aquatic vascular and non-vascular plant data as a screen. This screening-level analysis is based on the most sensitive EC_{50} value from all of the available non-vascular and vascular aquatic plant toxicity information. No known obligate relationship exists between the listed mussels and any single freshwater non-vascular or vascular plant species; therefore, endangered species RQs using the NOAEC/EC_{05} values for aquatic plants were not derived. If aquatic plant RQs exceed the Agency's non-endangered species LOC (because the assessed listed mussels rely on multiple plant species), potential community-level effects are evaluated using the threshold concentrations, as described in Section 4.2. RQs used to estimate potential indirect effects to the listed mussels from effects on aquatic plant primary productivity are summarized in Table 5.6.

Based on the results of this analysis, LOCs for direct effects to aquatic non-vascular plants are exceeded for all modeled atrazine use scenarios. LOCs for direct effects to aquatic vascular plants are also exceeded for modeled EECs based on corn, sorghum, fallow/idle land, and forestry (in all regions but the west); however, RQs are less than LOCs for use scenarios including forestry in the western region of the action area, residential, turf, and rights-of-way. Therefore, atrazine may indirectly affect the three listed mussels via effects to non-vascular aquatic plants for all modeled use scenarios and on vascular aquatic plants for the corn, sorghum, fallow/idle land, and forestry use scenarios. Further analysis of the potential effects to aquatic plant communities, as they relate to food availability and primary productivity for the assessed species, is used to determine if potential risks to aquatic plants are likely to adversely affect the assessed mussels. This refined analysis is presented in Section 5.2.1.4.

Table 5.6 Summary of RQs Used to Estimate Indirect Effects to the Listed Mussels via Effects to Aquatic Plants

Indirect Effect to Listed Mussels	Use (appl. Method; rate; # appl.; interval between appl.)	Range of Peak EECs (μg/L)[a]	Non-vascular plant RQ (EC_{50} = 1 μg/L)[b]	Vascular plant RQ (EC_{50} = 37 μg/L)[c]	LOC Exceedance
Reduced Habitat and/or Primary Productivity via Direct Toxicity to Aquatic Plants	Corn (aerial liquid; 2.5 lb ai/A; 2 appl.)	83 – 109	**83 – 109**	**2.24 – 2.95**	**Yes[d]**
	Sorghum (aerial liquid; 2 lb ai/A; 1 appl.)	57 – 69	**57 – 69**	**1.54 – 1.86**	**Yes[d]**
	Fallow/Idle land (aerial liquid; 2.25 lb ai/A; 1 appl.)	49 – 103	**49 – 103**	**1.32 – 1.59**	**Yes[d]**
	Forestry (aerial liquid; 4 lb ai/A; 1 appl.)	South: 46 North: 49 West: 27 UGP: 65 East: 44	**South: 46** **North: 49** **West: 27** **UGP: 65** **East: 44**	**South: 1.24** **North: 1.32** West: 0.73 **UGP: 1.76** **East: 1.19**	**South: Yes[d]** **North: Yes[d]** **West: Yes[e]** **UGP: Yes[d]** **East: Yes[d]**
	Residential (granular; 2 lb ai/A; 2 appl.; 30 d interval) and (liquid; 1 lb ai/A; 1 appl.)	7.6 – 20	**7.6 – 20**	0.21 – 0.54	**Yes[e]**
	Turf (granular; 2 lb ai/A; 2 appl.; 30 d interval) and (liquid; 1 lb ai/A; 1 appl.)	7.1 - 18	**7.1 - 18**	0.18 – 0.49	**Yes[e]**
	Rights-of-Way (liquid; 1 lb ai/A; 1 appl.)	2.4 – 3.8	**2.4 – 3.8**	0.06 – 0.10	**Yes[e]**

[a] Peak screening-level EECs include the range of modeled concentrations from all five regions of the action area (Table 3.7). Peak screening-level EECs from each of the five regions are provided for the forestry scenario in order to differentiate where vascular plant RQs exceed LOCs.

[b] Based on 1-week EC_{50} value of 1 µg/L for four species of freshwater algae (MRID # 000235-44).
[c] Based on 14-day EC_{50} value of 37 µg/L for duckweed (MRID # 430748-08).
[d] RQs > non-endangered aquatic plant species LOC of 1.0 for non-vascular and vascular plants. Direct effects to non-vascular and vascular aquatic plants are possible. Further evaluation of the EECs relative to the threshold concentrations (for community-level effects) is necessary.
[e] RQ > non-endangered aquatic plant species LOC of 1.0 for non-vascular plants; RQ < non-endangered plant species LOC of 1.0 for vascular plants. Direct effects to non-vascular aquatic plants are possible. Further evaluation of the EECs relative to the threshold concentrations (for community-level effects) is necessary.

5.1.2.4 Evaluation of Potential Indirect Effects via Reduction in Terrestrial Plant Community (Riparian Habitat)

Potential indirect effects to the listed mussels resulting from direct effects on riparian vegetation were assessed using RQs from terrestrial plant seedling emergence and vegetative vigor EC_{25} data as a screen. Based on the results of the submitted terrestrial plant toxicity tests, it appears that emerging seedlings are more sensitive to atrazine via soil/root uptake than emerged plants via foliar routes of exposure. However, all tested plants, with the exception of corn in the seedling emergence and vegetative vigor tests, and ryegrass in the vegetative vigor test, exhibited adverse effects following exposure to atrazine. The results of these tests indicate that a variety of terrestrial plants that may inhabit riparian zones may be sensitive to atrazine exposure. RQs used to estimate potential indirect effects to the listed mussels from seedling emergence and vegetative vigor effects on terrestrial plants within riparian areas are summarized in Tables 5.7 and 5.8, respectively.

As shown in Table 5.7, terrestrial plant RQs are above the Agency's LOC for all species except corn. For species with LOC exceedances, RQ values based on aerial application of atrazine to forestry at 4.0 lb ai/A range from 1.8 to 113; the maximum RQ value based on an equivalent ground application is 35, approximately a three-fold reduction as compared to aerial applications. Granular application of atrazine to residential lawns at 2.0 lb ai/A could also impact terrestrial plants with RQs ranging from <1 (corn and soybeans) to 13 (carrots). Monocots and dicots show similar sensitivity to atrazine; therefore, RQs are similar across both taxa.

Table 5.7 Non-target Terrestrial Plant Seedling Emergence RQs			
Surrogate Species	**EC_{25} (lbs ai/A)[a]**	**EEC Dry adjacent areas[b]**	**RQ Dry adjacent areas**
Monocot - Corn	> 4.0	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	<LOC
Monocot - Oat	0.004	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: 43 - 85 Ground: 13 - 26 Granular: 10**
Monocot - Onion	0.009	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: 19 - 38 Ground: 5.8 - 12 Granular: 4.4**
Monocot - Ryegrass	0.004	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: 43 -85 Ground: 13 - 26 Granular: 10**

Table 5.7 Non-target Terrestrial Plant Seedling Emergence RQs			
Surrogate Species	**EC_{25} (lbs ai/A)[a]**	**EEC Dry adjacent areas[b]**	**RQ Dry adjacent areas**
Dicot - Carrot	0.003	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: 57 - 113** **Ground: 17 - 35** **Granular: 13**
Dicot - Soybean	0.19	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: <LOC – 1.8** Ground: <LOC Granular: <LOC
Dicot - Lettuce	0.005	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: 34 - 68** **Ground: 10 - 21** **Granular: 8**
Dicot - Cabbage	0.014	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: 12 - 24** **Ground: 3.7 – 7.4** **Granular: 2.9**
Dicot - Tomato	0.034	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: 5 - 10** **Ground: 1.5 – 3.1** **Granular: 1.2**
Dicot - Cucumber	0.013	Aerial: 0.17 – 0.34 Ground: 0.05 – 0.10 Granular: 0.04	**Aerial: 13 - 26** **Ground: 4 - 8** **Granular: 3.1**

[a] From Chetram (1989); MRID 420414-03.
[b] Range of EECs based on use scenarios presented in Table 3.16 (i.e., aerial and ground: forestry, fallow/idle land, corn, sorghum; and granular residential).

Vegetative vigor studies indicate that terrestrial plants are generally less sensitive to foliar exposure of atrazine as compared to soil/root uptake. As shown in Table 5.8, vegetative vigor RQs exceed the Agency's LOC for only three dicot species (soybeans, cabbage, and cucumber), based on aerial application of atrazine at 2 to 4 lbs ai/A, with RQs ranging from 5 to 33. For ground applications, LOCs are exceeded for two dicot species, cabbage and cucumber, at application rates of 2 lbs ai/A with RQs ranging from 1.5 to 3. Vegetative vigor RQs do not exceed LOCs for any of the tested monocot species.

Table 5.8 Non-target Terrestrial Plant Vegetative Vigor Toxicity RQs			
Surrogate Species	**EC_{25} (lbs ai/A)[a]**	**Drift EEC (lbs ai/A)[b]**	**Drift RQ**
Monocot - Corn	> 4.0	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	<LOC
Monocot - Oat	2.4	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	<LOC
Monocot - Onion	0.61	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	<LOC
Monocot - Ryegrass	> 4.0	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	<LOC
Dicot - Carrot	1.7	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	<LOC
Dicot - Soybean	0.026	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	**Aerial: 5 - 10** Ground: <LOC
Dicot - Lettuce	0.33	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	<LOC
Dicot - Cabbage	0.014	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	**Aerial: 9.3 - 19** **Ground**: <LOC – **1.7**

Table 5.8 Non-target Terrestrial Plant Vegetative Vigor Toxicity RQs			
Surrogate Species	**EC_{25} (lbs ai/A)[a]**	**Drift EEC (lbs ai/A)[b]**	**Drift RQ**
Dicot - Tomato	0.72	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	<LOC
Dicot - Cucumber	0.008	Aerial: 0.13 – 0.26 Ground: 0.01 – 0.02	**Aerial: 16 - 33** **Ground: 1.5 – 3.0**

[a] From Chetram (1989); MRID 420414-03.
[b] Range of EECs based on use scenarios presented in Table 3.16 (i.e., aerial and ground: forestry, fallow/idle land, corn, and sorghum).

In summary, potential indirect effects to the three listed mussels resulting from direct effects on riparian vegetation may occur, based on the results of the screening-level analysis. Further evaluation of the potential for atrazine to affect the three listed mussels via reduction in riparian habitat, including a description of the importance of riparian vegetation to the assessed species and types of riparian vegetation that may potentially be impacted by atrazine use within the action area, is provided in Section 5.2.1.5.

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 three listed mussels.

If the RQs presented in the Risk Estimation (Section 5.1) show no indirect effects and LOCs for the three listed mussels are not exceeded for direct effects (RQs < LOC), a "no effect" determination is made, based on atrazine's use within the action area. If, however, direct or indirect effects to the individual listed mussels are anticipated (RQs > LOC), the Agency concludes a preliminary "may affect" determination for the FIFRA regulatory action regarding atrazine. A summary of the results of the risk estimation (i.e., "no effect" or "may affect" finding) presented in Sections 5.1.1 and 5.1.2 is provided in Table 5.9 for direct and indirect effects to the listed mussels. Conclusions of "may effect" based on RQs presented in Section 5.1 are further evaluated to distinguish actions that are likely to adversely affect ("LAA") from those that are not likely to adversely affect ("NLAA") the assessed mussel species.

Table 5.9 Preliminary Effects Determination Summary for the Assessed Listed Mussels Based on Risk Estimation		
Assessment Endpoint	**Preliminary Effects Determination**	**Basis for Preliminary Determination[a]**
1. Survival, growth, and reproduction of assessed mussel individuals via direct effects	Acute direct effects: No effect	No acute LOCs are exceeded (Table 5.1).
	Chronic direct effects: May affect	Chronic LOCs are exceeded for corn, sorghum, fallow/idle land, and forestry uses of atrazine, based on available chronic toxicity data from surrogate freshwater invertebrates (Table 5.2).
2. Indirect effects to	Phytoplankton: May affect	LOCs for phytoplankton are exceeded for all labeled uses

assessed mussel individuals via reduction in food items (i.e., freshwater phytoplankton and zooplankton)		of atrazine (Table 5.3).
	Zooplankton: May effect	Acute and chronic LOCs are exceeded for corn, sorghum, fallow/idle land and forestry uses (Tables 5.3 and 5.2).
3. Indirect effects to assessed mussel individuals via reduction in host fish for mussel glochidia	Acute indirect effects: No effect	Acute RQs for freshwater fish are less than LOCs (Table 5.4).
	Chronic indirect effects: May affect	Chronic LOCs are exceeded for corn, sorghum and fallow/idle land use of atrazine (Table 5.5)
4. Indirect effects to assessed mussel individuals via reduction of habitat and/or primary productivity	May affect	LOCs are exceeded for non-vascular aquatic plants for all modeled atrazine use scenarios (Table 5.6). LOCs are exceeded for vascular plants for the corn, sorghum, fallow/idle land, and forestry use scenarios (Table 5.6).
5. Indirect effects to assessed mussel individuals via reduction of terrestrial vegetation (i.e., riparian habitat) required to maintain acceptable water quality and habitat	May affect	LOCs are exceeded for all tested species except corn based on seedling emergence (Table 5.7). LOCs are exceeded for soybeans, cabbage, and cucumbers based on vegetative vigor (Table 5.8).
[a] All screening-level EECs for the preliminary effects determination are based on modeled scenarios for surface water (Table 3.7) and terrestrial plants (Table 3.16); toxicity values are based on the most sensitive endpoint summarized in Table 4.3.		

Following a “may affect” determination, additional information is considered to refine the potential for exposure at the predicted levels based on additional modeling and monitoring data, the life history characteristics (i.e., habitat range, feeding preferences, etc.) of the three listed mussels, and potential community-level effects to aquatic plants.

Two separate refined analyses were considered for the listed mussels, based on the species’ location within and outside the boundaries of vulnerable watersheds and site-specific flow information for occupied streams, as discussed in Section 3.2.1.

Based on the best available information, the Agency uses the refined evaluations to distinguish those actions that “may affect, but are not likely to adversely affect” (“NLAA”) from those actions that are “likely to adversely affect” (“LAA”) the three listed mussels (within and outside the boundary of vulnerable watersheds).

The criteria used to make determinations that the effects of an action are “not likely to adversely affect” the three listed mussels 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.

- Likelihood of the Effect Occurring: 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.

- Adverse Nature of Effect: Effects that are wholly beneficial without any adverse effects are not considered adverse.

A description of the risk and effects determination for each of the established direct and indirect assessment endpoints for the three listed mussels in occupied streams within and outside the boundaries of vulnerable watersheds, based on consideration of site-specific flow information, is provided in Section 5.2.1.

5.2.1 Direct and Indirect Effects to the Listed Mussels

5.2.1.1 Direct Effects to the Listed Mussels

The acute RQ of <0.003 (based on the peak EEC of 109 µg/L from the southern corn scenario) is well below the Agency's endangered species LOC for all modeled uses of atrazine within the action area. In addition, non-targeted NAWQA monitoring data (Section 3.2.6.2; Table 3.10) were also considered to provide context to the peak screening-level modeled EECs. The NAWQA data show that atrazine was detected within the action area at a similar peak concentration of 129 µg/L in Sugar Creek, Indiana. This watershed is located within the action area and within the boundary of vulnerable watersheds defined by WARP. Further analysis of the NAWQA monitoring data shows that the 99.9th percentile of all peak atrazine detections (from over 20,000 samples) is 61 µg/L (Table 3.15). Therefore, the 1997 Sugar Creek peak concentration of 129 µg/L is likely to overestimate current peak exposures of atrazine within this watershed. However, use of the peak value of 129 µg/L, would yield an acute RQ value of <0.004 (EEC = 129 µg/L/mussel LC_{50} = >36,000), which is also below the acute endangered species LOC of 0.05. The Agency, consistent with the Overview Document (U.S. EPA, 2004) and the alternative consultation agreement with the Services (USFWS/NMFS, 2004b), interprets RQs below the endangered species LOC to be consistent with a finding of no effect for direct effects on the listed species for the taxa being assessed.

To provide additional information, the probability of an individual mortality to the assessed mussels was calculated for acute RQs < 0.003. A probit slope value for the acute mussel toxicity test is not available; therefore, the effect probability was calculated based on a default slope assumption of 4.5 with upper and lower bounds of 2 and 9 (Urban and Cook, 1986). Based on the default dose response curve slope of 4.5, the corresponding estimated chance of an individual acute mortality to the listed mussels at an RQ level of <0.003 (based on the acute toxic endpoint for freshwater mussels) is 1 in 2.7E+29. It is recognized that extrapolation of very low probability events is associated with considerable uncertainty in the resulting estimates. In order to explore the possible bounds to such estimates, the upper and lower default bounds (2 to 9) were used to calculate upper and lower estimates of the effects probability associated with the acute RQ. The respective lower and upper effects probability estimates are 1 in 4.4E+06 and 1 in 5.1E+113.

In order to characterize potential acute direct effects to populations of fat pocketbook and northern riffleshell mussels that occur in streams with flow < 200 ft^3/sec (or for which no flow rate information is available) within the boundary of vulnerable watersheds, peak concentrations from the AEMP data were considered as an upper bound of exposure. Based on the AEMP data discussed in Section 3.2.6.1 and Table D-3 of Appendix D, atrazine was detected at a maximum peak concentration of 209 µg/L at sampling location IN 11. Atrazine was also detected at peak concentrations exceeding the PRZM/EXAMS pond screening-level EEC of 109 µg/L at an additional two locations including MO 01 (183 µg/L) and NE 07 (112 µg/L); however, peak concentrations from the remaining 37 watersheds were less than 109 µg/L with values ranging from 0.13 to 83 µg/L. Refinement of the peak EEC from 109 µg/L to 209 µg/L, based on the maximum detected peak concentration of atrazine from the AEMP data, would result in an acute RQ value of <0.006 (refined EEC: 209 µg/L/freshwater mussel LC_{50}: >36,000 µg/L). The acute RQ of < 0.006 is also well below the Agency's LOC.

In summary, the Agency concludes a "no effect" determination for acute direct effects to the three listed mussels, via acute mortality, based on all available lines of evidence.

Chronic toxicity data for freshwater mussels are not available; therefore, the most sensitive NOAEC value from the available freshwater invertebrate data was used as a surrogate. Chronic RQs, based on modeled screening-level EECs from Table 3.7 and the surrogate chronic freshwater invertebrate endpoint value for the scud (NOAEC = 60 µg/L), exceed the Agency's LOCs with RQ values ranging from 1.37 to 1.78. However, chronic RQs are likely to be overestimated given the available acute toxicity data for freshwater unionid mussels, which shows that mussels are less sensitive to atrazine than freshwater invertebrates routinely used in aquatic toxicity testing (i.e., cladocerons and amphipods). Available chronic data from the open literature suggest that growth effects to freshwater mollusks may occur at concentrations between 125 µg/L and 1,000 µg/L (NOAEC of 125 µg/L reported in Baturo, 1995; LOAEC of 1,000 µg/L reported in Streit and Peter, 1978). Although these studies were not considered appropriate for use in RQ calculations due to limitations in the study design and the lack of definitive NOAEC values (see Table A-21b of Appendix A), they suggest that chronic effects to freshwater mollusks may occur at concentrations between 125 µg/L and 1,000 µg/L.

Alternatively, potential use of an acute to chronic ratio (ACR) to estimate a chronic NOAEC for freshwater mussels was considered. ACRs were calculated for all freshwater invertebrate species where data allowed. However, there is a high degree of uncertainty in this analysis because acute and chronic studies conducted on the same species within the same laboratory were not available. Also, some acute studies reported non-discrete (i.e., "greater than") LC_{50} values. Non-discrete values were considered in the analysis only if they resulted in the highest (most conservative) ACR. The highest ACR across all freshwater invertebrate taxa is >300 (midge LC_{50} of >33,000 µg/L / midge NOAEC of 110 µg/L). If the ACR value of >300 was applied to the acute LC_{50} in freshwater mussels (>36,000 µg/L), the resulting estimated NOAEC would be approximately 120 µg/L. Therefore, use of the midge NOAEC of 60 µg/L is more conservative than the ACR-estimated NOAEC of 120 µg/L for freshwater mussels.

In addition, screening-level chronic EECs derived from the standard ecological water body are not likely to be representative of actual exposure concentrations in flowing water bodies where the fat pocketbook, PCPP mussel, and northern riffleshell occur. These listed mussel's inhabit flowing water bodies, which are subject to extensive mixing and dilution. In contrast, the standard ecological water body is assumed to be static.

As described in Section 3.2.5, additional modeling was completed to characterize the potential effect of flow on the screening-level EECs and provide refined chronic exposures for listed mussels that occupy streams outside the boundary of vulnerable watersheds and larger streams/rivers with flow rates > 200 ft^3/sec that are within the boundary of vulnerable watersheds.

Based on this analysis, flow-adjusted 21-day EECs are approximately 91 to 97% lower than 21-day EECs modeled using the static water body. This analysis suggests that screening-level EECs derived using the standard ecological water body may over-estimate exposure in water bodies with flowing water, including those where populations of the listed mussels occur. As shown in Table 3.8, 21-day flow-adjusted EECs (for the scenario yielding the highest screening-level EEC within each of the assessed geographic regions) range from 5 to 12 µg/L. Refined chronic RQ values based on the 21-day flow-adjusted EECs and most sensitive NOAEC of 60 µg/L range from 0.08 to 0.2, well below the Agency's LOC of 1.0 for chronic risk to aquatic invertebrates. Although predicted 21-day atrazine concentrations from the non-targeted monitoring data (21 µg/L; see Table 3.11) are approximately 2 times higher than the maximum predicted concentration based on flow-adjusted modeled EECs, consideration of the non-targeted monitoring data would also result in chronic RQs less than LOCs. Furthermore, consideration of the available open literature on freshwater mollusks indicates that potential chronic effects do not occur at estimated atrazine chronic exposure concentrations.

Consideration of the available targeted AEMP data from vulnerable watersheds confirms that longer-term screening-level EECs are likely to be overestimated by the static water body scenario. However, the highest flow-adjusted 21-day EEC of 12 μg/L may under-represent actual chronic exposure concentrations of atrazine in highly vulnerable areas based on the available AEMP data, which show a range of 21-day concentrations from 0.11 to 62 μg/L. Use of the maximum 21-day average AEMP concentration of 62 μg/L would result in a refined chronic RQ value that slightly exceeds the LOC of 1.0 (EEC of 62 μg/L / NOAEC of 60 μg/L = 1.03). Further review of the AEMP data shows that atrazine was detected at a concentration exceeding the freshwater invertebrate NOAEC (60 μg/L) in only one out of 40 sampled watersheds at NE 07. The range of 21-day average AEMP concentrations from the remaining 39 watersheds (excluding NE 07) is 0.11 to 44 μg/L. In addition, as discussed above, RQs for direct chronic effects to freshwater mussels based on the most sensitive aquatic invertebrate (freshwater scud) NOAEC value of 60 μg/L across all taxa are likely to be overestimated. The available chronic data from the open literature on freshwater mollusks suggest that direct effects may occur at concentrations between 125 and 1,000 μg/L, approximately two-fold higher than the maximum 21-day AEMP concentration of atrazine from highly vulnerable watersheds.

Therefore, atrazine's use within the action area is not likely to adversely affect the three listed mussels because refined flow-adjusted EECs and available targeted AEMP and non-targeted monitoring data indicate that chronic (21-day monitoring) exposure concentrations are unlikely to cause adverse chronic effects in mollusks. The effects determination for the assessment endpoint of direct chronic effects to the three listed mussels is "may affect, but not likely to adversely affect" or "NLAA." This finding is based on discountable effects (i.e., chronic effects to atrazine at the refined levels of exposure are not likely to occur and/or result in a "take" of a single fat pocketbook, PCPP mussel and northern riffleshell within the action area).

5.2.1.2 Indirect Effects via Reduction in Food Items (Freshwater Zooplankton and Phytoplankton)

Although data on the relative percentages of each type of food item in the listed mussel's diet are unavailable, freshwater mussels primarily consume phytoplankton, as well as detritus, zooplankton, and other microorganisms (Ukeles, 1971; Coker et al., 1921; Churchill and Lewis, 1924; and Fuller, 1974). Based on the screening-level analysis, LOCs are exceeded for phytoplankton for all labeled uses of atrazine within the action area. In addition, both screening-level acute and chronic RQs for zooplankton exceed their respective LOCs for agricultural (i.e., corn, sorghum, fallow/idle land) and forestry uses of atrazine. A description of the refined analysis for potential indirect effects to the listed mussels via reduction in zooplankton and phytoplankton as food items is provided below.

<u>Zooplankton</u>

With respect to zooplankton, screening-level acute RQs were based on the lowest LC_{50} value of 720 μg/L for the midge (*Chironomus* spp.). Consideration of all acute toxicity data for the midge shows a wide range of sensitivity within and between species of the same genus (2 orders of magnitude) with values ranging from 720 to >33,000 μg/L. Although effects data for the midge was used as a surrogate for dietary zooplankton, given that its lowest LC_{50} value is the most sensitive value for freshwater invertebrates, this species is generally not considered as zooplankton and is, therefore, unlikely to be consumed by the listed mussels. Freshwater zooplankton are dominated by four major groups of animals: protozoa, rotifers, and two subclasses of the Crustacea including the cladocerans and copepods. Out of the four major groups of animals considered as zooplankton, toxicity data for atrazine is available for cladocerons (*Daphnia*) only. As previously discussed in Section 4.1.3.1, acute atrazine LC_{50} values for *Daphnia* range from 3,500 to >30,000 μg/L. The acute RQ value for zooplankton, based on the maximum peak screening-level modeled EEC of 109 μg/L and the most sensitive *Daphnia* LC_{50} value of 3,500 μg/L is 0.03, less than the acute LOC. As previously discussed in Section 5.2.1.1, the available non-targeted NAWQA monitoring data from Sugar Creek show that atrazine was detected at a maximum peak concentration of 129 μg/L in 1997, similar to modeled peak screening-level EEC of 109 μg/L. However, it is unlikely that the NAWQA peak value from 1997 is representative of current peak exposures, given more recent 2000-2004 data from Sugar Creek, which show detected concentrations ≤ 29 μg/L, and the 99.9^{th} percentile of 61 μg/L from all peak NAWQA data. Based on the peak monitoring concentration of 129 μg/L, the refined acute RQ for zooplankton of 0.04 (refined EEC: 129 μg/L/*Daphnia* LC_{50}: 3,500 μg/L) is less than the acute LOC value of 0.05. In addition, LOCs are also not exceeded based on the 99.9^{th} percentile of all peak NAWQA monitoring data or recent data from 2000-2004 that is specific for Sugar Creek within the action area. Slope information on the toxicity study used to derive the RQ for zooplankton is not available. Therefore, the probability of an individual effect was calculated using the probit slope of 2.43 from an acute *Daphnia* study on the formulated product (80% ai) of atrazine; 95% confidence intervals could not be calculated based on the available data (MRID #420414-01). Based on the probit dose response curve slope of 2.42, the corresponding estimated chance of an individual effect to zooplankton at an RQ level of 0.04 is 1 in 2,790 (0.03%). Interpolation of the dose response curve shows an acute effect level (i.e., death or immobilization) for zooplankton of <1% at a peak exposure concentration of 129 μg/L.

Refined analysis of potential acute impacts to zooplankton in vulnerable watersheds where the three listed mussels occur is based on the AEMP data discussed in Section 3.2.6.1 and summarized in Appendix D. As previously discussed, the AEMP data show that atrazine was detected at a maximum peak concentration of 209 μg/L at sampling location IN 11, approximately two-fold higher than the modeled peak screening-level EEC of 109 μg/L. Based on the peak AEMP concentration of 209 μg/L, the revised acute RQ for zooplankton of 0.06 (refined EEC: 209 μg/L/*Daphnia* LC_{50}: 3,500 μg/L) exceeds the acute LOC value of 0.05. Therefore, the refined analysis for potential acute direct effects to zooplankton in vulnerable watersheds suggests that acute exposure to atrazine "may affect" the fat pocketbook and northern riffleshell mussels that occur in those vulnerable watersheds with flow rates < 200 ft^3/sec (or for which no flow rate information is available) via a reduction in food items. However, based on the probit dose response curve slope of 2.42, the corresponding estimated chance of an individual effect to zooplankton at an RQ level of 0.06 is 1 in 644 (0.2%). Interpolation of the dose response curve shows an acute effect level of approximately 1.5% for zooplankton at a peak exposure concentration of 209 μg/L.

In summary, zooplankton are not the primary food source for the listed freshwater mussels and there is a low probability of an individual level acute effect to zooplankton food items throughout the action area. In addition, available monitoring data within the action area suggest that estimates of atrazine concentrations in water are highly unlikely to cause acute effects to zooplankton. Therefore, the effects determination for indirect effects to the three listed mussels via direct acute effects on zooplankton as prey in vulnerable watersheds is "may affect, but not likely to adversely affect" or "NLAA". This finding is based on discountable and insignificant effects. The effects are discountable, given that refined exposures are not likely to cause acute effects to zooplankton and the low probability of an individual acute effect to zooplankton. Coupling the extremely low level of effect (i.e., <2% effect level at predicted levels of exposure) with the expectation that the sensitivity of the most sensitive species of zooplankton is likely to overestimate the sensitivity of the majority of zooplankton species available as food items, any predicted effects are also expected to be insignificant in context of "take" of a single listed fat pocketbook, PCPP mussel and northern riffleshell.

The screening-level chronic RQ for zooplankton, based on the highest modeled 21-day screening-level EEC of 107 μg/L and the most sensitive chronic freshwater invertebrate NOAEC of 60 μg/L for the scud, exceeds the Agency's LOC (see Table 5.2). However, as previously discussed, freshwater invertebrates including the scud are not considered as zooplankton; therefore, the effects data were refined to consider the available chronic atrazine toxicity data for cladocerons. The lowest NOAEC value for *Daphnia magna*, based on a reduction in the survival of F_0 young/female at 250 μg/L, is 140 μg/L. This NOAEC value of 140 μg/L is greater than the highest modeled 21-day screening-level EEC of 107 μg/L, as well as the highest refined 21-day flow-adjusted EEC of 12 μg/L. In addition, consideration of the 21-day atrazine concentrations from the non-targeted monitoring data (21 μg/L; see Table 3.11), although approximately 2 times higher than the maximum predicted based on flow-adjusted modeled EECs, would also result in chronic RQs less than LOCs.

Further refinement of potential chronic effects to zooplankton and resulting indirect effects to the fat pocketbook and northern riffleshell in vulnerable lower flowing watersheds (<200 ft^3/sec) is based on the targeted AEMP data. The AEMP data show that the maximum 21-day average concentration for atrazine is 62 μg/L, approximately two-fold lower than the corresponding 21-day screening-level EEC predicted by modeling. Use of the maximum 21-day average AEMP concentration of 62 μg/L would result in a refined chronic RQ value that exceeds the LOC (based on the most sensitive freshwater invertebrate NOAEC value of 60 μg/L). However, refined chronic effects data specific to cladocerons, which are considered to be representative of zooplankton, indicate that they are less sensitive to atrazine than the most sensitive freshwater invertebrate, with a corresponding NOAEC value of 140 μg/L. Therefore, chronic effects to zooplankton are not expected to occur at 21-day AEMP concentrations of 62 μg/L.

Given that all refined measures of exposure (i.e., 21-day flow adjusted EECs, non-targeted and targeted AEMP data) are well below levels that produced chronic effects in cladocerons, the effects determination for the three listed mussels via direct chronic effects on zooplankton as dietary food items is “may affect, but not likely to adversely affect” or “NLAA”. This finding is based on discountable effects (i.e., chronic effects to atrazine at the refined levels of exposure are not likely to occur and/or result in a “take” of a single listed fat pocketbook, PCPP mussel, and northern riffleshell via a reduction in zooplankton as food items).

Phytoplankton

As shown in Table 5.3, direct adverse effects to non-vascular aquatic plants (i.e., phytoplankton), which are the primary component of the listed mussel’s diet, are possible, based on all screening-level modeled atrazine uses. Direct effects to non-vascular plants are expected in all watersheds of the action area, based on peak detected concentrations of atrazine in the AEMP data (209 µg/L), the non-targeted NAWQA data from Sugar Creek (129 µg/L), as well as peak refined flow-adjusted EECs (109 µg/L). Based on these potential effects, atrazine may indirectly affect the three listed mussels via a reduction in food items required for growth and viability of juvenile and adult stages. In order to determine whether potential effects to individual plant species would likely result in community-level effects to the listed mussels, the time-weighted screening-level EECs (for 14-, 30-, 60-, and 90-day averages from Table 3.8) were compared to their respective time-weighted threshold concentrations. As discussed in Section 4.2, concentrations of atrazine from the exposure profile at a particular use site and/or action area that exceed any of the following time-weighted threshold concentrations indicate that changes in the aquatic plant community structure (including food items for the mussels) could be affected:

- 14-day average = 38 µg/L
- 30-day average = 27 µg/L
- 60-day average = 18 µg/L
- 90-day average = 12 µg/L

A comparison of the range of the screening-level 14-, 30-, 60-, and 90-day EECs for the listed mussels with the atrazine threshold concentrations representing potential aquatic community-level effects is provided in Table 5.10.

Table 5.10 Summary of Modeled Scenario Time-Weighted Screening-Level EECs with Threshold Concentrations for Potential Community-Level Effects

Use Scenario	14-day		30-day		60-day		90-day	
	EECs (µg/L) [a]	Threshold Conc. (µg/L)	EECs (µg/L) [a]	Threshold Conc. (µg/L)	EECs (µg/L) [a]	Threshold Conc. (µg/L)	EECs (µg/L) [a]	Threshold Conc. (µg/L)

Corn	**82 - 108**		**81 - 106**		**80 - 104**		**78 - 101**	
Sorghum	**57 - 68**		**56 - 68**		**54 - 66**		**53 - 64**	
Fallow / idle land	**49 - 103**		**49 - 103**		**49 - 103**		**49 - 103**	
Forestry	27 - **48**	38	26 - **60**	27	**26 - 58**	18	**25 - 57**	12
Residential	8 - 20		8 - 19		8 - **19**		8 - **18**	
Turf	7 - 18		7 - 18		7 - **18**		7 - **17**	
Rights-of-Way	2 - 4		2 - 4		2 - 4		2 - 4	

[a] Screening-level EECs from Table 3.7.

Based on the results of this comparison, predicted screening-level 14-, 30-, 60-, and 90-day EECs for corn, sorghum, fallow/idle land, and forestry modeled uses exceed their respective threshold concentrations for community level effects. In addition, predicted 60- and 90-day EECs for residential and turf uses of atrazine exceed their respective threshold concentrations. These screening-level EECs were estimated using PRZM/EXAMS and the non-flowing standard water body scenario, which is intended to be representative of exposures in headwater streams. As previously discussed, these chronic screening-level EECs are expected to over-estimate exposure in both vulnerable and less vulnerable water bodies with flowing water, where the listed mussels are known to occur. All of the listed mussels included in this assessment require flowing waters over relatively stable sand, gravel, cobble substrates for normal feeding, growth, and viability of all life stages; therefore, chronic EECs based on a non-flowing water body are expected to over-estimate actual exposure concentrations of atrazine for the assessed mussels in their expected range. Additional flow-adjusted EECs and available non-targeted and targeted monitoring data was used to refine exposure concentrations of atrazine for the three assessed mussels, relative to those presented for the standard water body scenario. Analyses of flow-adjusted EECs and relevant monitoring data are presented in detail in Sections 3.2.5 and 3.2.6, respectively, and summarized below.

In order to characterize the potential impact of flowing water on the longer-term exposures (i.e., 14 through 90-days) in less vulnerable watersheds and for large streams/rivers with flow > 200ft^3/sec within the boundary of vulnerable watersheds, further modeling was conducted to provide a general sense of the relative reduction in long term exposure that might occur in water bodies where flow is higher than small headwater streams. The results of this analysis show that the flow-adjusted modeling yields longer-term EECs that are reduced as compared to screening-level EECs derived using the standard water body. A comparison of the maximum flow-adjusted 14-, 30-, 60-, and 90-day EECs for two atrazine corn use scenarios in the southern and northern regions (that are representative of the distribution of all three listed mussels) with the atrazine threshold concentrations representing potential aquatic community-level effects is provided in Table 5.11.

Table 5.11 Summary of Flow-Adjusted EECs with Threshold Concentrations for Potential Community-Level Effects in Less Vulnerable Watersheds

Use Scenario	14-day		30-day		60-day		90-day	
	EECs (µg/L)	Threshold Conc. (µg/L)	EECs (µg/L)	Threshold Conc. (µg/L)	EECs (µg/L)	Threshold Conc. (µg/L)	EECs (µg/L)	Threshold Conc. (µg/L)
Corn[a]	14 - 16	38	7 - 8	27	3 - 4	18	2- 3	12

[a] Range of flow-adjusted EECs for corn are based on the percentage decrease in maximum screening-level EECs using USGS mean seasonal flow data for the southern and northern regions (Table 3.8).

As shown in Table 5.11, refined flow-adjusted 14-, 30-, 60- and 90-day EECs based on atrazine use patterns that yield the highest screening-level EECs for the regions occupied by the listed mussels (i.e., corn in the southern and northern regions), are well below their respective threshold concentrations. Although monitoring data from non-targeted areas show that longer-term concentrations of atrazine exceed the maximum flow-adjusted EECs by approximately a factor of 2, consideration of similar duration exposures from non-targeted monitoring data (Table 3.12) confirm that all long-term atrazine concentrations are also less than their respective threshold concentrations. It should be noted that the non-targeted data were collected from the Sadusky watershed, which is located within the boundary of vulnerable watersheds; therefore, use of this data is considered as a conservative estimate of exposure in less vulnerable watersheds. The flow-adjusted 14- through 90-day EECs would have to increase by a factor of approximately three to four to exceed the threshold concentrations. However, it is unlikely that flow-adjusted EECs underpredict atrazine exposure in streams and rivers that are outside the boundary of vulnerable watersheds or those watersheds within the boundary of vulnerable watersheds with flow rates > 200 ft^3/sec.

Although atrazine use may indirectly affect individual aquatic non-vascular plants that comprise the majority of the listed mussel's diet, its use within less vulnerable watersheds as well as larger streams/rivers (with flow rates > 200 ft^3/sec) within the boundary of vulnerable watersheds is not likely to indirectly affect the three listed mussels via a reduction in phytoplankton food items. This finding is based on insignificance of effects (i.e., although effects to individual plants may occur, community-level effects to non-vascular plants cannot be meaningfully measured, detected, or evaluated in the context of a "take" of a single PCPP mussel within the entire action area, and fat pocketbook and northern riffleshell mussel located in less vulnerable watersheds or higher flowing streams/rivers within the boundary of vulnerable watersheds). Therefore, the effects determination for the assessment endpoint of indirect effects on the PCPP mussel (in the entire action area) and fat pocketbook and northern riffleshell (in less vulnerable watersheds and larger streams/rivers with flow rates > 200ft^3/sec in vulnerable watersheds) via direct effects on prey (i.e., phytoplankton) is "may affect, but not likely to adversely affect" or "NLAA."

In addition to the modeling exercises, the Agency used the AEMP data to further characterize atrazine concentrations in the lower flow (< 200 ft^3/sec) portions of vulnerable watersheds where the fat pocketbook and northern riffleshell mussels are known to occur. Consideration of the AEMP data from vulnerable watersheds in Section 3.2.6.1 and Appendix D confirms that longer-term screening-level EECs are likely to be overestimated by the static water body scenario. However, the flow-adjusted 14-, 30-, 60-, and 90-day EECs presented in Table 5.11 appear to under-represent actual chronic exposure concentrations of atrazine in vulnerable areas under some conditions based on the AEMP data. As shown in Tables D-3 and D-4 of Appendix D, the flow-adjusted chronic EECs are less than their corresponding rolling averages from the AEMP data in approximately 25 to 43% of the sampled watersheds. Therefore, the AEMP data rolling averages are used to determine whether community-level effects may occur for aquatic non-vascular plants in vulnerable areas (with flow rates < 200 ft^3/sec or for which no information is available) that are occupied by the fat pocketbook and northern riffleshell mussels. Comparison of the range of rolling averages from the AEMP data with their corresponding threshold concentrations is provided in Table 5.12.

Table 5.12 Summary of AEMP Data Rolling Averages with Threshold Concentrations for Potential Community-Level Effects in Vulnerable Watersheds

14-day		**30-day**		**60-day**		**90-day**	
Range of EECs (µg/L)[a]	**Threshold Conc. (µg/L)**	**Range of EECs (µg/L)[a]**	**Threshold Conc. (µg/L)**	**Range of EECs (µg/L)[a]**	**Threshold Conc. (µg/L)**	**Range of EECs (µg/L)[a]**	**Threshold Conc. (µg/L)**
0.11 – **80**[a] (7.5%)[e]	38	0.10 – **62**[b] (12.5%)[e]	27	0.10 – **26**[c] (5%)[e]	18	0.10 – **18**[d] (5%)[e]	12

[a] Range of 14-day rolling averages from the AEMP data in Table D-3 of Appendix D. Maximum 14-day average concentrations exceed the threshold concentration of 38 µg/L at the following locations: IN 11 (65 µg/L), MO 01 (40-78 µg/L), and NE 07 (80 µg/L).
[b] Range of 30-day rolling averages from the AEMP data in Table D-3 of Appendix D. Maximum 30-day average concentrations exceed the threshold concentration of 27 µg/L at the following locations: IN 11 (32 µg/L), MO 01 (29-43 µg/L), MO 02 (27-32 µg/L), NE 04 (27 µg/L) and NE 07 (45 µg/L).
[c] Range of 60-day rolling averages from the AEMP data in Table D-3 of Appendix D. Maximum 60-day average concentrations exceed the threshold concentration of 18 µg/L at the following locations: MO 01 (19-26 µg/L)and NE 07 (23 µg/L).
[d] Range of 90-day rolling averages from the AEMP data in Table D-3 of Appendix D. Maximum 90-day average concentrations exceed the threshold concentration of 12 µg/L at the following locations: MO 01 (12-18 µg/L) and MO 02 (12 µg/L).
[e] Percentage of watersheds (N = 40) that exceed the corresponding threshold concentration.

As shown in Table 5.12, 14-, 30-, 60- and 90-day rolling averages based on the AEMP data exceed their respective threshold concentrations for a small number of watersheds ranging from approximately 5 to 12.5 percent of the total. Data from the following sites exceeded at least one of the threshold concentrations: IN 11, MO 01, MO 02, NE 04, and NE 07. It should be noted, however, that a number of watersheds, particularly in Nebraska (NE), experienced dry periods where scheduled sampling did not take place; therefore, the statistics for watersheds including NE 04 and NE 07 may not represent actual conditions expected in normal or wetter years. In addition, it is unlikely that freshwater mussels would inhabit these types of streams. Although it is uncertain if these sites are representative of the streams and rivers where the fat pocketbook and northern riffleshell occur, it is assumed, until further analysis is available, that data from these watersheds may be representative of chronic atrazine exposure conditions in vulnerable lower flow (< 200 ft^3/sec) watersheds within the action area where these species occur. Therefore, community-level effects are possible for non-vascular plants within vulnerable watersheds (with flow rates < 200 ft^3/sec or for which no flow data are available) of the action area where the fat pocketbook and northern riffleshell feed on phytoplankton. The effects determination for the assessment endpoint of indirect effects on the fat pocketbook and northern riffleshell mussels via direct effects on phytoplankton as food is "may affect and likely to adversely affect" or "LAA" for populations that occur in highly vulnerable, lower flow (< 200 ft^3/sec) watersheds of the action area. With the exception of large streams/rivers with flow > 200 ft^3/sec, the range of the LAA determination for the fat pocketbook and northern riffleshell mussels is depicted in Figures 3.7 and 3.8, respectively.

Given this "LAA" finding, the Agency has completed a summary of the environmental baseline and cumulative effects for the fat pocketbook and northern riffleshell mussel species included in this assessment in Appendix H. The environmental baseline is defined as the effects of past and ongoing human induced and natural factors leading to the status of the species, its habitat, and ecosystem, within the action area. The baseline information provides a snapshot of the assessed mussel's status at this time. A summary of all USFWS biological opinions that are relevant to the fat pocketbook and northern riffleshell mussels that have been made available to EPA included in this assessment is also provided as part of the baseline status. Cumulative effects include the effects of future state, tribal, local, private, or other non-federal entity activities on endangered and threatened species that are reasonably expected to occur in the action area.

5.2.1.3 Indirect Effects via Reduction in Host Fish

The highest RQ based on the highest PRZM/EXAMS screening-level EEC (southern corn scenario) and the lowest freshwater fish LC_{50} value is 0.02, which is less than the acute LOC of 0.05. As previously discussed, recent targeted AEMP and non-targeted monitoring report peak EECs that are approximately 2-fold higher than the highest peak screening-level EEC used to calculate RQs. Based on the highest peak EEC from the AEMP data in vulnerable watersheds, the acute RQ would be 0.04 (EEC of 209 µg/L / LC_{50} of 5,300 µg/L = RQ of 0.04), which is also below the acute LOC. Given that acute LOCs are not exceeded based on screening-level and refined EECs, the effects determination for direct acute effects on freshwater host fish necessary for mussel glochidia of the three listed mussels is "no effect".

Chronic RQs, which are based on modeled screening-level 60-day EECs and the surrogate freshwater fish chronic endpoint value for brook trout (NOAEC = 65 µg/L), exceed the Agency's LOCs for corn, sorghum, and fallow/idle land uses with RQ values ranging from 1.02 to 1.6 (see Table 5.5). However, as previously discussed, chronic RQs based on screening-level EECs (derived using the PRZM/EXAMS pond scenario) are likely to be overestimated given that freshwater mussels are known to occur in flowing water bodies, where chronic atrazine exposures are expected to be lower than 60-day exposure concentrations in a static pond. Based on the analysis conducted in Section 3.2.5, flow-adjusted 60-day EECs are approximately 96 to 98% lower than 60-day EECs modeled using the static water body. As shown in Table 3.8, 60-day flow-adjusted EECs (for the scenarios yielding the highest screening-level EEC from within each of the four geographic regions) range from 2 to 4 µg/L. In addition, the previously discussed non-targeted and targeted AEMP data report maximum 60-day rolling averages of 21 and 26 µg/L, respectively. All of the 60-day AEMP EECs are lower than the most sensitive life-cycle NOAEC of 65 µg/L by roughly a factor of three. The refined chronic RQ value based on the 60-day flow-adjusted EEC is 0.06, and the chronic RQs based on the 60-day EECs from non-targeted and AEMP monitoring data are <0.4. Therefore, all refined RQ values are below the Agency's LOC of 1.0 for chronic risk to freshwater fish.

As discussed in Section 4.1.2.3, several open literature studies raise questions about sublethal effects of atrazine on plasma steroid levels, behavior modifications, gill physiology, neurophysiological, and endocrine-mediated functions in freshwater fish and anadromous fish. Consideration of the sublethal data indicates that effects associated with alteration of gill physiology and endocrine-mediated olfactory functions may occur in anadromous fish including salmon at atrazine concentrations as low as 1 µg/L (Waring and Moore, 2004; Moore and Lower, 2001). In addition, Tierney et al. (2007) observed hyperactivity and neurophysiological responses in juvenile rainbow trout exposed to atrazine at 1 and 10 µg/L, respectively. However, there are a number of limitations in the design of these studies, which are addressed in detail in Sections A.2.4 of Appendix A, that preclude quantitative use of the data in this risk assessment. For example, Moore and Lower (2001) and Tierney et al. (2007) exposed epithelial tissue (after removal of skin and cartilage) and not intact fish to atrazine, and potential solvent effects could not be reconciled (i.e., no negative control was tested). Furthermore, no quantitative relationship is established between reduced olfactory response (measured as electrophysiological response) of epithelial tissue to the priming hormone and/or amino acid odorant in the laboratory and reduction in fish imprinting and homing, alarm response, and reproduction (i.e., the ability of trout to detect, respond to, and mate with ovulating females) in the wild. Other sublethal effects observed in fish studies have included behavioral modifications, alterations of plasma steroid levels, and changes in kidney histology at atrazine concentrations ranging from 5 to 35 µg/L (see Section 4.1.2.3). However, a number of uncertainties were also identified with each of the studies, which are discussed in Section A.2.4 of Appendix A.

In summary, it is not possible to quantitatively link the sublethal effects to the selected assessment endpoints for the listed mussels (i.e., survival, growth, and reproduction of individuals). Also, effects to reproduction, growth, and survival were not observed in the four submitted fish life-cycle studies at levels that produced the reported sublethal effects (Appendix A).

Although atrazine RQs based on the static water body EECs and a NOAEC of 65 µg/L exceed the chronic LOC of 1.0, its use within the action area is not likely to adversely affect the listed mussels via reduction in available fish hosts because flow-adjusted EECs and available monitoring data indicate that atrazine concentrations are not likely to result in adverse chronic growth effects to fish. Therefore, the effects determination for the assessment endpoint of indirect effects to the listed mussels via direct chronic effects to host fish is "may affect, but not likely to adversely affect" or "NLAA." This finding is based on discountable effects (i.e., chronic exposure to atrazine is not likely to result in a "take" of a single listed fat pocketbook, PCPP mussel, and northern riffleshell because direct chronic effects to host fish are unlikely to occur).

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

Based on the static pond scenario, the non-vascular aquatic plant LOC of 1.0 was exceeded for all modeled uses. In addition, vascular plant RQs also exceeded the LOC of 1.0 for corn, sorghum, fallow/idle land, and forestry uses of atrazine. Direct effects to vascular and non-vascular plants are expected in both vulnerable and less vulnerable watersheds of the action area, based on peak detected concentrations of atrazine in the AEMP data and non-targeted NAWQA data, which are up to two-fold higher than predicted peak modeled EECs. Based on these potential screening-level direct effects to aquatic plants, atrazine may indirectly affect the three listed mussels by reducing food supply and primary productivity. Therefore, screening-level time-weighted EECs (for 14-day, 30-day, 60-day, and 90-day averages) were compared to their respective community level effects threshold concentrations to determine whether potential effects to individual plant species are likely to result in community level effects.

A comparison of the screening-level 14-, 30-, 60-, and 90-day EECs for the listed mussels with the atrazine threshold concentrations representing potential aquatic community-level effects is provided in Table 5.12 as part of the risk description for indirect effects to listed mussels based on a reduction of dietary phytoplankton. The results of this analysis (Section 5.2.1.2) show that screening-level EECs exceed threshold concentrations indicative of community-level effects for all durations and modeled atrazine uses with the exception of rights-of-ways. The screening-level EECs were refined by considering site-specific flow data and available non-targeted and targeted AEMP monitoring data because screening-level EECs are expected to over-estimate exposure in flowing water bodies where the listed mussels occur.

Comparison of the refined flow-adjusted EECs with respective threshold concentrations is shown in Table 5.11 and also discussed in Section 5.2.1.2. The results of this comparison show that flow-adjusted EECs for all atrazine uses and available non-targeted monitoring data are well below threshold concentrations (all durations) for community level effects; therefore, atrazine use in the less vulnerable watersheds of the action area and large streams/rivers with flow > 200ft^3/sec in vulnerable watersheds is not likely to adversely affect the three listed mussels that occupy these types of watersheds via community-level effects to aquatic vegetation. As previously discussed, the flow-adjusted 14- through 90-day EECs would have to underpredict exposures by a factor of approximately three to four to result in exceedance of the threshold concentrations. However, it is unlikely that flow-adjusted EECs underpredict longer-term atrazine exposure in higher flow (> 200 ft^3/sec) streams and rivers of the action area for reasons previously discussed. Therefore, the effects determination for the assessment endpoint of indirect effects on the PCPP mussel (in the entire action area) and the fat pocketbook and northern riffleshell (in occupied streams in less vulnerable watersheds and larger streams/rivers with flow rates > 200 ft^3/sec within the boundary of vulnerable watersheds) via direct effects on habitat and/or primary productivity of aquatic plants is "may affect, but not likely to adversely affect" or "NLAA". This finding is based on insignificance of effects (i.e., although effects to individual plants may occur, community-level effects to aquatic plants cannot be meaningfully measured, detected, or evaluated in the context of a "take" of a single PCPP mussel (within the entire action area), and fat pocketbook and northern riffleshell (in less vulnerable watersheds and higher flowing streams/rivers > 200 ft^3/sec within the boundary of vulnerable watersheds).

AEMP data were also used to further characterize atrazine concentrations for populations of the fat pocketbook and northern riffleshell that occur in vulnerable watersheds of the action area where the flow rate is < 200 ft^3/sec (and/or flow rate information is not available). As shown in Table 5.12, 14-, 30-, 60-, and 90-day rolling averages based on the AEMP data from vulnerable watersheds exceed their respective threshold concentrations. Therefore, community-level effects are possible for aquatic plants within vulnerable lower flow watersheds of the action area where the fat pocketbook and northern riffleshell mussels occur (Figures 3.7 and 3.8). However, as previously discussed, there is uncertainty associated with use of the AEMP data from Nebraska, where the sampling locations went "dry", and listed species of mussels are not expected to occur. Despite these uncertainties, the effects determination for the assessment endpoint of indirect effects via direct effects on habitat and/or primary productivity of aquatic plants is "may affect and likely to adversely affect" or "LAA" for fat pocketbook and northern riffleshell mussels that occur in vulnerable watersheds with flows < 200 ft^3/sec of the action area. The range of the LAA determination for the fat pocketbook and northern riffleshell is depicted in Figures 3.7 and 3.8, respectively; however, it should be noted that these figures also include the species' range within larger streams and rivers (> 200 ft^3/sec) in vulnerable watersheds that are not included in the "LAA" determination. As previously discussed, a summary of the environmental baseline and cumulative effects for the fat pocketbook and northern riffleshell mussel species included in this assessment is provided in Appendix H.

5.2.1.5 Indirect Effects via Alteration in Terrestrial Plant Community (Riparian Habitat)

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

- Atrazine may enter riparian areas via runoff where it may be taken up through the root system of sensitive plants.

- Comparison of seedling emergence EC_{25} values to EECs estimated using TERRPLANT suggests that inhibition of new growth may occur. Inhibition of new growth could result in degradation of high quality riparian habitat over time because as older growth dies from natural or anthropogenic causes, plant biomass may be prevented from being replenished in the riparian area. Inhibition of new growth may also slow the recovery of degraded riparian areas that function poorly due to sparse vegetation because atrazine deposition onto bare soil would be expected to inhibit the growth of new vegetation.

- Because LOCs were exceeded for most species tested (9/10) in the seedling emergence studies, it is likely that many species of herbaceous plants may be potentially affected by exposure to atrazine in runoff.

A number of dicots in riparian habitats may also be impacted via foliar exposure from atrazine in spray drift as evidenced by vegetative vigor LOC exceedances in three dicots. Therefore, riparian habitats comprised of herbaceous plants sensitive to atrazine may be adversely affected by spray drift. However, comparison of the seedling emergence and vegetative vigor RQs indicates that runoff, and not spray drift, is a larger contributor to potential risk for riparian vegetation. Vegetative vigor risk quotients were not exceeded for monocots; therefore, drift would not be anticipated to affect riparian zones comprised primarily of monocot species such as grasses.

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

Riparian plants beneficially affect water and stream quality in a number of ways (discussed below) in both adjacent river reaches and areas downstream of the riparian zone. Atrazine use in the action area, which is inclusive of the listed mussels range, may potentially affect these species by impacting riparian vegetation and subsequently causing sedimentation that results in degraded water quality and alteration of available habitat. In order to characterize the potential indirect effects caused by atrazine-related impacts to riparian vegetation, a general discussion of riparian habitat and its relevance to the listed mussels and a description of the types of riparian zones that may be potentially impacted by atrazine use in the action area for the listed mussels are discussed below.

Importance of Riparian Habitat to the Listed Mussels

Riparian vegetation provides a number of important functions in the stream/river ecosystem, including the following:

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

The specific characteristics of a riparian zone that are optimal for the listed mussels are expected to vary with developmental stage, the use of the reach adjacent to the riparian zone, and the hydrology of the watershed. Criteria developed by Fleming et al. (2001) have been used to assess the health of riparian zones and their ability to support habitat for aquatic communities. These criteria, which include the width of vegetated area (i.e. distance from cropped area to water), structural diversity of vegetation, and canopy shading, are summarized in Table 5.13.

Table 5.13 Criteria for Assessing the Health of Riparian Areas to Support Aquatic Habitats (adapted from Fleming et al. 2001)

	Quality			
Criteria	**Excellent**	**Good**	**Fair**	**Poor**
Buffer width	>18m	12 - 18m	6 - 12m	<6m
Vegetation diversity	>20 species	15 - 20 species	5 - 14 species	<5 species
Structural diversity	3 height classes grass/shrub/tree	2 height classes	1 height class	sparse vegetation
Canopy shading	mixed sun/shade	sparse shade	90% sun	no shade

To maintain at least "good" water quality for aquatic habitats in general, riparian areas should contain at least a 12 m (~40 feet) wide vegetated area, 15 plant species, vegetation of at least two height classes, and provide at least sparse shade (>10% shade). In general, higher quality riparian zones (wider vegetated areas with greater plant diversity) are expected to have a lower probability of being affected by atrazine than poor quality riparian areas (narrower areas with less vegetation and little diversity).

The following three attributes of riparian vegetation habitat quality were evaluated for this assessment: stream bank stability, sedimentation, and thermal stability. Each of these attributes and their relative importance with respect to the listed mussels is discussed briefly below.

<u>Stream and river bank stabilization:</u> Riparian vegetation typically consists of three distinct height classes of plants, which include a groundcover of grasses and forbs, an understory of shrubs and young trees, and an overstory of mature trees. These plants serve as structural components for streams, with the root systems helping to maintain stream stability, and the large woody debris from the mature trees providing instream cover. Riparian vegetation has been shown to be essential to maintenance of a stable stream (Rosgen, 1996). Destabilization of the stream can have a severe impact on aquatic habitat quality. Following a disturbance in the watershed bank, the stream may widen, releasing sediment from the stream banks and scouring the stream bed. Changes in depth and or the width/depth ratio via physical modification to the stability of stream and river banks may also affect light penetration and the flow regime of the listed mussel's habitat. Destabilization of the stream can have severe effects on aquatic habitat quality by increasing sedimentation within the watershed. The effects of sedimentation are summarized below.

<u>Sedimentation:</u> Sedimentation refers to the deposition of particles of inorganic and organic matter from the water column. Increased sedimentation is caused primarily by disturbances to river bottoms and streambeds and by soil erosion. Riparian vegetation is important in moderating the amount of sediment loading from upland sources. The roots and stems of riparian vegetation can intercept eroding upland soil (USDA NRCS, 2000), and riparian plant foliage can reduce erosion from within the riparian zone by covering the soil and reducing the impact energy of raindrops onto soil (Bennett, 1939).

Freshwater mussels require flowing, silt free streams and rivers in order to survive. Therefore, they are susceptible to adverse effects caused by sedimentation in waterways. Specific biological impacts on mussels from excessive sediments include reduced feeding and respiratory efficiency from clogged gills, disrupted metabolic processes, reduced growth rates, increased substrata instability, limited burrowing activity and physical smothering (Ellis, 1936; Stansbery, 1971; Markings and Bills, 1979; Kat, 1982; Vannote and Minshall, 1982; Aldridge et al., 1987; and Waters, 1995). Physical effects of sediment on the listed mussels appear to be multifold, and include changes in suspended and bed material load; alteration in bed sediment composition; changes in channel form, position, and degree of stability; alteration of light penetration via turbidity; active aggrading (filling) or degrading (scouring) of channels; and changes in channel position that may reduce suitable habitat for mussels (Vannote and Minshall, 1982; Kanehl and Lyons, 1992; Brim Box and Mossa, 1999).

Interstitial spaces in the substrate also provide crucial habitat for juvenile mussels. When clogged due to sedimentation, interstitial flow rates and spaces become reduced (Brim Box and Mossa, 1999), thus reducing juvenile mussel habitat. Sediments also act as a means of transport for delivering contaminants such as nutrients and pesticides to streams. Juveniles can readily ingest contaminants adsorbed to silt particles or in interstitial pore water during normal feeding activities (Yeager et al., 1994; Newton, 2003).

According to the USFWS Recovery Plans for the three listed mussel species (USFWS, 1989, 1992, and 1994), the greatest impact on the habitat of the assessed mussels is related to navigation and flood control activities associated with channelization and impoundments. These impoundments and other related flood control measures cause increased siltation, reduce the availability of riverine habitat, and likely affect the distribution and availability of the mussel's host fish. Associated impacts related to sedimentation are also cited as a primary cause for the decline of freshwater mussels in the USFWS recovery plans for the three listed species. Excessive sediments deposited on stream bottoms can smother and kill relatively immobile bottom-dwelling species such as freshwater mussels and can eliminate more mobile aquatic species (such as host fish) by making their habitat unsuitable for feeding or reproduction (Brookes, 1994; National Research Council, 1992; Waters, 1995; Hartfield and Hartfield, 1996). Increased sedimentation may affect the spawning habitat of host fish by settling on spawning gravel and reducing flow of water and dissolved oxygen to the eggs and fry (Everest et al., 1987). In addition, fine particles settling on the streambed can also disrupt the food chain by reducing habitat quality for aquatic invertebrates, and adversely affect groundwater-surface water interchange (Nelson et al., 1991). Increased turbidity from sediment loading may also reduce light transmission, potentially affecting aquatic plants (Cloern, 1987; Weissing and Huisman, 1994) that are important source of food for the listed mussels.

Thermal stability. Riparian habitat including mature woody trees provides stream shading resulting in thermal stability. Although the sensitivity of the listed mussels to fluctuations in water temperature are unknown, stream shading has been shown to be positively correlated with freshwater unionid mussel species richness and density (Arbuckle and Downing, in press; obtained from http://limnology.eeob.iastat.edu/Studies/MusselStudies/FinalReport/Chapter4.htm; January 25, 2007).

Sensitivity of Forested Riparian Zones to Atrazine

As previously summarized in Table 5.13, the parameters used to assess riparian quality include buffer width, vegetation diversity, vegetation cover, structural diversity, and canopy shading. Buffer width, vegetation cover, and/or canopy shading may be reduced if atrazine exposure impacts plants in the riparian zone or prevents new growth from emerging. Plant species diversity and structural diversity may also be affected if only sensitive plants are impacted (Jobin et al., 1997; Kleijn and Snoeijing, 1997), leaving non-sensitive plants in place. Atrazine may also affect the long term health of high quality riparian habitats by affecting seed germination. Thus, if atrazine exposure impacted these riparian parameters, water quality within the action area for the listed mussels could be affected.

Because the majority of woody plants (i.e., shrubs and trees) are not sensitive to environmentally-relevant atrazine concentrations (MRID #46870400-01), effects on shading, streambank stabilization, and structural diversity (in terms of height classes) of woody forested vegetation are not expected. Effects are expected to be limited to herbaceous (non-woody) plants (e.g., grasses), which are not generally associated with shading.

The riparian health criteria described in Fleming et al. (2001; Table 5.13) and the characteristics associated with effective vegetative buffer strips suggest that healthier riparian zones would be less sensitive to the impacts of atrazine runoff than poorer riparian zones. Although riparian zones rich in species diversity and woody species may contain sensitive species, it is unlikely that they would consist of a high proportion of very sensitive plants. Wider buffers have more potential to reduce atrazine residues over a larger area, resulting in lower loading levels. According to Fleming et al. (2001), buffer distances of >18 m (approximately 60 feet) are characterized as "excellent" in supporting aquatic habitats. It should be noted that the label requirements for atrazine specify no use within 66 feet of intermittent and perennial streams. While this "buffer" area was established to decrease atrazine loading to waterbodies resulting from drift, if maintained with other good to excellect (Table 5.13) riparian habitat attributes, it is likely to reduce atrazine runoff to adjacent waterbodies. In addition, trees and woody plants in a healthy riparian area act to filter spray drift (Koch et al., 2003) and push spray drift plumes over the riparian zone (Davis et al., 1994), thus reducing exposure to lower height classes of plants (i.e., grassy and non-woody vegetation), which tend to be more sensitive. Therefore, higher quality riparian zones are expected to be less sensitive to atrazine than riparian zones that are narrow, low in species diversity, and comprised of young herbaceous plants or unvegetated areas. The available data suggest that riparian zones comprised of herbaceous plants and grasses would likely be most sensitive to atrazine effects, while woody vegetation within forested riparian zones would be tolerant of exposure to atrazine. Bare ground riparian areas and areas with sparse vegetation could also be adversely affected by prevention of new growth of grass, which can be an important component of riparian vegetation for maintaining water quality.

Based on the low sensitivity of forested areas containing woody shrubs and trees to atrazine, it is unlikely that atrazine will adversely affect these types of riparian vegetation adjacent to use sites and watersheds within the action area of the listed mussels.

Potential for Atrazine to Indirectly Affect the Listed Mussels via Effects on Riparian Vegetation

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

Quantification of risk to the listed mussels from potential effects to riparian areas is precluded by the following factors:

- The relationship between distance of soil input into the watershed and sediment deposition in areas critical to survival, reproduction, and growth of the listed mussels is not known;
- Riparian areas within the action area are highly variable in their composition and location with respect to atrazine use; therefore, their sensitivity to potential damage is also variable; and
- The action area for the listed mussels, specifically the fat pocketbook, is a large geographic area, encompassing 8 states.

In addition, even if plant community structure was quantifiably correlated with riparian function, it may not be possible to discern the effects of atrazine on species composition separate from other agricultural actions or determine if atrazine is a factor in altering community structure. Plant community composition in agricultural field margins is likely to be modified by many agricultural management practices. Vehicular impact and mowing of field margins and off-target movement of fertilizer and herbicides are all likely to cause changes in plant community structure of riparian areas adjacent to agricultural fields (Jobin et al., 1997; Kleijn and Snoeijing, 1997; Schippers and Joenje, 2002). Although herbicides are commonly identified as a contributing factor to changes in plant communities adjacent to agricultural fields, some studies identify fertilizer use as the most important factor affecting plant community structure near agricultural fields (e.g. Schippers and Joenje, 2002) and community structure is expected to be affected by a number of other factors (de Blois et al., 2002). Specifically, the alteration and destruction of stream habitat due to impoundments for flood control, navigation, hydroelectric power, and recreation are critical factors that may impact water quality for the listed mussels within the defined action area (USFWS 1989, 1992, 1994). Thus, the effect of atrazine alone on riparian community structure is complicated by other multiple stressors likely to occur within the action area for the listed mussels. Although the data do not allow for a quantitative estimation of risk from potential riparian habitat alteration, a qualitative discussion is presented below.

In summary, terrestrial plant RQs are above LOCs for all uses; therefore, riparian vegetation may be affected by use of atrazine. As previously discussed, the potential for atrazine to affect the listed mussels via impacts on riparian vegetation depends primarily on the extent of potentially sensitive (herbaceous and grassy) riparian areas and their impact on water quality in the streams and rivers where the listed mussels are known to occur. Because woody plants are generally not sensitive to atrazine at expected exposure concentrations, riparian areas which have predominantly forested vegetation containing woody shrubs and trees are not likely to be impacted by atrazine use. Therefore, atrazine is not likely to adversely affect populations of listed mussels in watersheds with predominantly forested riparian areas. Conversely, atrazine may affect grassy and herbaceous riparian vegetation, resulting in increased sedimentation which could impact the listed mussels in ways previously described.

Further evaluation of the potential for atrazine to indirectly impact the listed mussels via potential effects to riparian vegetation was completed for the three listed mussels. This evaluation, which is included in Appendix I, was based on an analysis of land cover/use data and the type of riparian vegetation (i.e., grassy versus forested) adjacent to occupied streams for PCPP mussel, and northern riffleshell. As previously mentioned, the action area for the fat pocketbook mussel is a large area, encompassing eight states. Therefore, spatial analysis of land cover data and type of riparian vegetation adjacent to occupied streams for the fat pocketbook was conducted for a number of example watersheds, intended to encompass the range of larger rivers (and surrounding land cover types) that this species inhabits. These watersheds include the Big Sunflower River in Mississippi, the Wabash River in Illinois, the White River and Lower Ohio River in Indiana, the Upper Ohio River in Kentucky, and the St. Francis and White Rivers in Arkansas. It should be noted, however, that the fat pocketbook also inhabits other smaller streams and chutes, for which no land cover data is available. In these areas, the effects determination for indirect effects to the fat pocketbook mussels via direct atrazine effects on riparian vegetation is dependant on the presence of forested (woody shrubs and trees) versus herbaceous (grassy and non-woody) riparian vegetation adjacent to the streams and rivers within the fat pocketbook mussel's action area. For areas where the riparian habitat is predominantly forested with shrubs and trees, the effects determination for the fat pocketbook in small streams and chutes is "may affect, but not likely to adversely affect" or "NLAA". This finding is based on insignificance of effects (i.e., the effect cannot be meaningfully measured, detected or evaluated in the context of a level of effects where "take" occurs for a single fat pocketbook mussel). For watersheds of the fat pocketbook mussels that are in close proximity to potential atrazine use sites and where the riparian vegetation is comprised of grasses and non-woody plants, the effects determination is "may affect and likely to adversely affect" or "LAA". In addition, the extent of specific land management practices, which may result in reduced sedimentation to occupied watersheds, is unknown. Until further analysis on specific land management practices and sensitivity of riparian vegetation adjacent to fat pocketbook mussel habitat in smaller streams and chutes is completed, potential effects to riparian vegetation are presumed to potentially adversely affect the fat pocketbook in these watersheds. A graphic representation of the effects determination for the fat pocketbook mussel located in small streams and chutes, based on evaluation of the sedimentation, streambank stability, and thermal stability attributes for riparian vegetation is provided in Figure 5.1.

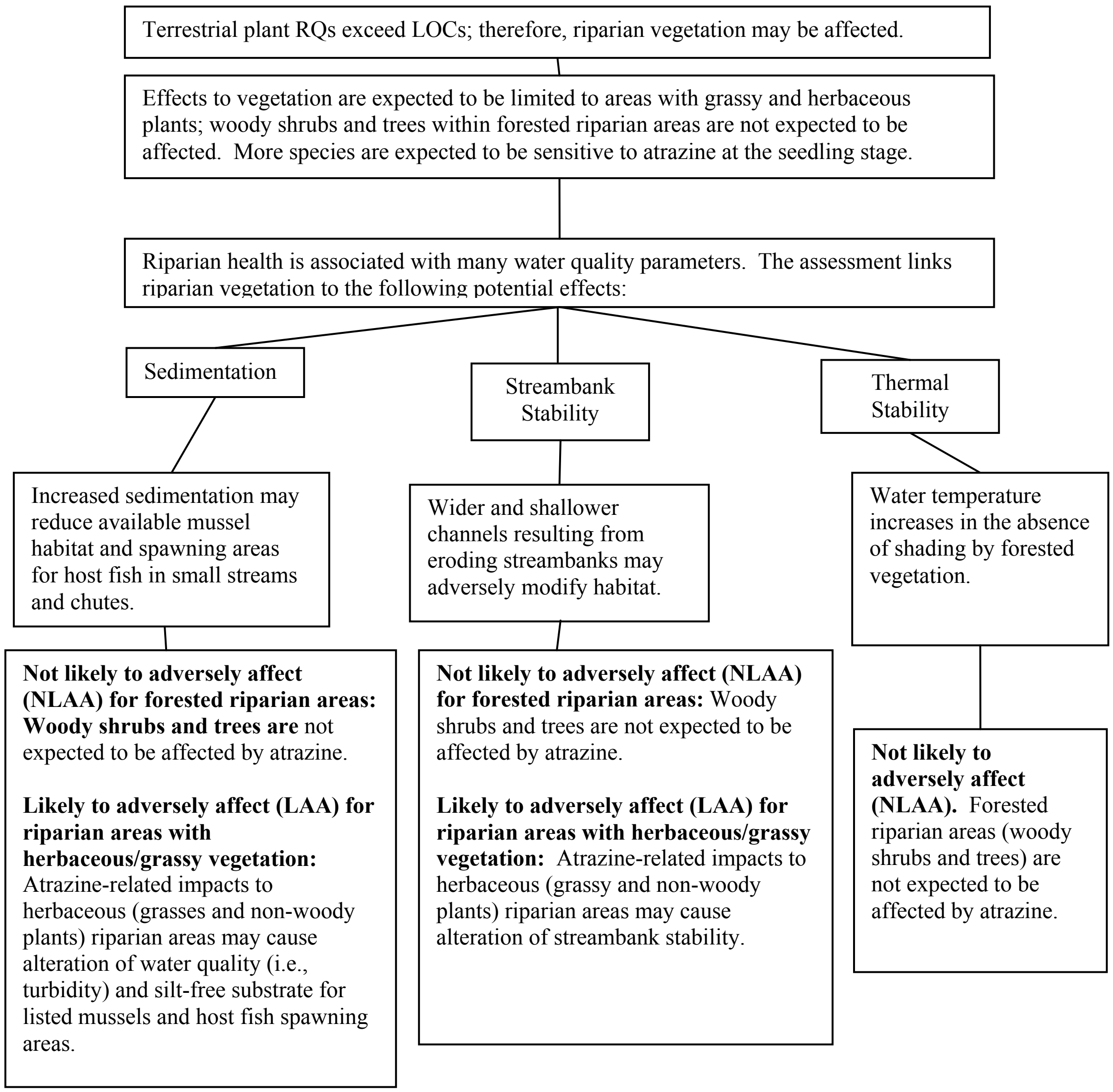

Figure 5.1 Summary of the Potential of Atrazine to Affect the Fat Pocketbook Mussel via Riparian Habitat Effects in Small Streams and Chutes

The results of the spatial analysis of occupied big rivers for the fat pocketbook mussel (i.e., Big Sunflower River in Mississippi, the Wabash River in Illinois, the White River and Lower Ohio River in Indiana, the Upper Ohio River in Kentucky, and the St. Francis and White Rivers in Arkansas) show that very little, if any, sensitive herbaceous riparian vegetation or barren land is present in the riparian area adjacent to these watersheds. The majority of land cover directly adjacent to these types of occupied watersheds appears to be either cultivated crop, forested vegetation, or woody wetlands. Given the lack of sensitive herbaceous vegetation and barren land, atrazine-related impacts to riparian vegetation adjacent to these larger rivers are not expected. Although it is possible that the fat pocketbook may occupy similar watersheds where the percentage of herbaceous land cover surrounding the watershed is higher than that observed in the seven example watersheds, the available land cover data for all three listed species indicates the majority of riparian vegetation directly adjacent to occupied rivers is comprised of deciduous forest and woody wetlands that are not sensitive to atrazine at environmentally relevant concentrations. Therefore, potential indirect effects via atrazine-related impacts to riparian areas adjacent to large rivers occupied by the fat pocketbook are expected to be insignificant (i.e., cannot be meaningfully measured, detected or evaluated in the context of a level of effects where "take" occurs for a single fat pocketbook), and the resulting effects determination is "may affect, but not likely to adversely affect" or "NLAA".

Based on the spatially-explicit evaluation of potential impacts to the PCPP mussel and northern riffleshell, effects to riparian vegetation adjacent to occupied watersheds are not expected (see Appendix I). This evaluation is based on combination of land cover and county-specific land use data, including the type of riparian vegetation (i.e., grassy versus forested) adjacent to occupied streams, and land cover and aerial imagery. Land cover and land use data (as well as aerial satellite imagery) surrounding the occupied streams/rivers of the PCPP mussel and northern riffleshell suggest that the predominant riparian area adjacent to occupied watersheds is not likely to be sensitive to atrazine and/or riparian vegetation exposure to atrazine is expected to be minimal. Potential indirect effects via atrazine-related impacts to riparian areas adjacent to occupied streams/rivers are expected to be insignificant, such that they cannot be meaningfully measured, detected or evaluated in the context of a level of effects where "take" occurs for a single PCPP mussel or northern riffleshell. Therefore, atrazine is not likely to adversely affect the PCPP mussel and northern riffleshell in occupied streams/rivers via effects to riparian vegetation, and the resulting effects determination for the two species is "may affect, but not likely to adversely affect" or "NLAA".

6. Uncertainties

6.1 Exposure Assessment Uncertainties

6.1.1 Uncertainties in the Aquatic Exposure Assessment

While peak exposures in available monitoring data are within a factor of two of modeling, longer term concentrations (e.g. 30-day averages) are generally higher in screening-level modeling than in monitoring data. Conversely, refined modeling using flow through the Index Reservoir water body (typically used for drinking water assessments) are similar when comparing peak concentrations, but are lower than the longer term concentrations seen in a subset of monitoring sites in the most vulnerable watersheds. However, the majority of atrazine concentrations from monitored sites that are greater than modeled EECs are within 2 to 3 times of the refined flow-adjusted modeled EECs. Viewed in the context of exposure for all atrazine use areas, the refined modeling is likely to represent a reasonable approximation of high end atrazine exposure.

The primary factor that may result in over-estimation of exposure in the screening-level modeling is the assumption of no flow in the modeled water body. Factors that may account for under-estimation of exposure in the refined modeling relative to the most vulnerable watersheds may include differences between reservoir volume, watershed size, and flow dynamics relative to stream characteristics, as well as differences in the flow rates used in the refined modeling (taken from occupied streams generally at 4th order and higher) compared to flow rates in the 2nd and 3rd order streams represented by most of the vulnerable watershed sites. Furthermore, the impact of setbacks on runoff estimates has not been quantified, although well-vegetated setbacks are likely to result in a reduction in runoff loading of atrazine.

Overall, analysis indicates that increasing flow will result in reduction of exposure relative to screening level model estimates, particularly for longer-term durations of exposure (14-day, 30-day, etc.).

6.1.2 Modeling Assumptions

Overall, the uncertainties addressed in this assessment cannot be quantitatively characterized. Given the available data and use of conservative modeling assumptions, it is expected that the screening-level modeled EECs over-predict exposure for longer-term durations, but are within a factor of two as compared with peak monitored concentrations. However, refined flow-adjusted EECs are likely to be conservative for all but a subset of watersheds most vulnerable to atrazine runoff.

In general, the simplifying assumptions used in this assessment appear from the characterization in Section 3.2.7 to be reasonable given the analysis completed and the available monitoring data. There are also a number of assumptions that tend to result in over-estimation of exposure. Although these assumptions cannot be quantified, they are qualitatively described. For instance, modeling in this assessment for each atrazine use assumes that all applications have occurred concurrently on the same day at the exact same application rate. This is unlikely to occur in reality, but is a reasonable conservative assumption in lieu of actual data.

6.1.3 Comparison of Modeling and Monitoring Data

A number of factors add uncertainty to the direct comparison of flow-adjusted modeling EECs with the monitoring data (including other sources discussed previously). For example, the selection process for the AEMP sites was focused on the most vulnerable sites relative to atrazine runoff, and as seen in Figure 3.10, do not directly correlate with the majority of streams that are occupied by the three listed mussels. The AEMP sites represent highly vulnerable 2nd and 3rd order streams (by the Strahler system) and are considered to be representative of lower flow watersheds (< 200 ft^3/sec) that co-occur within the 1,172 watersheds. Therefore, the AEMP data are directly representative of exposures expected in lower flow streams where the fat pocketbook and northern riffleshell live within the boundary of vulnerable watersheds. However, occupied areas outside the range of the 1,172 watersheds and higher flow streams/rivers (> 200 ft^3/sec) within the boundary of 1,172 vulnerable watersheds are not represented by these data. These types of occupied streams/rivers are best represented by modeled flow-adjusted EECs and non-targeting monitoring data. Flow-adjusted EECs used to characterize exposures are based on flow data from higher order streams with flow rates that are higher than those found in all of the ecological monitoring sites.

There are also uncertainties associated with modeling using the Index Reservoir water body (used principally for human health exposure assessments) because the water body volume of the Index Reservoir may not be representative of the larger rivers where the three listed mussel's lives. The Index Reservoir was developed to represent a small drinking water reservoir.

Additional uncertainties should be considered when comparing the modeled static water body EECs with various habitat types and monitoring data. Specifically, the modeled water body represents static water; however, in reality, many water bodies have some amount of flow. For the action area, it is expected that no-flow and low-flow water bodies are representative of the headwater streams adjacent to agricultural field. In general, it is expected that modeled atrazine concentrations in the static water body will over-estimate exposure in settings where flow is greater than those modeled and where the volume of the water body is greater than that modeled (20,000,000 liters). As demonstrated in the various comparisons between modeling and monitoring data described above, it is apparent that peak concentrations are well represented by modeling with both the static water body and flow-adjusted modeling using the Index Reservoir although some of the more vulnerable sites may be under-represented. However, longer-term concentrations (e.g. 14-, 30-, 60-, and 90-day averages) appear to be over-represented by modeling with the static water body, while these same duration-exposure concentrations may be under-represented by flow-adjusted modeling in the most vulnerable watersheds with low flow rates.

6.1.4 Impact of Vegetative Setbacks on Runoff

Unlike spray drift, models are currently not available to evaluate the effectiveness of a vegetative setback on runoff and loadings. The effectiveness of vegetative setbacks is highly dependent on the condition of the vegetative strip. For example, a well-established, healthy vegetative setback can be a very effective means of reducing runoff and erosion from agricultural fields (USDA, NRCS, 2000). Alternatively, a setback of poor vegetative quality or a setback that is channelized can be ineffective at reducing loadings. Until such time as a quantitative method to estimate the effect of vegetative setbacks on various conditions on pesticide loadings becomes available, the aquatic exposure predictions are likely to overestimate exposure where healthy vegetative setbacks exist and underestimate exposure where poorly developed, channelized, or bare setbacks exist.

6.1.5 PRZM Modeling Inputs and Predicted Aquatic Concentrations

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

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

Additionally, the rate at which atrazine is applied and the percent of crops that are actually treated with atrazine may be lower than the Agency's default assumption of the maximum allowable application rate being used and the entire crop being treated. The geometry of a watershed and limited meteorological data sets also add to the uncertainty of estimated aquatic concentrations.

6.2 Effects Assessment Uncertainties

6.2.1 Age Class and Sensitivity of Effects Thresholds

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

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

6.2.2 Use of Acute Freshwater Invertebrate Toxicity Data for the Midge

The initial acute risk estimate for freshwater invertebrates was based on the lowest toxicity value from *Chironomus* studies, which showed a wide range of sensitivity within and between species of the same genus (2 orders of magnitude). Further evaluation of the species sensitivity distribution shows that the majority of freshwater invertebrate species are unaffected by atrazine at environmentally relevant concentrations. Therefore, screening-level acute RQs based on the most sensitive toxicity endpoint for freshwater invertebrates may represent an overestimation of potential indirect effects to the listed mussels via direct effects on freshwater invertebrates as dietary food items.

6.2.3 Impact of Multiple Stressors on the Effects Determination

The influence of length of exposure and concurrent environmental stressors to the listed mussels (i.e., construction of dams and locks, fragmentation of habitat, change in flow regimes, increased sedimentation, degradation of quantity and quality of water in the watersheds of the action area, predators, etc.) will likely affect the species' response to atrazine. Additional environmental stressors may increase the listed mussel's sensitivity to the herbicide, and there is the possibility of additive/synergistic reactions. Timing, peak concentration, and duration of exposure are critical in terms of evaluating effects, and these factors are expected to vary both temporally and spatially within the action area. Overall, the effect of this variability may result in either an overestimation or underestimation of risk. However, as previously discussed, the Agency's LOCs are set to be protective given the wide range of possible uncertainties.

6.2.4 Use of Threshold Concentrations for Community-Level Endpoints

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

The threshold concentrations are intended to be predictive of potential atrazine-related community-level effects in aquatic ecosystems, such as those that occur in known locations for the listed mussels, where the species composition may differ from those included in the micro- and mesocosm studies. Although it is not possible to determine how well the responses observed in the micro- and mesocosm studies reflect the action area watersheds for the listed mussels, estimated chronic atrazine exposure concentrations in less vulnerable watersheds of the action area (from modeled EECs assuming flow) are predicted to be between 5 to 12 times lower than the community-level threshold concentrations, depending on the modeled atrazine use and averaging period. However, an evaluation of targeted monitoring data from vulnerable watersheds suggests that chronic exposure concentrations of atrazine exceed these threshold concentrations in a small number of watersheds ranging from approximately 5 to 12.5 percent of the total. Given that threshold concentrations were derived based on the best available information from available community-level data for atrazine, these values are intended to be protective of the aquatic community, including the listed mussels. Additional uncertainties associated with use of the thresholds to estimate community-level effects are discussed in Appendix B (Section B.8) of the previous atrazine endangered species assessment for eight listed mussels (U.S. EPA, 2007c).

6.2.5. Sublethal Effects

The assessment endpoints used in ecological risk assessment include potential effects on survival, growth, and reproduction of the assessed mussels and organisms on which mussels depend for survival such as fish. A number of studies were located that evaluated potential sublethal effects to fish from exposure to atrazine. Although many of these studies reported toxicity values that were less sensitive than the submitted studies, they were not considered for use in risk estimation. In particular, fish studies were located in the open literature that reported effects on endpoints other than survival, growth, or reproduction at concentrations that were considerably lower than the most sensitive endpoint from submitted studies.

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

Reported sublethal effects including changes in hormone levels, behavioral effects, neurophysiological responses, kidney pathology, gill physiology, and potential olfaction effects have been observed at concentrations lower than 65 μg/L (see Appendix A and Section 4.1.2.). In accordance with the Overview Document (U.S. EPA, 2004) and the Services Evaluation Memorandum (USFWS/NMFS, 2003), these studies were not considered appropriate for risk estimation in place of the life cycle studies because quantitative relationships between these effects and the ability of fish to survive, grow, and reproduce has not been established. The magnitude of the reported sublethal effect associated with reduced survival or reproduction has not been established; therefore it is not possible to quantitatively link sublethal effects to the selected assessment endpoints for this endangered species risk assessment. In addition, in the fish life cycle studies, no effects were observed to survival, reproduction, and/or growth at levels associated with the sublethal effects. Also, there were limitations to the studies that reported sublethal effects that preclude their quantitative use in risk assessment (see Appendix A and Section 4.2.1). Nonetheless, if future studies establish a quantitative link between the reported sublethal effects and fish survival, growth, or reproduction, the conclusions with respect to potential effects to host fish may need to be revisited.

6.2.6. Exposure to Pesticide Mixtures

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

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

6.3 Assumptions Associated with the Acute LOCs

The risk characterization section of this endangered species assessment includes an evaluation of the potential for individual effects. The individual effects probability associated with the acute RQ is based on the mean estimate of the slope and an assumption of a probit dose response relationship for the effects study corresponding to the taxonomic group for which the LOCs are exceeded.

Sufficient dose-response information was not available to estimate the probability of an individual effect on the midge (one of the dietary food items of the host fish). Acute ecotoxicity data from the midge were used to derive RQs for freshwater invertebrates. Based on a lack of dose-response information for the midge, the probability of an individual effect was calculated using the only probit dose response curve slope value reported in available freshwater invertebrate ecotoxicity data for technical grade atrazine.

Therefore, a probit slope value of 4.4 for the amphipod was used to estimate the probability of an individual effect on the freshwater invertebrates. It is unclear whether the probability of an individual effect for freshwater invertebrates other than amphipods would be higher or lower, given a lack of dose-response information for other freshwater invertebrate species. However, the assumed probit dose response slope for freshwater invertebrates of 4.4 would have to decrease to approximately 1 to 2 to cause an effect probability ranging between 1 in 10 and 1 in 100, respectively, for freshwater invertebrates.

6.4. Uncertainty in the Potential Effect to Riparian Vegetation vs. Water Quality Impacts via Increased Sedimentation

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

7. Summary of Direct and Indirect Effects to the Listed Mussels

In fulfilling its obligations under Section 7(a)(2) of the Endangered Species Act, the information presented in this endangered species risk assessment represents the best data currently available to assess the potential risks of atrazine to the fat pocketbook, PCPP mussel and northern riffleshell. A summary of the risk conclusions and effects determination for the three listed mussels, given the uncertainties discussed in Section 6, by assessment endpoint, is presented in Tables 7.1. Table 7.2 provides a summary of the direct and indirect effects determinations for each of the three assessed listed mussels.

<table>
<tr><th colspan="5">Table 7.2 Effects Determination Summary for the Assessed Listed Mussels (by Assessment Endpoint)</th></tr>
<tr><th colspan="5">Direct and Indirect Effects to Listed Mussels</th></tr>
<tr><th rowspan="2">Assessment Endpoints for Aquatic Animals and Plants</th><th colspan="2">Effects Determination and Basis for PCPP Mussel (in all occupied streams) and Fat Pocketbook and Northern Riffleshell Mussels (located in less vulnerable watersheds and larger river/streams with flow > 200 ft^3/sec in vulnerable watersheds)</th><th colspan="2">Effects Determination and Basis for Fat Pocketbook and Northern Riffleshell (located in highly vulnerable watersheds with stream flow < 200 ft^3/sec or for which no flow data is available)</th></tr>
<tr><th>Effects Determination[a]</th><th>Basis</th><th>Effects Determination[a]</th><th>Basis</th></tr>
<tr><td rowspan="2">1. Survival, growth, and reproduction of assessed mussel individuals via direct acute or chronic effects</td><td>Acute direct effects: NE</td><td>No acute LOCs are exceeded.</td><td>Acute direct effects: NE</td><td>No acute LOCs are exceeded.</td></tr>
<tr><td>Chronic direct effects: NLAA</td><td>Chronic LOCs are exceeded based on screening-level EECs; however, RQs based on flow-adjusted EECs and non-targeted monitoring data are less than concentrations shown to cause adverse effects in freshwater mollusks. This finding is based on discountable effects (i.e., chronic effects at refined levels of exposure are not likely to occur and/or result in “take” of a single listed mussel).</td><td>Chronic direct effects: NLAA</td><td>Chronic LOCs are exceeded based on screening-level EECs; however detected concentrations of atrazine in monitoring data from vulnerable watersheds are less than those shown to cause adverse effects in freshwater mollusks. This finding is based on discountable effects (i.e., chronic effects to atrazine at refined levels of exposure are not likely to result in “take” of a single fat pocketbook and northern riffleshell mussel located in highly vulnerable watersheds).</td></tr>
<tr><td>2. Indirect effects to assessed mussel individuals via reduction in food items (i.e., freshwater phytoplankton and zooplankton)</td><td>Phytoplankton: NLAA</td><td>Individual aquatic plant species may be affected. However, refined 14-, 30-, 60- and 90-day EECs, which consider the impact of flow and non-targeted monitoring data, are less than the threshold concentrations representing community-level effects. This finding is based on insignificance of effects (i.e., community-level effects to aquatic plants cannot be meaningfully measured, detected, or evaluated in the context of a “take” of a single listed mussel via a reduction in food items).</td><td>Phytoplankton: LAA[b]</td><td>Individual aquatic plant species within vulnerable watersheds of the action area may be affected. 14-, 30-, 60-, and 90- day rolling averages, based on the AEMP data, exceed their respective threshold concentrations for 5 to 12.5% of the sampled vulnerable watersheds. Therefore, community-level effects are possible for phytoplankton, resulting in indirect effects to the food supply of the fat pocketbook and northern riffleshell mussels, within lower flow (< 200 ft^3/sec) vulnerable watersheds of the action area.</td></tr>
</table>

	Acute direct effects to zooplankton: NLAA	Acute LOCs are exceeded based on screening-level EECs and the most sensitive freshwater invertebrate toxicity data. Based on the refined analysis, which considered flow-adjusted EECs, non-targeted monitoring data, and effects data specific to zooplankton, acute effects to zooplankton are not likely to occur at refined levels of exposure. Effects are discountable because refined exposures are not likely to cause adverse effects to zooplankton and the probability of an individual effect to zooplankton is low (i.e., 0.03%). Effects are also insignificant because the level of effect at predicted levels of exposure is low (i.e., <2%) and zooplankton are not the primary food source for listed mussels. Therefore, "take" of a single listed mussel is not expected to occur).	Acute direct effects to zooplankton: NLAA	Acute LOCs are exceeded based on screening-level EECs and the most sensitive freshwater invertebrate toxicity data. Based on the refined analysis, which considered flow-adjusted EECs, non-targeted monitoring data, and effects data specific to zooplankton, acute effects to zooplankton are not likely to occur at refined levels of exposure. Effects are discountable because refined exposures are not likely to cause adverse effects to zooplankton and the probability of an individual effect to zooplankton is low (i.e., 0.03%). Effects are also insignificant because the level of effect at predicted levels of exposure is low (i.e., <2%) and zooplankton are not the primary food source for listed mussels. Therefore, "take" of a single listed fat pocketbook and northern riffleshell mussel is not expected to occur).
	Chronic direct effects to zooplankton: NLAA	Chronic LOCs are exceeded based on screening-level EECs and the most sensitive freshwater invertebrate toxicity data. However, all refined measures of exposure (21-day flow-adjusted EECs and non-targeted monitoring data) are well below levels of chronic effects in cladocerons. This finding is based on discountable effects (i.e., chronic effects to atrazine at refined levels of exposure are not likely to occur and/or result in a "take" of a single listed mussel via a reduction in zooplankton as food items).	Chronic direct effects to zooplankton: NLAA	Chronic LOCs are exceeded based on screening-level EECs and the most sensitive freshwater invertebrate toxicity data. However, 21-day rolling averages based on the ecological monitoring data are well below levels of chronic effects in cladocerons. This finding is based on discountable effects (i.e., chronic effects to atrazine in highly vulnerable watersheds are not likely to occur and/or result in a "take" of a single fat pocketbook and northern riffleshell mussel via a reduction in zooplankton as food items).

3. Indirect effects to assessed mussel individuals via reduction in host fish for mussel glochidia (i.e., larvae)	Acute direct effects to host fish: NE	No acute LOCs are exceeded.	Acute direct effects to host fish: NE	No acute LOCs are exceeded.
	Chronic direct effects to host fish: NLAA	Chronic LOCs are exceeded based on screening-level EECs; however refined flow-adjusted EECs and non-targeted monitoring data are not likely to result in adverse chronic effects to fish. This finding is based on discountable effects (i.e., chronic exposure to atrazine is not likely to result in "take" of a single listed mussel because direct chronic effects to host fish are unlikely to occur).	Chronic direct effects to host fish: NLAA	Chronic LOCs are exceeded based on screening-level EECs; however, detected concentrations of atrazine in monitoring data from vulnerable watersheds are not likely to result in adverse chronic effects to fish. This finding is based on discountable effects (i.e., chronic exposure to atrazine is not likely to result in "take" of a single fat pocketbook and northern riffleshell because direct chronic effects to host fish in vulnerable watersheds are unlikely to occur).
4. Indirect effects to assessed mussel individuals via direct effects to aquatic plants (i.e., reduction of habitat and/or primary productivity)	Direct effects to aquatic plants: NLAA	Individual aquatic plant species may be affected. However, flow-adjusted 14-, 30-, 60-, and 90-day EECs and similar durations of exposure based on non-targeted monitoring data, are less than the threshold concentrations representing community-level effects. This finding is based on insignificance of effects (i.e., community-level effects to aquatic plants cannot be meaningfully measured, detected, or evaluated in the context of a "take" of a single listed mussel via direct effects on habitat and primary productivity).	Direct effects to aquatic plants: LAA[b]	Individual aquatic plant species within vulnerable watersheds of the action area may be affected. 14-, 30-, 60-, and 90- day rolling averages based on the AEMP data from vulnerable watersheds exceed their respective threshold concentrations for a small percentage of the data set. Therefore, community-level effects are possible for phytoplankton, resulting in indirect effects to the fat pocketbook and northern riffleshell, via direct effects on habitat and primary productivity, within lower flow (< 200 ft^3/sec) vulnerable watersheds of the action area.
Assessment Endpoints for Terrestrial Plants	**Effects Determination[a]**	**Basis**	**Effects Determination[a]**	**Basis**
5a. Indirect effects to *fat pocketbook* individuals via reduction of terrestrial vegetation (i.e., riparian habitat)	Direct effects to forested riparian vegetation: NLAA	Riparian vegetation may be affected because terrestrial plant RQs are above LOCs. However, woody shrubs and trees are generally not sensitive to atrazine; therefore, listed mussels in watersheds with predominantly forested	Direct effects grassy/herbaceous riparian vegetation: LAA	Riparian vegetation may be affected because terrestrial plant RQs are above LOCs. The LAA effects determination for listed mussels that are in close proximity to grassy/herbaceous riparian areas is based on the sensitivity of herbaceous vegetation to atrazine. Until further analysis on specific land

required to maintain acceptable water quality and habitat[c]		riparian vegetation (i.e., woody shrubs and trees) are not likely to adversely affected. This finding is based on insignificance of effects (i.e., effects to forested riparian vegetation in the action area are not likely to result in "take" of a single listed mussel).		management practices and sensitivity of grassy riparian vegetation adjacent to fat pocketbook mussel habitat is completed, potential indirect effects via sedimentation are presumed to adversely affect the fat pocketbook.
	Indirect effects to fat pocketbook mussels that occur in big rivers: NLAA	Land cover data from seven example watersheds (i.e., big rivers including the Big Sunflower River in Mississippi, the Wabash River in Illinois, the White River and Lower Ohio River in Indiana, the Upper Ohio River in Kentucky, and the St. Francis and White Rivers in Arkansas) indicates that the majority of riparian vegetation directly adjacent to occupied rivers is comprised of deciduous forest and woody wetlands that are not sensitive to atrazine at environmentally relevant concentrations. Therefore, potential indirect effects via atrazine-related impacts to riparian areas adjacent to large rivers occupied by the fat pocketbook are expected to be insignificant (i.e., cannot be meaningfully measured, detected or evaluated in the context of a level of effects where "take" occurs for a single fat pocketbook).		
5b. Indirect effects to *PCPP mussel* and *northern riffleshell* individuals via reduction of terrestrial vegetation (i.e., riparian habitat) required to maintain acceptable water quality and habitat[d]	NLAA	Land cover and land use data (as well as aerial satellite imagery) surrounding the occupied streams/rivers of the PCPP mussel and northern riffleshell suggest that the predominant riparian area adjacent to occupied watersheds is not likely to be sensitive to atrazine and/or riparian vegetation exposure to atrazine is expected to be minimal. Therefore, potential indirect effects via atrazine-related impacts to riparian areas adjacent to occupied streams/rivers are expected to be insignificant, such that they cannot be meaningfully measured, detected or evaluated in the context of a level of effects where "take" occurs for a single PCPP mussel or northern riffleshell.		

[a] NE = "no effect"; NLAA ' "may affect, but not likely to adversely affect"; and LAA = "may affect and likely to adversely affect".

[b] Further analysis of the AEMP data is required to determine the representativeness of the data to other watersheds within vulnerable areas where the listed mussel species occur. If the analysis suggests that the AEMP data are representative of atrazine concentrations in vulnerable watersheds where the fat pocketbook and northern riffleshell mussels occur, the effects determination will remain as "LAA." However, if further analysis reveals that the monitoring data are not representative of atrazine concentrations in vulnerable watersheds where these listed mussels occur, the effects determination will be revised to "NLAA".

[c] The effects determinations for indirect effects to the fat pocketbook mussel based on direct impacts to riparian habitat is applicable to its entire action area including riparian areas adjacent to both vulnerable and less vulnerable watersheds. Separate effects determinations are based on the presence of forested or herbaceous/grassy riparian vegetation adjacent to the streams and rivers within the fat pocketbook mussel's action area. In addition, a separate effects determination for fat pocketbook mussels located in big rivers was made, based on available land cover data.

[d] Given the limited range of the PCPP mussel and northern riffleshell, an analysis of land cover and county-level use data was completed as part of the effects determination for this endpoint.

Table 7.2 Effects Determination Summary for Each of the Three Assessed Listed Mussels[a]										
Assessed Mussel Species	Direct Effects		Indirect Effects							
	Acute	Chronic	Food Items		Host Fish		Aquatic Habitat: community-level effects	Riparian Vegetation		Big Rivers[b]
			Phytoplankton	Zooplankton	Acute	Chronic		Herbaceous/ Grassy	Forested	
Fat Pocketbook	NE	NLAA	**LAA**[c]	NLAA	NE	NLAA	**LAA**[c]	**LAA**	NLAA	NLAA
Purple Cat's Paw Pearlymussel	NE	NLAA	NLAA	NLAA	NE	NLAA	NLAA	NLAA		
Northern Riffleshell	NE	NLAA	**LAA**[c]	NLAA	NE	NLAA	**LAA**[c]	NLAA		

[a] NE = "no effect"; NLAA ' "may affect, but not likely to adversely affect"; and LAA = "may affect and likely to adversely affect". See Table 1.1 for the basis of the effects determinations for each of the assessed mussel species.

[b] Big Rivers include the Big Sunflower River in Mississippi, the Wabash River in Illinois, the White River and Lower Ohio River in Indiana, the Upper Ohio River in Kentucky, the St. Francis and White Rivers in Arkansas, and other similarly sized watersheds where the fat pocketbook mussel occurs.

[c] This LAA determination applies to populations of the fat pocketbook and northern riffleshell that are located in highly vulnerable watersheds with stream flow < 200 ft^3/sec or for which no data are available. Further analysis of the AEMP data is required to determine the representativeness of the data to other watersheds within vulnerable areas where the fat pocketbook and northern riffleshell mussels occur. If the analysis suggests that the AEMP monitoring data are representative of atrazine concentrations in vulnerable watersheds where these listed mussels occur, the effects determination will remain as "LAA." However, if further analysis reveals that the AEMP monitoring data are not representative of atrazine concentrations in vulnerable watersheds where these listed mussels occur, the effects determination will be revised to "NLAA".

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