LOTIC DIPTERA OF KHAO YAI NATIONAL PARK, THAILAND, WITH EMPHASIS ON THE DIVERSITY, TAXONOMY, AND ECOLOGY OF BLACK FLIES (SIMULIIDAE)

INTRODUCTION

Few studies have been made of the diversity of aquatic Diptera in Thailand. Consequently, data are lacking on ecosystem structure, especially biodiversity. The Diptera are an interesting group of aquatic insects. They play an important role in aquatic ecosystems, as larvae and pupae are a food resource for other invertebrates, fish, amphibians, reptiles, birds, and mammals (Courtney, 1994; Courtney *et al*, 1996). Adults of many aquatic families are important amongst the bloodsucking insect. Many diseases of humans and animals are vectored by aquatic families such as Culicidae (mosquitoes), Simuliidae (black flies), Ceratopogonidae (biting midges), Psychodidae (sand flies), and Tabanidae (horse flies) (Williams and Feltmate, 1992). Biting habits can be a direct health hazard by taking quantities of blood, inflicting pain, and sometime causing allergic reactions (McCafferty, 1998). For example nonbiting midges in the family Chironomidae may damage property and create public health problems in some people (Hilsenhoff, 1991). Aquatic Diptera can serve roles in the monitoring of water quality as well as scientific research on the structure and function of aquatic ecosystems, structural and ecological adaptation to aquatic environments, and patterns and mechanisms of evolutionary divergence (Courtney *et al*, 1996).

Taxonomic characters of Diptera are diverse and complex. The order is recognizable by the presence of only one pair of wings. This character distinguishes the Diptera from other insects. However, within some groups flies are similar morphologically. For example, some black flies are morphologically indistinguishable, but are biologically separate species in nature known as sibling species. In these situations, these groups of species are referred to as species complex (Crosskey, 1990). Chromosome studies have been very important in black fly taxonomy (Rothfels, 1979). They have provided not only a valuable supplement to basic morphological species identification, but have led to the detection of many isomorphic sibling species.

This study was conducted on lotic habitats (running water) in Khao Yai National Park. Data on the species richness, abundance, distribution of lotic Diptera was expected to provide insights into potential bioindicators of water quality.

Objectives

The objectives of this study were to:

1. Characterize streams of various size, altitude, and physical characteristics by lotic Diptera species encountered.

2. Analyze morphologically and identity of the Simuliidae species in Khao Yai National Park.

3. Study chromosomal characters of a selected group of Simuliidae.

4. Document patterns of species richness and abundance, seasonality, and habitat in Simuliidae communities.

LITERATURE REVIEW

The Diptera, two-winged or true flies, is a large order of endopterygote Neoptera. It is estimated that the order contains approximately 200,000 species, worldwide (Williams and Feltmate, 1992). Hilsenhoff (1991) estimated that about 40% of all species of aquatic insects belong to this order, and at least a third of aquatic Diptera species are in one family, Chironomidae. It is difficult to estimate the number of species of aquatic Diptera because there exists a lack of knowledge about the biology and ecology of some species. Merritt *et al.* (1996) noted that aquatic Diptera belong mainly to the suborder Nematocera, where 15 of the 25 extant families contain aquatic species. The suborder Brachycera has 15 of 84 extant families with aquatic or semiaquatic species.

Life History

Aquatic Diptera are typical holometabolous insects with complete metamorphosis that includes four stages: egg, larva, pupa, and adult. In general, eggs are small and elongate-oval. The eggs are deposited singly, in small clusters, or irregular masses in or near the water and attached to rocks or vegetation (Courtney *et al*., 1996). They may also be laid in well-defined rafts or rosettes (some Culicidae) or in a gelatinous matrix (most Chironomidae) (Colless and McAlpine, 1991). Hatching usually occurs within a few days or weeks, but sometimes much longer, depending on species and water temperature.

The number of larval instars varies among Diptera; typically 3 in Brachycera, 4 in nematocerous Diptera although some species have more (e.g. Simuliidae usually have seven) (Courtney *et al*., 1996; Peterson, 1996). For a particular species, all larval instars usually occur in the same habitat except lentic Chironomidae (Peterson, 1996). Larval form is variable but usually elongate and cylindrical or dorsoventrally flattened. The integument usually bears rows of microscopic spines or setae that may provide important taxonomic characters. Segmentation is normally distinct including the head, 3 thoracic, and 8 to 10 abdominal segments (these segments are occasionally fused into fewer apparent segments or appear to be subdivided). The head may be well developed or not. Antennae are variously developed. Mouthparts are often highly modified. The larval thorax has no true legs, although one or two anterior prolegs occur in some family (e.g. Chironomidae and Simuliidae). The abdomen may have prolegs on various segments and the terminal segment may have prolegs or other fleshy or filamentous processes (Colless and McAlpine, 1991; McCafferty, 1998).

Diptera larvae have a variety of respiratory adaptations. Respiration can be directly from the atmosphere (e.g. Dixidae, most Culicidae, Tipulidae, and Syrphidae), from plant tissues (e.g. some Culicidae), or through absorbing dissolved oxygen directly from the water through the skin (particularly Chironomidae, Blephariceridae, Deuterophlebiidae, Nymphomyiidae, Simuliidae, and Ceratopogonidae). The presence of haemoglobin in the blood of certain bottom-dwelling Chironomidae may assist the absorption of oxygen.

The duration of larval development may range from several weeks to as long as months depending on the species, water temperature, and food conditions (Williams and Feltmate, 1992). Larval growth of many species fluctuates as a reaction to seasonal changes. It is interesting that among Chironomidae, a few species can survive extreme dehydration (a phenomenon known as cryptobiosis), freezing, or heat during unfavorable growing conditions. Many Diptera species have both summer and overwintering generations (McCafferty, 1998).

The characters of Diptera pupa are relatively simple and adecticous. In the Nematocera and Orthorrhapha almost all are obtect, whereas the Cyclorrhapha are secondarily exarate and coarctate, being enclosed within a hardened puparium (capsule like case) that conceals developing wings and legs. Obtect forms are either free-living or live within a structured cocoon. The pupae are usually immobile, though in many Culicomorpha are active. The pupal thorax has one pair of developing wings. Appendages are either fused to the body or held rigidly apart from it. The puparium or pupa often has a pair of dorsal thoracic respiratory horns or gills. Free-living pupae commonly also have a terminal breathing apparatus.

Pupation sometime occurs in the larval habitat, with current also influencing microhabitat selection (e.g. Simuliidae) (Eymann, 1991). Other species leave the water to pupate in marginal areas, usually in thick-matted vegetation or in mud or sand (e.g. some Tipulidae, Tabanidae and Dixidae). The pupal stage lasts for only a short time, except in species that overwinter as pupae.

When the adult has become fully formed with the pupal skin, it is called a "pharate adult". Before adult emergence, the pupa or pharate adult moves to the surface (e.g. Culicidae, Chironomidae, and Tabanidae), unless the pupa is attached to the substrate in which case the adult emerges under water and floats quickly to the surface (e.g. Simuliidae and Blephariceridae). The pharate adult bursts from the pupal skin or puparium usually by the pressure of swallowed air. It emerges from a lengthwise split in obtect pupae; in coarctate forms, however the adult protrudes the ptilinum to push off the anterior end of puparium (Colless and McAlpine, 1991; Grillott, 1995; Daily *et al*., 1998; McCafferty, 1998).

Structures of the adult generally include a single pair of membranous wings. Hind wings are modified into small knobbed structures, known as halteres whereas the fore wings are complete or rarely reduced or absent. Hairs are sometimes variously developed on the wings. Compound eyes are large, sometimes meeting on dorsal side of head. Antennae are variably shaped. Adult mouthparts are often reduced or modified for piercing and sucking or cutting and lapping. The thorax is well developed and somewhat humplike. The legs of some flies are very long (e.g. Tipulidae), whereas the legs of other flies (e.g. Simuliidae) are quite robust. Tarsi always have 5-segments. The abdomen is usually elongate but with greatly reduced cerci (McCafferty, 1998).

In some newly emerged adults, it takes several minutes before structures become hardened enough for flight. In others, particularly flies in rapid or swiftly flowing water (e.g. some Chironomidae, some Simuliidae, and Blephariceridae), the adults are adapted for flying immediately upon emergence. Most adult flies of aquatic taxa are found around or very near water but some, such as piercing and sucking feeders that require a blood meal for ovarian development, can occur miles away from the nearest watersource (e.g. Ceratopogonidae, Culicidae, Simuliidae, and Tabanidae). The adult stage lasts from a few hours to several months, depending on the species.

Mating occurs in flight, on the ground, vegetation, or even occasionally on the water. Many species mating is preceded by various kinds of swarming or courtship behavior. In many Nematocera and some Brachycera have aerial swarming behavior, in which males form dense, dancing masses, often near characteristic landmarks or over water. Among some flies (e.g. Ceratopogonidae) mate recognition is a response to the wing beat tone made by the female when flying into the swarm. Other Diptera apparently use visual cues. A female usually enters the swarm, pairs with a male, then the coupled pair leaves the swarm. Some females require a blood before laying eggs (Merritt *et al*., 1996; Daly *et al*., 1998; McCafferty, 1998).

Oviposition behavior in Diptera is very variable. Depending on species, females lay eggs directly on the water, submerged substrates, shoreline substrate, aquatic vegetation, (McCafferty, 1998). In most groups, females select oviposition sites that are suitable for larval development. In some groups (e.g. Deuterophlebiidae, Nymphomyiidae, and some Simuliidae), the female crawls beneath the water, losing its wings in the process of oviposition (Courtney *et al*., 1996; Peterson, 1996). The hue and intensity of light reflected from substrates can influence ovipositing black flies (Golini and Davies, 1974).

Some aquatic Diptera pass through one (univoltine) or two (bivoltine) generations a year and adults are usually present during the spring or summer (e.g. some Tipulidae, Tanyderidae, Simuliidae, Blephariceridae, and Stratiomyidae). Other groups may undergo several generations a year (multivoltine) and adults may be found during all seasons (e.g. some Culicidae, Ceratopogonidae, and Ephydridae) (Merritt *et al*., 1996; Service, 1996).

Habitats

Aquatic habitats can be divided into freshwater and marine ecosystems. Freshwater ecosystems are characterized as having lotic (flowing water) habitats and lentic (standing water) habitats. Marine ecosystems include the oceans of Earth, bays, and estuaries (Mix *et al.*, 1992; Kormondy, 1996). Diptera may be found in every aquatic habitat except the open ocean (Courtney *et al.*, 1996). However aquatic Diptera have many adaptations to live in a wide variety of aquatic habitats (Table 1) including in the drains of kitchens and restrooms (McCafferty, 1998).

Feeding

The taxonomic and ecological diversity of Diptera is reflected in the wide range of larval feeding behavior and food preferences (Courtney *et al*., 1996). Food sources include fine detritus, algae, plants, nectar, decaying wood, microorganisms, insects, and other invertebrates. The food preferences and feeding habits of some larvae (e.g. Chironomidae) change with the age of the larvae and the season of the year (McCafferty, 1998).

Functional feeding groups are based on food acquisition mechanisms (Cummins and Merritt, 1996). Courtney *et al*. (1996) discussed larval feeding habits among aquatic Diptera, which many include the following functional feeding groups:

1. Shedders, which consume live plants or decomposing plant fragments, are particularly well represented by larvae of the Tipulidae and Ephydridae.

2. Collector-gatherers feed on decaying, fine organic matter and associated microorganisms (e.g. Tipulidae, Ephydridae, Psychodidae, Ptychopteridae, and Syrphidae).

3. Collector-filterers consume diatoms, protozoa, plankton, and other small organisms using modified mouth-brushes or labral fans to catch particles from the water (e.g. most Culicidae and Simuliidae). Other groups (e.g. Stratiomyidae and Syrphidae) lack external filtering devices, but accomplish the same task with a filter-like pharynx.

Food particle size may correlate with larval size. For example, large larvae of simuliidae (*Simulium noelleri* Friederichs, 1920) have a longer gut retention time than small larvae, so they can digest particles greater than 52 µm in diameter, whereas small larvae are better suited to digest particles smaller than 13µm. Capture of different sized particles by larvae of different ages may be a mechanism for reducing intraspecific competition for food (Wotton, 1984). However, Alencar *et al*. (2001) reported larval sizes of *Simulium perflavum* Roubaud, 1906 were not associated with food type. Moreover, current velocity has been shown to affect ingestion rates (Charpentier and Morin, 1994).

4. Grazers, which feed on the thin periphytic film of algae and organic matter on rocks and other substrates, include the highly specialized families Blephariceridae, Deuterophlebiidae, and Thaumaleidae, and various members of the Psychodidae (e.g. *Maruina* Müller), Ephydridae (e.g. *Parydra* Stenhammar), Simuliidae (e.g. *Gymnopais* Stone and *Twinnia* Stone & Jamnback), the latter of which lack labral fans (Craig, 1973).

5. Predators feed primarily or exclusively on other animals (e.g. Ceratopogonidae, Athericidae, Dolochopodidae, and Tabanidae) whereas the larvae of many families (e.g. Chironomidae, Culicidae, Tipulidae, and Ephydridae) contain a few predaceous species.

6. Parasitoid habits are rare among aquatic Diptera larvae. For example, the Sciomyzidae inhabit freshwater snails and fingernail clams, and Dryomyzidae (e.g. *Oedoparena* Curren) feed on barnacles.