



Aquatic Toxicology Notes: Predicting the Fate and Effects of Aquatic and Ditchbank Herbicides¹

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INTRODUCTION

The potential for a given pesticide to cause adverse effects on aquatic organisms depends on 1) its inherent toxicity to the specific organism and 2) the organism's exposure to the compound in terms of concentration and duration (Table 1). The inherent toxicity is a specific relationship between the organism and the chemical, which cannot be changed (i.e. specific mode of action). On the other hand, exposures can be highly variable due to a variety of factors including application rates and techniques, chemical and physical properties of the chemical and environmental conditions at the time of application.

The purpose of this document is to introduce readers to: 1) factors affecting the exposure of aquatic organisms to aquatic herbicides, and 2) useful methods for estimating potential risks of acute mortality for non-target aquatic organisms. A series of companion EDIS documents containing herbicide-specific information is accessible at <http://edis.ifas.ufl.edu/>

TOPIC_AQUATIC_TOXICOLOGY. These

documents contain specific information concerning chemical properties, environmental fate, toxicity to non-target organisms, and references.

FACTORS INFLUENCING EXPOSURE OF NON-TARGET ORGANISMS TO AQUATIC HERBICIDES

The fate of a pesticide refers to what happens to it once it is released into the environment. To describe the fate of a given pesticide, the environment is usually thought of as having several compartments, including: soil, water, air, and biota (living creatures) (Figure 1). The fate of a given pesticide is important because it ultimately dictates the duration (e.g. 1 hour, 1 day, 1 week, etc.) and route (e.g. sediments, food, water column, etc.) of exposure for aquatic organisms. Many factors influence the fate of pesticides in the environment, and the likelihood that aquatic organisms may be exposed to them. Herbicide application rates are important because

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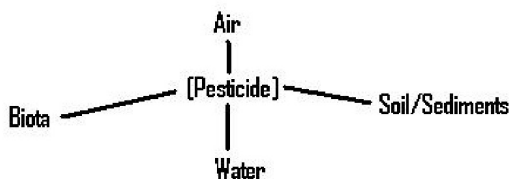
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All chemicals should be used in accordance with directions on the manufacturer's label. The use of trade names in this publication is solely for the purpose of providing specific information. UF/IFAS does not guarantee or warranty the products named, and references to them in this publication does not signify our approval to the exclusion of other products of suitable composition.

Table 1. Factors affecting potential toxicity of an aquatic herbicide.

Factor	Description
Mode of Action	Specific biochemical pathway that is impacted by the pesticide, resulting in negative effects such as death, reduced reproduction, reduced health, or some other negative impact.
Concentration	The actual amount of pesticide in the water column and/or sediments to which the organisms are exposed.
Exposure Duration	The amount of time the organism is exposed to the pesticide. The probability of toxic effects generally increases as exposure duration increases if a mode of action is present.

they dictate the total amount of active ingredient that organisms may be exposed to. In addition, the solubility and partitioning behavior of the herbicide will determine whether organisms in the water column or sediments will likely be exposed, and the persistence of the herbicide will influence the potential exposure period. These factors are discussed in the following sections.

**Figure 1.** Distribution of a pesticide between the different environmental compartments.

Water Solubility

Water solubility refers to the amount of the pesticide that will completely dissolve in a given volume of water. When a solid or liquid completely dissolves in water, the resulting liquid mixture is clear with no suspended materials present. This is one of the most important properties influencing the presence of a pesticide in the water column. The typical units for expressing solubility are:

- g/L (~ parts per thousand)
- mg/L (~ parts per million)
- µg/L (~ parts per billion)

Note equivalents,

- 1 g/L = 1,000 mg/L = 1,000,000 µg/L
- 1 mg/L = 0.001 g/L = 1000 µg/L
- 1 µg/L = 0.001 mg/L = 0.000001 g/L

Because solubility is affected by temperature, solubility values are usually reported with the temperature at which they were measured. Pesticides having large water solubility values dissolve to a greater extent relative to those with smaller solubility values. For instance, a pesticide with a water solubility value of 33,000 mg/L at 80° F is 1000 times more soluble than one with a solubility of 33 mg/L at 80° F.

Partitioning Behavior

Once the herbicide is applied to the water or to surrounding ditch banks, it will preferentially move or transfer into one or more of the environmental compartments depending on the physical and chemical properties of the herbicide and the environment. By definition, partition refers to the act or process of distributing something into different phases or compartments. Two major partitioning routes influencing the fate of pesticides are 1) partitioning between soil/sediments and water, and 2) partitioning between soil/sediments/water and air. These concepts are discussed below.

Soil-Water Partitioning

When an herbicide is applied directly to the soil (ditchbanks) or water, some of it will preferentially sorb (stick) to soil particles, particularly organic matter (OM), and some will dissolve and mix with the water. The process of sorption greatly influences the partitioning of some chemicals into the soil and sediment environmental compartments. Sorption is a

general term referring to the physical associations of an herbicide with the surface (adsorption) and the interior (absorption) of solid matrices. The sorptive properties of herbicides vary greatly. Those that are less strongly sorbed may be slowly released back into the water column through the process called desorption. Sorption can also influence the susceptibility of the herbicides to degradation processes in the environment by removing them from or attracting them to active degradation zones. The partitioning coefficient (K_d) is an indicator of the sorptive properties of a pesticide (Figure 2). This value is calculated from experiments where a soil and water is mixed into a slurry along with the pesticide. Once equilibrium is achieved, the concentration of pesticide associated with the solids and dissolved fractions are measured. The K_d is then calculated as,

$$[\text{sorbed pesticide concentration}] / [\text{pesticide concentration in solution}]$$

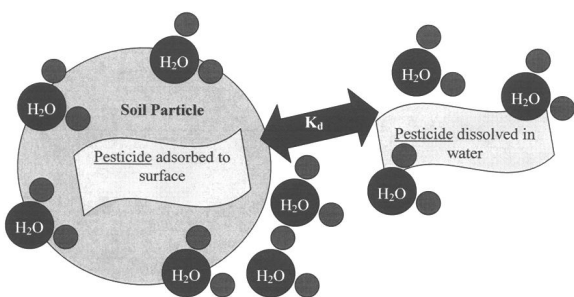


Figure 2. The partitioning coefficient (K_d) estimates the relative distribution of a pesticide between the sorbed and dissolved states.

K_d values for a given herbicide, especially nonpolar compounds, can be used to qualitatively predict whether it will preferentially partition into the water column or sediments and soil (ditchbanks) using the following criteria:

- $K_d = 1$; Equally distributed between solid and water

- $K_d > 1$; More than 50% likely present in sorbed states
- $K_d \gg 1$; Most present is highly sorbed, very little in soluble state (water)
- $K_d < 1$; More than 50% likely present in soluble state (water column)
- $K_d \ll 1$; Most present in highly soluble state, very little sorbed

For nonpolar pesticides, sorption is primarily related to associations with organic materials in the soil and sediments. Because of the importance of organic carbon, the Organic Carbon partitioning coefficient (K_{oc}) is more commonly reported. The K_{oc} is derived as,

$$K_d / F_{oc} (\% \text{ organic carbon in soil})$$

If one has the K_{oc} for one soil-pesticide combination (reported in literature), the K_d for other soil types can be estimated by multiplying the K_{oc} by the fraction (%) of soil organic carbon ($K_{oc} \times F_{oc}$). For a given nonpolar pesticide, sorption increases as the soil/sediment organic carbon content increases. Thus, pesticide leaching in ditch bank soils with high OM is expected to be slower compared to soils low in OM due to increased sorption. Pesticides with a K_{oc} or $K_d = 0$ do not sorb to soil/sediment, and would likely move with the water in a totally dissolved state.

While sediments and soils are similar in many ways, it is important to note that pesticides may interact with each in very different fashions. Possibly contributing to the different behaviors is the increased amorphous material and organic matter content found in many sediments as opposed to soils.

The use of K_{oc} for estimating sorptive potential is appropriate for nonpolar pesticides, and assumes that all partitioning is influenced primarily by hydrophobic (water-hating) interactions with insoluble organic materials. This may not be the case with ionizable pesticides. Ionizable pesticides are those in which the uncharged, neutral form is capable of gaining or losing protons, resulting in a change into cationic (positively charged) or anionic (negatively charged) forms relative to pH. Pesticides capable of ionization are typically indicated by the

presence of pK_a or pK_b values. When not in the neutral state, pesticides having a pK_a will be anionic; while those with a pK_b will be cationic. A summary of the expected environmental behavior of ionizable pesticides is shown in Table 2. Consult Wauchope et al. (1992) for a more thorough discussion.

Table 2. Summary of the expected environmental behavior of ionizable pesticides based on ionization constants. Table taken from Wauchope et al. (1992).

pK_a/pK_b Value	Dominant form ¹	Environmental Behavior
$pK_a < 3$	anion	Highly mobile in soil unless chemical complex formed; less mobile under very acidic conditions; very soluble; nonvolatile.
$pK_a > 10$	neutral	Behaves like nonionic material except at extremely high pH; less mobile than anion; probably much less soluble than anion; volatilization possible.
pK_a 3-10	mixture of neutral and anions depending on pH	If pH is near the pK_a , mobility, solubility and volatility will be sensitive to pH.
$pK_b < 4$	cation	Immobile (clay surface sorption); very soluble; extreme soil sorption leads to long half-life but little biological activity; nonvolatile.
pK_b 4-11	mixture of neutral and anions depending on pH	If pH is near pK_b , mobility, solubility, and volatility will be sensitive to pH.
$pK_b > 11$	neutral	Behaves like nonionic material except at extremely low pH; much more mobile, and less soluble than cation; volatilization possible.

¹ Assumes soil pH ranges from 5-8.

Sorption mechanisms for these pesticides include:

1. binding of cations (positively charged herbicides) to negatively charged sites on clay surfaces (very strong interaction)
2. binding of anions (negatively-charged herbicides) to soil anion-exchange sites (a very weak interaction)
3. chemical specific binding mechanisms.

Soil/Water-Air Partitioning

One other partitioning behavior affecting exposure of nontarget organisms is between the solid or liquid state and air. This behavior is primarily influenced by the vapor pressure or volatility of the pesticide. Volatility is the tendency for a liquid or a solid to change into a gas. It describes how quickly a liquid will evaporate when it is in contact with air. Highly volatile chemicals are easily lost to the atmosphere. Chemical volatility is generally inversely related to water solubility (i.e. highly water soluble compounds are not very volatile and vice versa).

Environmental Persistence

The persistence of the herbicide in the environment refers to how long it remains in a biologically active or inactive (for degradation products) form. The typical measure of persistence is called the half-life ($T_{1/2}$), or the amount of time needed for half of the chemical to disappear. The $T_{1/2}$ of pesticides is highly variable. In fact, the same pesticide may have a very different $T_{1/2}$ in various environments. Persistence increases as the $T_{1/2}$ increases, and is highly influenced by the susceptibility of the herbicide to a variety of degradative processes such as: photodecomposition, microbial decomposition, and chemical degradation. These are discussed below.

Photodecomposition

Photodecomposition reactions account for the degradation of many herbicides in both aquatic and terrestrial environments. These reactions require absorption of light energy by the herbicide molecule in order to progress. Substances that absorb UV light in the spectral region of sunlight (Wavelengths >290 nm) may either (1) undergo direct photolysis, or (2)

undergo indirect photolysis in which other constituents in the water absorb the light energy, which is either transmitted to the herbicide (sensitization) or leads to the formation of reactive species that enter into chemical reactions. Water turbidity (the presence of suspended sediments and foreign particles) directly affects light penetration into the surface water, and indirectly affects light absorption by the herbicides. Thus, the half-life for an herbicide that is normally readily photodegraded may be extended when applied in conditions where light penetration into the water column is limited. This increased half-life may also extend the exposure time for aquatic organisms.

Microbial Decomposition

The uppermost sediment layer in a surface water body is the most biologically active area, and encompasses the primary reaction zone for sorptive and microbially-mediated degradative processes. Microbial populations within the uppermost strata may be 10 times greater than in lower strata (i.e., 1 meter). As a result of microbial respiration, dissolved oxygen concentrations decrease causing changes in localized water chemistry. These changes in chemistry favor the accumulation of organic matter, which provides additional adsorbent and an energy source for microbial respiration and possible degradation of some pesticides.

Water Chemistry

Water pH can affect the adsorption of some pesticides to sediments, especially those that are ionizable (have a pK value). Lake water sometimes possesses higher pH values than the incoming water. As a result, weakly basic pesticides such as substituted ureas (i.e. diuron), and acidic herbicides (i.e. 2,4-D) may desorb from sediment/soil complexes. Often, pH values approaching the pK of specific compounds results in maximum adsorption to organic matter. Changes in pH may also result in increased desorption of pesticides from sediments and in some cases pH has

no effect on adsorption/desorption phenomena. Basic compounds are sorbed most strongly when the surface acidity of the soil/sediment particle is 1 to 2 pH units lower than that of the lowest pK value of the molecule. In addition to its influence on sorption, pH also stimulates hydrolysis of some pesticides. Hydrolysis refers to the breakdown of the active ingredient by reactions with water molecules.

Limnology

Pond characteristics such as water body size, depth, thermal stratification, and lake age can have important effects on mixing, water chemistry, and sediment distribution within the water body. The effects on mixing are especially important during herbicide applications. If the water is not well mixed, herbicide concentrations may be much higher in the uppermost strata, possibly intensifying toxicity to target and non-target organisms. Stratification within ponds may also change the water chemistry due to the lack of oxygen exchange to bottom areas. The resulting conditions may increase or decrease degradation of specific herbicides. Sediment distribution effects are especially important relative to the input of pesticide-containing soil eroded from ditchbanks and other areas. In this case, the sorbed complexes segregate on a particle size basis with the largest sized particles remaining close to the runoff inflow points and the smaller sized particles moving progressively farther from the inflow point based on decreasing density. Clay distribution is usually more uniform in small ponds that are subjected to large inflows of runoff and no stratification. These clay particles eventually settle to the bottom and distribute vertically according to size.

AQUATIC TOXICOLOGY

Basic Principles

Aquatic toxicology can be defined as the study of the effects of potentially toxic chemicals on aquatic organisms, with special emphasis on the harmful effects. Historically, this discipline has used toxicity tests to identify the harmful effects. Standard tests

evaluate dose-response relationships (toxicity at different concentrations) and mechanisms of action in a variety of organisms that are representative of different ecosystem niches. These tests may evaluate the response of individuals or populations to varying concentrations of the chemical. The dose-response relationship is based on the following three assumptions:

1. The response (toxicity) is due to the chemical administered
2. The magnitude of the response (toxicity) is related to the dose
3. There exists both a quantifiable method for measuring and a precise means of expressing toxicity.

Types of Effects

Effects may be of such minor significance that the organism can function normally. However, under stressful conditions (i.e., pH change, low dissolved oxygen, high temperatures, changes in hardness, etc.), the same chemical exposure may become very lethal. The toxicity of some chemicals may also be enhanced or mitigated in the presence of other chemicals. In addition to killing the organisms, some pesticides can have negative but non-lethal effects on individual organisms and populations, such as reduced reproduction, reduced mobility to escape predation, or alterations in behavior.

Toxicity Measurement and Estimation

One common measurement used to describe toxicity of pesticides to organisms is the LC_{50} , or the statistically derived concentration in water that can be expected to cause death in 50 percent of the animals exposed (Figure 3).

For estimation of non-lethal effects on processes such as growth and reproduction, the EC_{50} , or the statistically derived concentration in water that can be expected to cause a reduction of 50 percent in the process being measured, is used. Toxicity tests

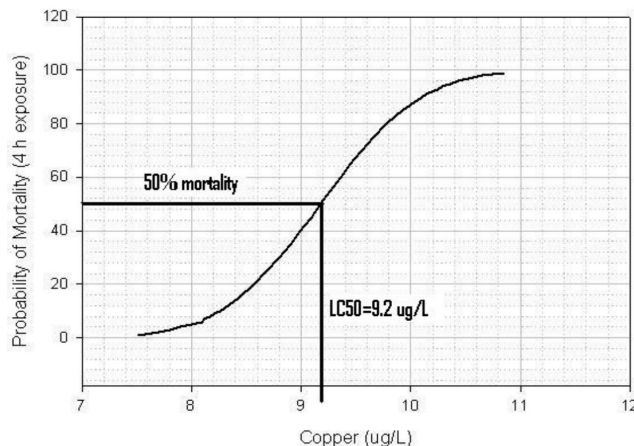


Figure 3. Sample toxicity curve showing water flea mortality. The sigmoidal line represents actual percent mortality. The $LC_{50} = 9.2 \mu\text{g/L}$ in this example.

usually fall into one of two categories, acute or chronic. Acute tests are designed to evaluate the effects of pesticides on survival following exposures for a short period of their lifespan. Animals used in these tests are normally exposed for 24-, 48-, 72-, or 96 hours in order to estimate acute toxicity. In contrast, chronic toxicity tests evaluate effects over a significant portion ($1/10^{\text{th}}$ of lifetime or longer) of the organism's life span. These tests often evaluate sublethal effects on reproduction, growth, and behavior, as well as mortality. Relative to acute effects, chronic effects may occur following exposures to lower concentrations of the pesticide. This chronic toxicity information is not always readily available because of the considerable expense associated with testing.

Standard Toxicity Testing Organisms

It is important to recognize that toxicity data will not always be available for all potential species in a given environment. Given this limitation, the overall objective of test organism selection is to choose surrogates that are representative of the major ecosystem components. Aquatic algae and plants are representative of organisms that convert sunlight to carbon-based energy (the base of the food chain). Invertebrate species such as scuds and water fleas feed on algae and decaying plant materials and bacteria. These organisms are an important food source for larger invertebrates and fish. Fish species

serve as an important source of food for a variety of larger fish, birds and mammals. Fathead minnows and sunfish often represent temperate warm-water fish, and trout represent coldwater fish species.

A listing of commonly used species in North America is provided in Tables 3, Table 4 and Table 5, along with a brief description of the significance of each. Of those listed, several organisms are more routinely used for estimating pesticide toxicity because of their sensitivities, ease of culturing, and because much is known about their life histories (i.e. physiology, genetics, behavior, etc.). These organisms include: water fleas (*Daphnia* sp.), fathead minnows (*Pimephales promelas*), bluegill (*Lepomis macrochirus*), rainbow trout (*Oncorhynchus mykiss*/*Salmo gairdneri*), mysid shrimp (*Mysidopsis* sp.), and sheepshead minnow (*Cyprinodon variegatus*). The use of these surrogates assumes that organisms within a group will respond similarly, which may not always be the case (see "Note Uncertainty" section for more information).

RISK ESTIMATION

The risk of toxicity between a given herbicide and aquatic organism depends on the organism-specific, inherent toxicity of the compound and the concentration and duration of the exposure. The inherent toxicity is associated with the presence of a specific mode-of-action for causing toxic effects, and cannot be changed. The exposure concentration and duration can be affected by all of the processes discussed in the previous sections.

Herbicides labeled for application directly to aquatic systems or to ditch banks may be of special concern in aquatic systems because of the more direct exposure routes, compared to applications in a terrestrial situation. While the labels for these herbicides have been formulated to minimize potential toxicity in treated water bodies, it is important to recognize that there is a margin of safety associated with use of each at the labeled application rates; and that margin of safety can be significantly reduced with improper use.

There are several methods for estimating the potential risks of toxicity to nontarget organisms. Below are descriptions of several common methods that may be useful for estimating risks to aquatic organisms.

Note Uncertainty. *Uncertainty refers to doubt associated with estimation. When trying to predict the ecological risks of any chemical to aquatic organisms, it is important to recognize sources of uncertainty within the estimate. Differing degrees of uncertainty exist for all aspects of a risk assessment, from estimating exposures to effects on the organism. Results of toxicity assays can be influenced by many factors including the initial health of testing-organisms, age and sex of the organisms, as well as treatment of those organisms before and during toxicity assays. Results generated in the lab may exaggerate or understate toxicity for a given species under actual local conditions in the environment. Likewise, organisms may vary in their response to pesticide exposures between species, genera, families, etc. Thus, results from toxicity tests should not be considered as absolute values, but rather as possible values occurring within a range of probable values. These values are especially useful for comparing relative toxicity of pesticides to organisms. The common practice when toxicity data are not available for a given species within the environment of interest is to use a related species. It should be recognized that the response of the chosen species may or may not be similar.*

Ratio of Environmental Concentration / Toxic Concentration

One simple (but less precise) toxicity screening method is to compare expected environmental concentrations of the pesticide to levels causing toxicity to relevant organisms in that environment. Environmental concentrations greater than the levels causing toxicity constitute significant risks. This approach is useful as a screening tool, and is easy to calculate with herbicides applied directly to the water in order to achieve a target concentration. However, estimation of concentrations in the water resulting from ditchbank or drained-ditch/pond applications is more difficult because of partitioning and degradation uncertainties. Estimation of environmental concentrations in these situations may

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Table 3. Listing of common fish used for toxicity testing in North America, and their ecological significance.

Common Name	Latin Name	Description and Ecological Significance
Bluegill	<i>Lepomis machrochirus</i>	One of largest and most common sunfish; native to eastern/central North America; Habitat: shallow, weedy, warm water of lakes, ponds, and heavily vegetated slowly moving areas of small rivers and larger creeks.
Channel catfish	<i>Ictalurus punctatus</i>	Commercially significant; freshwater fish; Range: throughout North America; feeds on small fish, crustaceans (i.e. crayfish), clams, snails, aquatic insects, and small mammals.
Fathead minnow	<i>Pimephales promelas</i>	Small, warm/fresh water species; Habitat: ponds, lakes, ditches, slow muddy streams; omnivorous, feeding on living invertebrates and detritus; widely distributed.
Rainbow trout	<i>Oncorhynchus mykiss</i>	Cold/fresh water species; widespread in NE North America east of the Mississippi river. Economically important.
Sheepshead minnow	<i>Cyrinodon variegatus</i>	Grows up to 3 inches long; tolerant of high temperatures and salinities; typically found in estuaries; shallow water.

Table 4. Listing of common aquatic plants used for toxicity testing in North America, and their ecological significance.

Common Name	Latin Name	Description and Ecological Significance
Duckweed	<i>Lemna</i> sp.	Small (2-4 mm) floating plant; food source for water fowl and small animals; provides food, shelter, and shade for fish; provides physical support for various invertebrates.
Green algae	<i>Selenastrum capricornutum</i>	Representative of freshwater primary producers, which are the base of freshwater aquatic food chains.
Marine algae	<i>Skeletonema costatum</i>	Representative of marine water primary producers, which are the base of marine aquatic food chains.

Table 5. Listing of common aquatic invertebrate organisms used for toxicity testing in North America, and their ecological significance.

Common Name	Latin Name	Description and Ecological Significance
Amphipods/Scuds/Sideswimmers	<i>Hyalella azteca</i>	Widely distributed throughout North American fresh and marine waters; Habitat: wide variety of unpolluted lakes, ponds, streams, brooks, springs, pools, and lakes; found in vegetation or hidden under and between debris and stones during day; typically found in shallow (<1m) waters. Valuable food source for larger organisms.
Grass shrimp	<i>Palaemonetes pugio</i>	Small (1-2 inches), non-edible shrimp; transparent; usually found in estuaries; feed on algae and detritus in grasses of salt marshes and salt ponds; use same grasses for shelter.
Marine amphipods	<i>Rheopoynius pugio/Ampleisca abdita</i>	Marine invertebrate; omnivorous, consume all kinds of plant and animal matter; seldom predacious; important food species for small fish.
Marine clam	<i>Macoma</i> sp.	Marine shellfish; bivalve, wafer-thin shells; found in sand or mud; grow up to 4 inches long; found 4-6 inches below surface of water in the middle of the intertidal zone.
Marine mussel	<i>Mytilus edulis</i>	Marine shellfish; roughly triangular-shaped, hinged shells (bivalve); commonly found in estuaries and from the middle shore to shallow sublittoral zone; filter feeds on bacteria, plankton, and detritus; serves as food for whelks, crabs, sea urchins, starfish, sea birds, and other animals.

Table 5. Listing of common aquatic invertebrate organisms used for toxicity testing in North America, and their ecological significance.

Common Name	Latin Name	Description and Ecological Significance
Marine worms	<i>Nereis virens</i>	Burrow in muddy sand areas of the littoral and sublittoral zones in both marine and estuarine habitats; found throughout northern hemisphere; preys on invertebrates, carrion, and algae; up to 8 inches long.
Mayflies	<i>Hexagenia</i> sp.	Habitat: lentic and lotic depositional sand-silt; burrowers; collectors-gatherers, possibly filtering at mouth of burrow; widespread.
Midges	<i>Chironomus</i> sp.	Burrowers found in lentic-littoral and profundal, lotic depositions sediments; widespread; herbivores, feeding as collectors-gathers, shredders.
Mysid (opposum) shrimp	<i>Mysidopsis bahia</i>	Salt water, colorless shrimp; very important invertebrate predator.
Oysters	<i>Crassostrea virginica</i>	Shellfish with two rough shells hinged together; typically 2-6 inches long; filter feeders, feeding on plankton and detritus; range includes eastern shore of North America and the Gulf of Mexico.
Penaid shrimp	<i>Penaeus</i> sp.	Commercially important; salt water; range generally restricted to Atlantic coast and Gulf of Mexico; omnivorous (eat both plants and animals); bottom feeders; may spawn in estuaries or sea.
Water fleas	<i>Daphnia</i> sp.	Ubiquitously distributed in freshwater bodies; ecologically important because they convert phytoplankton and bacteria into animal protein and form a major portion of the diet for many small fish species; relatively short lifecycle.