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Aspects of the ecology of two sympatric species of Thamnophis and heavy metal accumulation within the species

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ASPECTS OF THE ECOLOGY OF TWO SYMPATRIC SPECIES
OF THAMNOPHIS AND HEAVY METAL ACCUMULATION
WITHIN THE SPECIES

By

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A narrow fellow in the grass
Occasionally rides;
You may have met him did
  you not?
His notice sudden is.

The grass divides as with a
  comb,
A spotted shaft is seen;
And then it closes at your feet
And opens further on.

He likes a boggy acre,
A floor too cool for corn.
Yet when a child, and barefoot,
I more than once, at morn,

Have passed, I thought, a
  whip-lash
Unbraiding in the sun,-
When, stooping to secure it,
It wrinkled, and was gone.

Several of nature's people
I know, and they know me;
I feel for them a transport
Of cordiality;

But never met this fellow,
Attended or alone,
Without a tighter breathing,
And zero at the bone.

—Emily Dickinson

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Aspects of the ecology of two sympatric species of Thamnophis and heavy metal accumulation within the species (147 pp.)

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Two species of garter snakes, Thamnophis elegans vagrans and Thamnophis sirtalis parietalis were studied from eight selected sites in Western Montana. A four-faceted approach was followed in the study of the biology of these animals.

One part of the survey entailed the analysis of the tissues of some snakes and frogs for contamination by lead and mercury. Snakes from six sites were compared to those taken from East Helena from the vicinity of the ASARCO smelter, and from the area of the stamp mill along Fred Burr Creek at Phillipsburg. Thamnophis and frogs at East Helena ingested and absorbed more lead from their environment than did the snakes from other sites. Frogs and fish from Fred Burr Creek showed heavy concentrations of mercury.

Sympatric populations of Thamnophis were compared in relation to temperature and food preference. No obvious difference in activity temperature range was preferred by either species. The principal food items of the snakes included amphibians, leeches, and slugs. Leeches were the mainstay of both species throughout the summer.

Population size as an indicator of ecological disturbance was determined by the capture, marking, releasing, and recapture method. Marking of the snakes was done by scale-clipping.

Garter snakes from Western Montana exhibited a wide range of counts of scales, especially in the labial scales. A statistical analysis of the scale counts from the six selected sites were compared to those of preserved specimens and from an earlier study. Supplementary data on possible genetic inheritance were obtained from the broods of four snakes.

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CHAPTER I

INTRODUCTION

The use of organisms as biological indicators was initiated by Merriam (1894); this method was employed in the present study of heavy metal contamination and ecological relationships in the red-sided garter snake (Thamnophis sirtalis parietalis) and the wandering garter snake (Thamnophis elegans vagrans). Mercury and lead were chosen as the areas of concentration in heavy metal analysis; D'Itri (1972) and Friberg and Vostal (1972) together provided a comprehensive summary on the status of mercurial contamination, and the National Academy of Sciences (1972) provided data on the causes and effects of lead pollution. Background on existing problems in Montana for mercury and lead was obtained respectively from Van Meter (1974) and Gordon (1972,1973).

The incorporation of reptiles in environmentally-oriented studies has been ignored until this present decade. Dustman et al. (1972) included herpetofauna in their review of mercury in wild animals around Lake St. Clair. Brisbin et al. (1974) specifically chose snake species as indicators of radiocesium contamination; and Bauerele et al. (1975) selected species of snakes for a survey on chlorinated hydrocarbons and lead.

In this present study, garter snakes were chosen instead of other herpetofauna because of their relative abundance. As snakes, however, they also displayed two characteristics that were ideally suited for heavy metal analysis. Because it was the nature of snake species to maintain a home range of no more than a few kilometers (Hirth et al. 1969), an accurate measurement of contamination from a specific locality was possible. And, because snakes are a predatory species, they exhibit a higher concentration of pollutants than members of other, lower trophic levels in the same ecosystem.

DESCRIPTION OF THAMNOPHIS SIRTALIS PARIETALIS

Thamnophis sirtalis parietalis Say (1823), the red-sided garter snake, was predominantly olive brown on the dorsum; the venter and caudum varied from a slate grey, blue, cream, to yellow. The dorsal stripe was wide and distinct; it and the lateral stripes were yellow. Two rows of quadrate black blotches occurred on both sides between the dorsal and lateral stripes; the first was distinct, the other confluent (Schmidt and Davis 1941). Brick-red to orange bars broke up the olive dorsum, which had corresponding red skin below the bars. Orange coloration may be extended up to the posterior upper labials. Black dots occurred on either one or both sides of the upper labia, on the third scale from the rear. Extra scales, which were triangular, were found occasionally between the three posterior scales on the upper labia. The majority of specimens displayed the following scale counts: (from right to left) upper labials: 7-7; lower labials: 10-10; dorsal row: 19-19-17 or 19-19-16; ventrals: 150-178; caudals: 65-95.

The red-sided garter snake was first described as Coluber parietalis by Say; later, it was reclassified by Cope in 1875 as Eutaenia sirtalis parietalis. The first specimens to be collected of this species were found in 1873 by Dr. Coues (Cope 1900). The species was primarily riparian, and has been found from sea level to 8000 feet in elevation (Wright and Wright 1957). In western Montana, a sympatric association commonly occurred between this species and the wandering garter snake (Thamnophis elegans vagrans). The subspecific group parietalis was distributed more widely than the other subspecies in the United States; it has been found from the Mississippi River to Utah, and eastern Nevada and southern Idaho, southern Alberta, Saskatchewan and Manitoba, southward through the Great Plains to Oklahoma (Ruthven 1908, Stebbins 1966).

DESCRIPTION OF THAMNOPHIS ELEGANS VAGRANS

In 1853 Baird and Girard described Thamnophis elegans vagrans, the wandering garter snake. The overall color was olive to earthen. A yellow dorsal stripe, which was often indistinct, extended the length of the body. Two lateral stripes were present, also. A pair of black checkers paralleled the stripes and were located between the dorsal and lateral stripes. White specks were located interstitially among the dorsal scales; they were most obvious when the scales were distended. The ventral scales had a blackened center; a lessening in the melanistic pigment occurred at the anterior and posterior ventrals. Frequently, there was more than one undivided caudal randomly distributed among the other caudal scales. Labials tended to be melanistic in the corners of the mouth; and yellow dots were present in the center of the parietal scales. The normal scale counts for the species were: (from right to left) upper labials: 8-8; lower labials: 10-10; dorsal row count: 21-21-17 or 20-20-17; ventrals: 147-184; caudals: 60-93.

The wandering garter snake was first described as Eutaenia elegans vagrans by Baird and Girard, and it was first collected in Montana by Dr. Coues in the 1870's (Cope 1900). This species was correctly named for its habitat preference; it has been found in many variable ecotypes, terrestrial or aquatic. Its range in altitudinal distribution was from sea level to 10,500 feet (Stebbins 1966). Nationally, this species has been found from the Great Basin and the Rocky Mountain Ranges, from Alberta and British Columbia southward, eastward from western South Dakota and Oklahoma to the southern limits in high Sierra Nevada and southwestern New Mexico (Ruthven 1908, Wright and Wright 1957).

PURPOSES OF THIS STUDY

The primary objective of this study was to determine the potential of garter snakes as an indicator species. This study compared data from relatively non-contaminated areas with contaminated areas for both mercury and lead; muscle and liver tissue were analyzed in the survey.

The secondary objective was to investigate the sympatric relationship between the two species in the area of food preferences and body temperature activity ranges. The tertiary objective was to ascertain by a statistical test the validity in the usage of population size as an indicator of environmental disturbance. The final objective was to survey the genetic variation in the scale counts and patterns of both species of garter snakes.

A chapter on each of the objectives follows; they have been treated as individual units. Because of this approach, an introductory section has been included in each one of the chapters; thereby it provided the immediate background data which were pertinent to the specific objective.

CHAPTER II

DESCRIPTION OF COLLECTION SITES

Herpetofauna were collected from eleven sites; ten of these sites were in Western Montana. At most of these sites only _Thamnophis_ were collected; however, amphibians were collected at specific sites for heavy metal analysis. The sites were chosen from previous collection records. Three of the factors which were important in the selection of the sites were 1) the relative disturbances that occurred at the site from man-made causes, 2) the size of the population of _Thamnophis_ which inhabited the area, and 3) the presence of gravid individuals which were selected for a genetic study of the species.

WOODS POND

Woods Pond (Township 20N, Range 19W, Section 20, Lake County) was an area which contained a small permanent pond along the access road; two smaller bodies of water, and a marsh. It was approximately 32 kilometers south of Polson, and 3 kilometers east of Highway 93.

The permanent pond was split by the road. The north sector (142 meters in perimeter) was covered in abundance by the yellow water lily (_Nuphar advena_) and the water buttercup (_Ranunculus aquaticus_). The south sector (100 meters in perimeter) had the same plants on its surface; however, it was more sheltered by trees and shrubs. The ponderosa pine (_Pinus ponderosa_) and the sub-alpine fir (_Abies lasiocarpa_) caused the southern end of the pond to be significantly cooler than the northern end. Cattails (Typha latifolia) were in a small cluster in the northwest end. One of the smaller ponds, which was in the wooded area, was solely covered by duckweed (Lemna minor).

Most of the garter snakes (Thamnophis spp.) were found around the periphery of the permanent pond, in which two food sources were present. These were a species of leech, Erpobdella punctata, and the spotted frog, Rana pretiosa. Other garter snakes frequented the marsh, which contained metamorphosing long-toed salamanders (Ambystoma macrodactylum). The boreal toad (Bufo boreas) was occasionally seen in the adult stage at this site; and a population of western painted turtles (Chrysemys picta belli) lived in the permanent pond.

There were rare occurrences of white-tailed deer (Odocoileus virginianus) and great blue heron (Ardea herodias), and frequent visits by cattle and tractors in the area. Only once was another man-made disturbance found; two painted turtles, that were shot with .22 caliber bullets, were alongside the permanent pond.

In the wooded regions, which surrounded all these bodies of water, a few piles of brush and shrubs ostensibly served as hibernacula. These were sites in which a large number of snakes were found emerging from underground.

Collection trips were made to this site on June 19, 27; July 4, 12, 18, 29; August 5, 1974; and June 22, and September 14, 1975.

JETTE POND

Jette Pond (T23N, R21W, S2, Lake County) was a montane pond (ele. 2,124 m) in the Flathead Indian Reservation. It was under the supervision of the Tribal Council of the Confederated Flathead, Salish, and Kootenai Indians and was accessible by a road, which was principally used to log the pond's immediate vicinity and surrounding forest.

The pond (400 meters in perimeter) was surrounded by an association of ponderosa pine, lodgepole pine, Englemann spruce, and bunchgrass and was within an open range for an active herd of 40-50 cattle. The pond was a permanent source of water and of supplementary plants for the cattle. Consequently, as the water lowered in depth throughout the summer, the pond's banks became increasingly packed down by the cattle's hooves. The pond, therefore, retained an overall brownish cast from the runoff of all the organic material produced by the cattle.

The aquatic vegetation consisted of mainly smartweeds (*Polygonum sp.*), water buttercups (*Ranunculus aquaticus*), and several species of pondweed (*Potamogeton sp.*). A floating mat of cattails, which occurred along the northwestern shore, was frequented by a pair of ducks.

The southern end of the pond differed in shape and depth from the northern end. Although the southern end was not under direct exposure such as that of the northern end, the southwestern area completely dried up by the end of summer. This was because a small forested peninsula jutted out into the southern end of the pond and formed a shallow, western inlet. The other inlet, which was surrounded by trees, was deeper and remained cool.

The pond was thinned more on the western margin than on the eastern margin; however, felled trees occurred on both sides. Many of the trees jutted out into the pond and were used as resting sites for garter snakes and amphibians alike. The most logged area was around the northern end, which was closest to the road. Here the shoreline, which extended more than 46 meters from the road, was composed of barren ground, large rocks, burnt stumps, and scattered logs and branches.

The pond harbored a variety of aquatic animals. Many of these served as food for both species of garter snakes. Metamorphosing and adult individuals were seen of the Pacific treefrog (H. regilla), the leopard frog (R. pipiens), and the spotted frog (R. pretiosa). Bufo boreas, which were adults, were found near the pond at evening. Two types of leeches, Haemopis grandis and Erpobdella punctata, were prevalent in the water.

An anomaly was found in a small percentage of metamorphosing treefrogs. They possessed extra hind limbs. This was first noticed in 1958, and successive collections were made from 1959-1961 (Hebard and Brunson 1963). The anomalies could have been caused from many environmental factors among which was the radioactive fallout of 1958; however, its present occurrence implied that it developed into a genetic mutation. Extra limbs have not been found in any of the other amphibians which populated the pond.

Research trips were made to the site June 19, 28; July 4, 13, 17, 29; and August 4, 1974; and September 14, 1975.

JETTE LAKE

Jette Lake (T23N, R21W, S14, Lake County) was approximately 16 hectares in area and was located 16 kilometers northwest of Polson. The lake and surrounding acreage, which was under the process of being subdivided, were privately owned by Mr. Christian. Development was to be no closer than 61 meters from the water's edge; however, a disturbance in the form of piling and burning of brush took place within this margin.

A variety of deciduous trees (i.e. alder, willow, and cottonwood) and ponderosa pine emarginated the lake. The northeastern end of the lake contained an intermittent drainage stream, which was surrounded by a large open area of grass. This area was grazed by a herd of Hereford cattle; and on occasion, they could be found in the forested regions.

Polygonum and _Potamogeton_ were plentiful among the aquatic plants. The aquatic plants were fed upon by two families of ducks, which had been planted in the lake, muskrats (_Ondatra zibethica_), and western painted turtles. The painted turtles were also observed to use the shore of the lake for areas of oviposition.

The piled brush and logs served as a shelter for both the wandering and red-sided garter snakes. On one occasion, a nest of ground squirrels was found under one such log. An unknown scale of impact was caused in particular to the garter snakes, when the logs and brush were burned.

Salmonids, which were introduced into the lake, and shiners (_Richardsonius_) inhabited the lake itself. Fish served as part of the diet of _Thamnophis spp._; whereas _B. boreas_ and _R. pipiens_, which inhabited the margins of the lake, were not included in the diet of the research animals.

Herpetofauna were observed at this site on June 19, 27, 28; July 3, 12, 17; and August 4, 1974. A follow-up trip was made on June 22, 1975.

MUD LAKE

Mud Lake (T31N, R19W, S9, Flathead County) was situated 20 kilometers northwest of Columbia Falls. It was fed by the waters of Lake Five, which was a popular resort and recreation area. The two lakes were separated by approximately 1.6 kilometers in distance. Mud Lake (approximately 2.5 hectares) was the smaller lake and received occasional use (i.e. boating, swimming, fishing).

Both lakes were rehabilitated twice by the State Department of Fish and Game (i.e. in September, 1960, and May, 1968). The target species in Mud Lake were suckers and pumpkinseed. Brook trout and cutthroat trout were stocked; however, all four species of fish were present at the time of this study.

Part of the stream, which flowed from the end of the forested area to the inlet of the lake, was included as part of the habitat study of the wandering garter snake. The stream (ca. 213 meters in length) was paralleled by steeply-sloped banks that were composed of medium-sized rocks. On these banks, garter snakes were seen sunning and, inasmuch as they had fresh fish in their stomachs, it was assumed that the fish were caught from the schools in the stream.

The lake and stream were situated on a northeast-southwest axis. The eastern side of the stream was relatively undisturbed and contained a small spring-fed marsh. Rana pretiosa were commonly found in this marsh. Further north on both eastern and western banks, a few large logs were present. These often harbored a type of slug (Prophysaon), a form of prey for garter snakes, and were a cover for these snakes. The area of the western bank was periodically disturbed by the construction of an access road for the telephone line, which emarginated the stream and lake.

The stream widened at the mouth of the lake. A Carex-mat originated and spread widely on both sides of the lake from this point. Intermingled among these mats of Carex, random clumps of cattails and the yellow water lilies occurred. These mats supported various logs and shrubs, which were used by the garter snakes as resting areas, and were separated by channels of water from the lake's shore.

The lake itself (24,282 square meters in area) was surrounded by a growth of alders, willows, and hawthorne trees. A black bear frequented the forested area of the southern end of the lake. The area in which the largest congregation of snakes were found, was on the southeastern shore of the lake. It was protected by the trees and had many planks of wood that the snakes were either on or under.

No red-sided garter snakes were sighted at this lake. However, two of its food sources, which were a population of Rana pipiens that lived in the lake and a few adult Bufo boreas, were present at this site.

Collections and observations were made on June 21, 30; July 15, 16; and August 1, 2, 1974; and September 13, 1975.

SMITH LAKE

Smith Lake (T27N, R22W, S4 and 9, Flathead County) was approximately 16 kilometers southwest of Kalispell. It was situated in the Ashley Creek drainage, and the primary access site was the Kila Fishing

Access at the northwest side of the lake. The lake (ca. 180 hectares in area) was approximately 2.4 kilometers in length.

The northern end was a marsh and was designated by the State Department of Fish and Game as an area for hunting waterfowl and fishing. The lake was deeper in the southern end, and its outflow was used for private irrigation purposes. Private property comprised the major portion of both sides of the lake. Because of the lack of public access, this study was centered at Kila.

The total length of the western shoreline which was studied was approximately 800 meters; and a distance of 90 meters from the shoreline toward the road was also surveyed. The study area incorporated various environments; from north to south, the shore consisted of a hay field (200 meters), Kila Fishing Access (400 meters), an abandoned pig farm (80 meters), a waterfowl production area (80 meters) and part of a cow pasture (40 meters).

Both species of garter snakes were found among the grasses and reeds (i.e. Scirpus and Carex) along the shoreline, as well as inland under felled trees and planks of wood. However, only the red-sided garter snake was found in the water. Three types of amphibians, the leopard frog, boreal toad, and the Pacific treefrog, were present. Only the former two were seen in metamorphosing as well as adult stages.

The amphibians were fed on heavily by the garter snakes, but the leopard frog and boreal toadlets were caught indiscriminately by fishermen and children alike. A supplementary prey species, Haemopis grandis, a type of leech also inhabited the lake. In the shallow waters large populations of snails (Helisoma trivolvis and Lymnea stagnalis) were abundant.

The western painted turtle was present, but only found on the shore. (Of those found, all were infected by a fungus.) Many species of shore birds and waterfowl were sighted, among which were gulls, avocets, and great blue herons. The gulls were occasionally seen preying upon toadlets.

The water along the west shore contained many aquatic plant species. Potamogeton, Polygonum, Nuphar, and the blue-green alga, Nostoc, were predominant. Within the waters, members of the Percidae, Centrarchidae, Castostomidae, and Cyprinidae lived.

Research trips were made to the site on June 29; July 2, 13, 14, 15, 16, 17, 31; August 1, 3, 1974; and June 29, 1975.

LAKE ROGERS

Lake Rogers (T27N, R23W, S29, 30, 31, and 32, Flathead County) was located in the Salish Mountains. The lake (ele. 1,188 meters) was located approximately 30 kilometers southwest of Kalispell. The target species of the rehabilitation (in 1958 and 1967) by the State Department of Fish and Game was redside shiners (Richardsonius balteatus). Grayling (Thymallus signifer) were re-introduced, and a population was present at the time of this study (personal communication).

The lake (107 hectares in area) was situated on a northeast-southwest axis. It was entirely privately owned on its southern shore; the other side was equally divided into private lots and public access sites. Therefore, the larger portion of shoreline (i.e. 1600 meters of land) on the north and west was studied.

The northeastern corner of the lake was encompassed by a sandy beach; part of the beach was periodically inundated when the water level rose seasonally. Along the shore, detritus, water-lily leaves, and scattered logs, which were partially underwater, sheltered the red-sided and wandering garter snakes and juvenile western painted turtles. The large leech, Haemopis grandis, also used the wet, flat boards as a niche for breeding and the deposition of cocoons in mid-July through August. Scuds (Gammarus sp.) and large balls of blue-green algae (Nostoc sp.) were abundant in this area.

The northwestern shore was composed of various species of grasses, and at least three springs existed along this shoreline. A herd of cattle grazed on these grasses as well as some plants which extended three meters from shore into the water. Scirpus validus, Chara vulgaris, and Sterionema sp. were abundant among the aquatic plant species.

All along the northwestern shore were five very large felled trees, which extended into the water (i.e. up to 7 meters). These trees were used as shelter for the garter snakes; they were seen emerging from beneath them, as well as from under the bark. Other pieces of wood and shrubs gave shelter to the snakes. No snakes were observed emerging from the plentiful, deserted burrows of ground squirrels (Spermophilus columbianus), that lived on the northwestern shore.

The southwestern end of the lake was composed of standing dead trees, many springs, and a grove of poplar trees. Further away from the shore, remnants of a logging camp and stumps of Pinus ponderosa were present. Garter snakes were found at this site, which probably was the dwelling of Peromyscus and Microtus, occasional food items for these snakes.

The waterfowl was quite diverse at this site; the terrestrial birds, such as the members of the fringillid family, seemed to be an integral part of the garter snake's niche. Other parts of their niche included breeding populations of R. pretiosa, R. pipiens, and B. boreas. A few members of the species, H. regilla, were also found. The common slug species (Deroceras), which resided under the logs, also factored into the habitat.

Besides the herd of 20-30 cattle, other disturbances around the lake included occasional visits by deer and horses. The one disturbance that had the most detrimental impact was a man-made one. Local vacationers were observed on occasion to indiscriminately shoot ground squirrels, and presumably any other animals which were large and abundant, such as garter snakes.

The sympatric populations of Thamnophis were studied on June 20, 29, 30; July 2, 3, 14, 15, 17, 30, 31; August 3, 1974; and June 29, 1975.

FRED BURR CREEK

Fred Burr Creek (T6N, R13W, S8 and 9, Granite County), a tributary of Flint Creek, was chosen as a collection site for the study of heavy metals. It has been confirmed as an area that contained fish with a higher-than-normal content of mercury in their tissues (Van Meter 1974). The mercury contamination stemmed from the pan-amalgamation mills, which were active at the turn of the century.

Collection trips were made to the neighboring vicinity of the mill along Fred Burr Creek. This was approximately 5 kilometers from U. S. Highway 10A. Concentrations of mercury were found to increase from the mouth of Fred Burr Creek to where the mill stood (Van Meter, 1974).

Thamnophis sirtalis and *T. elegans* both inhabited the land around the creek. However, the snakes were relatively scarce; the most abundant vertebrate was the spotted frog (*R. pretiosa*). Jumping mice (*Zapus princeps*) were also seen along the creek.

The creek and the mill, which was deteriorating into mounds of brick, were the main areas investigated. The neighboring shrubs and cottonwoods were studied as time permitted, and particular areas with planks of wood and debris were selected as possible shelters. The mill itself always had disturbances by the general public. The citizens were engaged in collecting bricks and boards in great numbers, thus, disrupting the microenvironment for the herpetofauna.

The research trips to this site were made on June 11; July 12; and August 17, 1975.

EAST HELENA

A few areas within a 3.2 kilometer radius from the lead smelter, which was operated by the American Smelting and Refining Company (ASARCO), were selected for the collection of herpetofauna and fish. The smelter has been recognized as the major source of environmental contamination in East Helena; it has emitted effluents which contained high concentrations of arsenic, cadmium, and lead from its stack. These metals have been found within the whole ecosystem (i.e. air, water, soil, plants, animals) (Environmental Protection Agency 1972).

The food chain has not yet been studied; the main concern of past studies has been with herbivores, such as horses and cattle. In fact, Gordon (1972) investigated the uptake of lead and cadmium in several species of rodents, so that the effect from the consumption of native forage would be known. One of the rodents, the vole, has been part of the diet of garter snakes at other sites.

Four areas near East Helena were chosen for the collection of garter snakes for heavy metal analysis. The closest site was a pond (T10N, R3W, S36, Lewis and Clark County) which had an area of 16 hectares. It was an estimated 400 meters from the main stack of the smelter. Rana pretiosa and Thamnophis elegans vagrans were found at this pond. The pond was eutrophic in nature with strong odor of hydrogen sulfide and thick areas of cattails.

On another side of the smelter, a large slag pile was present. Prickly Pear Creek flowed beside this slag pile, and the land between the creek and slag pile supported a diverse community of vegetation. Among these were tall grasses, shrubs, prickly pear cactus, and members of the willow family. T. e. vagrans was found along the banks of the creek.

Approximately 1.6 kilometers south-southeast from the smelter, an irrigation overflow area, some springs, and the banks of the Prickly Pear Creek were surveyed. All of these places were on a private ranch (T19N, R3W, S1, Jefferson County). No snakes were sighted; however, a juvenile sucker (Catostomus macrocheilus) was found in the irrigation overflow, and R. pretiosa were at the springs.

The last area, McClellan Creek Quarry Lake (T9N, R2W, S7, Jefferson County), was approximately 40 hectares in area. It was privately owned and was approximately 3 kilometers southeast of the smelter. A sandy shoreline surrounded the lake, and a marshy region was at the eastern end. The marsh was chosen as the most probable habitat for Thamnophis; both T. e. vagrans and R. pretiosa were collected from this area. Young salmonids were observed in shallow channels of water in the marsh, and the wandering garter snake was caught with a live fish in its mouth.

It cannot be assumed that T. s. parietalis did not occur here because both species have lived together in similar habitats at other sites; and amphibians, which were prey for this species, were present. Frogs and fish were collected for a preliminary food-chain study.

The research trip to East Helena was on July 27, 1975.

McDONALD RESERVOIR

McDonald Reservoir (T19N, R19W, S11, Lake County) was examined as a potential site for the study of the population ecology of Thamnophis on a preliminary trip on June 19, 1974. It did not contain a very large population of Thamnophis; so it was used as a supplementary site in the comparison of levels of heavy metals in garter snakes from northwestern Montana. Two melanistic wandering garter snakes were collected on the floating loggs at the west end of the reservoir.

The collection trip was made on August 5, 1974.

DAHL LAKE

Dahl Lake (T28N, R26W, S24, Flathead County) was examined on the preliminary trip. At this time, one gravid T. elegans vagrans was collected. It was retained for the study of the genetic relationship between the mother and its young. The site itself was not used as a study site.

Collection of this specimen took place on June 20, 1974.

BRENAMEN'S MARSH

Brenamen's Marsh (T31W, R19N, S21, Flathead County) was surveyed as a possible study site. It was not chosen for a population study; however, one _T_. _elegans_ _vagrans_, which was gravid, was collected. It was retained to make a comparison of scale counts between the female and its young. _Chrysemys_ _picta_ _belli_ were also sighted at the marsh. The trip was made on July 31, 1974.

CHAPTER III

ANALYSIS OF HEAVY METALS

INTRODUCTION

In earlier studies ecologists have employed organisms as indicators for the evaluation of environments. Merriam (1894) was one of the first to use vertebrates and plants as indicators of temperature zones. Because snakes have been known to remain in an area no greater than a few kilometers in diameter for their lifetime (Fitch 1949, Hirth et al. 1969), garter snakes were selected in this study to demonstrate the uptake of contaminants from regional sources, and the "normal" content of heavy metals in non-industrial areas.

Both species, *T. sirtalis parietalis* and *T. elegans vagrans*, were used to investigate the levels of mercury and lead contained within the tissues of this predator at specific localities. Two areas of known contamination, Fred Burr Creek for mercury and East Helena for lead, were chosen to contrast to the sevel collection sites (i.e. Woods Pond, McDonald Reservoir, Jette Pond, Jette Lake, Smith Lake, Lake Rogers, and Mud Lake), which were not near a major industry.

Possible influences, however, existed at the collection sites for the introduction of heavy metals within the ecosystem. Although alkylmercury compounds were banned by the E.P.A. in 1971 as a seed dressing, the local uses of such pesticides with mercury, or lead arsenate, have been most difficult to control. Weigand (1973) had surveyed the effect of the ingestion of seed treated with Panogen 15®, a methylmercury fungicide, in north-central Montana; the birds were substantially concentrating this heavy metal in their livers even though their diet was not principally composed of treated seeds. This had serious implications for humans which could ingest an abnormal amount from successive meals of these fowls.

Further probable sources were, in the case of lead, the inorganic emission from the combustion of leaded gas by motorized vehicles (i.e. cars, boats, and snowmobiles (Adams 1975), and errant gun shot in the aquatic systems. In the case of mercury, natural sources such as the volatilization and erosion of mercury-rich soil, or volcanic action, or the use of many available mercury-containing products by man, such as latex paint, have caused the metal to be widely dispersed by air currents and natural waters.

Three approaches to this heavy metal study were emphasized. They were as follows: 1) The analysis of members of both species, which were within a total length range of 40-70 cm, from seven specific sites; 2) the analysis of both species from two known areas of contamination; 3) the analysis of preserved specimens, which were collected from two to twenty years ago, to discover the effect of formalin and ethyl alcohol upon the retention of heavy metals by the specimens. (It should be noted that McDonald Reservoir was chosen as the seventh site because of the scarcity of wandering garter snakes at Woods Pond. Because it was only eight kilometers northwest of Woods Pond, it was used in conjunction with Woods Pond as a typical site for the survey of heavy metals in individuals within this region.)

The essential food types for the maintenance of a population of both species of garter snakes were present at all the collection sites. Therefore, such a migration of up to 17 km by garter snakes in order to find a marsh-like habitat and food items, such as was observed in Canada (Gregory and Stewart 1975), was not expected to occur in this study. Thus, the results were more likely to be regional.

The selection of the two areas of contamination was done after a literature search on heavy metal analyses in Montana. Buhler (1973) indicated several drainages in western Montana which contained relatively high contents of mercury; however, the study of Van Meter (1974) on heavy metals in the Upper Clark Fork River was instrumental in the choice of Fred Burr Creek for analysis of mercury. The studies by the National Academy of Sciences (1972) in conjunction with the Environmental Protection Agency (1972) on lead were principal sources for the selection of the East Helena area.

Both sites were subjected to primarily chronic damage from the continual exposure of heavy metals into the ecosystems. The mercury contamination in Fred Burr Creek originated from early-day mining activities (circa 1881-1905); the pan-amalgamation process for isolating silver was used at this stamp mill. It was this process which was responsible for the dissemination of mercury in the soils, waters, and consequently, fish, in the area of Fred Burr Creek. Mercury in addition to cadmium, copper, lead, and zinc were found in high concentrations in the fish from the recent past to date (Van Meter 1974).

The immediate two-mile radius of the ASARCO smelter was chosen as a collection site in East Helena. The metals, lead, arsenic, cadmium, and zinc, plus the gas, SO_2, have been released into this area since the beginning of the smelting operations in 1888 (Gordon 1973). The animals (domestic and wild) and plants have been suffering from chronic poisoning from the effluents of the smelter's stack; however, only after the metals or sulfur had accumulated to threshold levels in the organisms was the lethal effect noticed by the citizens. After 1968, the ranchers, by their complaints concerning the death of their stock, and the E.P.A. caused the smelter to control its effluents in a reasonable fashion.

In 1970, an induced draft fan and stack heater were added to the 400-foot stack, which was the equivalent of adding 100 feet in height. This had a further dilution effect to the concentration of effluents that fell in the immediate vicinity, and a further dispersion of the effluents into a larger geographic area. It also was instrumental in causing the damage to plants to be primarily chronic rather than acute (Gordon 1973). Therefore, an overall change in the extent of damage to the animals must also be assumed.

In a situation in which chronic poisoning had been experienced, an unnatural selection occurred against the organisms which were unable to exist with an abnormal content of contaminants. In the case of East Helena, some of the original native plants, such as lichens, which were present in 1888, have been eliminated. Lichens have not existed within a six-mile radius of ASARCO for many decades (Gordon 1973). The lack of earthworms in the soil also had been attributed to the high heavy metal content in the top five centimeters of soil.

Under this stress many species disappeared, whereas other species evolved towards more tolerant varieties because of their diverse genetic pool (Gordon 1973). The observance of a paucity of snakes and frogs at East Helena, and the lack of snakes in conjunction with an abundance of frogs at Fred Burr Creek, must have been correlated to this phenomenon. It was the small number of snakes that were found during a long period of search that caused the divergence from the collection of solely garter snakes to the inclusion of frogs and fish in this present study.

The selection of preserved specimens was determined in order that a comparison by species and size with others from the present study could be made. The two sites, Jette Lake and Lake Rogers, were the sources of the garter snakes; they were collected by Brunson in 1949. The two ranids, one from Smith Lake and one from Mud Lake, both of which were preserved as stomach contents from the snakes in this study, were used to contrast heavy metal contents of the ranids from Fred Burr Creek.

METHODS AND MATERIALS

The accepted method of collection of specimens for analysis involved the freezing of specimens in plastic bags at the time of capture (Van Meter 1974, Gordon 1972). In the present study, the specimens were retained in a double layer of plastic bags until they were analyzed. Sections of a blood-containing organ (i.e. usually liver, or if necessary, additions from the kidney and heart) and of voluntary muscles were analyzed from each specimen, unless otherwise indicated. Because the procedure for the determination of lead was different from that of mercury, a part of a blood-containing organ and part of the muscle were divided between the analyses of each metal. Therefore, an average of four chemical tests per frozen specimen were performed. Analyses of the other organs, however, were performed in the case of the samples from the contaminated areas or of the preserved specimens.

Two standard methods of obtaining and preparing a tissue for analysis have been demonstrated to produce reliable results. These methods have been developed with economically valuable species, such as sport fish and game birds. D'Itri (1972) determined total mercury in a fish by obtaining 50 grams of the specimen, and homogenizing a subsample of 10 grams from this muscle, and using approximately 1 gram of the subsequent mixture. Baskett (1975), in his study of birds, and Van Meter (1974), in his study of fish, dissected a known amount, respectively 1-gram and 3-gram samples, of muscle. Subsamples were not used because a uniform distribution of heavy metals was assumed in the muscle and liver. Uniformity of mercury in tissue was determined in one particular study by the Fish and Wildlife Service (1970).

No homogenous subsamples were made in this study; and there was a wide range in sample weights, which were recorded on a wet-weight (w/w) basis, whether or not the sample was frozen or infiltrated with preservatives. Koch and Manning (1973) indicated that the detection of metals by the Atomic Absorption Spectroscopy (AAS) was improved as the sample increased in volume within the limit of practical sample handling. An attempt was made to use nothing lower in weight than 1.0 grams; the mean weight of the samples were 1.6 grams, and the range was 0.99-6.0 grams.

The larger samples were juvenile snakes, that were used in totality. A loss of some of the larger samples, with a range of 3.9-6.0 grams, was experienced because of the amount of gas pressure that built up within the sealed tube during the digestion of the sample caused the tube to explode. Subsequent divisions of large samples prevented further loss of samples. Other undetermined causative agents (such as a flaw in the glassware) caused the loss of smaller samples from explosion.

The samples were procurred from the specimens with sterilized dissection instruments; and care was taken to remove the tissue expediently to prevent any thawing in the case of the frozen specimens. The samples were placed in test tubes after weighing. The tubes, which were 30 cm in length, were made and sealed manually from 25 mm Pyrex tubing.

The digestion procedure, which involved the addition of 15 ml of a mixture of nitric (HNO_3) and sulfuric (H_2SO_4) acids, to the test tubes, and the insertion of the test tubes in a furnace for three hours at 150°C, was followed from the description in Van Meter (1974). The sealed tubes were placed in steel tubing to provide protection from possible explosions that could be caused by the increase of carbon dioxide inside the tubes from the digestion of organic material.

Following the cooling of the samples, the methods differed in the subsequent treatments for the specific metals. These methods, the preparation of reagents and the mode of operation of the AAS, were patterned after Van Meter (1974). The analysis was accomplished by the use of the Beckman Atomic Absorption attachment for the Model DB Spectrophotometer. The cold-vapor method was applied in the measurement of mercury; and the flame aspiration method was used in the measurement of lead.

Blanks, or samples that consisted of reagents and with no known content of heavy metals, and standards of known concentrations in parts per million were interspersed between the analyses of the samples for heavy metals. Standards were made up every two weeks to prevent any change in concentration. Random tests were made to find any source of error in the methods by a second analysis of the same samples for lead, and, different samples of the same type of tissue from the same specimen, for mercurial content.

RESULTS

Collection Sites

Both lead and mercury were present in all the specimens from each one of the seven sites. The mean wet-weight concentrations of lead in the muscles of Thamnophis was .18 w/w ppm (range .02-.40 ppm); in the liver samples, the mean was .12 w/w ppm (range .02-.30 ppm). The mean wet-weight concentrations of mercury in the muscles and liver were correspondingly .26 w/w ppm (range .01-1.0 ppm) and .24 w/w ppm (range .05-.49 ppm). The muscle tissue consistently had a higher content than the liver in most of the individual specimens for both metals (Table 3.1). No comparison was made in the area of specific concentration of heavy metals by female snakes versus males from the same site because of the small sample size; however, in mammals and in rattlesnakes, the tendency of the females to concentrate a higher amount of metals has been substantiated in former studies (Jeffries and French 1972, Bauerele et al. 1975).

A comparison of each site in descending order of use by automobiles, and occasionally boats, was made for each metal. Smith Lake was the most visited site and Jette Lake was the least (Figure A).

Table 3.1—Heavy Metal Concentrations in *Thamnophis* from Collection Sites

Site	Species	Sex	Snout-Vent Length (cm)	Lead (w/w ppm)		Mercury (w/w ppm)	
				Liver	Muscle	Liver	Muscle
Woods Pond	*T. s. parietalis*	M	42.3	.18	.22	.05	—
MacDonald	*T. e. vagrans*	M	48.7	.22	.18	—	.23
		juv.	24.7	.54		—	—
Jette Pond	*T. e. vagrans*	M	47.7	.05	.03	.15	.04
	T. s. parietalis	M	52.6	.02	.02	.49	.28
Jette Lake	*T. e. vagrans*	F	43.8	.03	.13	—	.33
	T. s. parietalis	F	44.2	.07	.03	.2	.3
Smith Lake	*T. e. vagrans*	F	33.6	—	.4	—	1.0
		F	55.3	.03	.03	.20	.09
	T. s. parietalis	M	44.8	.2	.25	.43	.21
Lake Rogers	*T. e. vagrans*	F	43.4	.2	.1	.21	.63
		-	48.4	.1	.15	.47	—
	T. s. parietalis	F	47.5	.2	.25	.15	.16
Mud Lake	*T. e. vagrans*	M	45.9	.3	.3	.09	.11
		M	48.9	.14	.1	—	.01

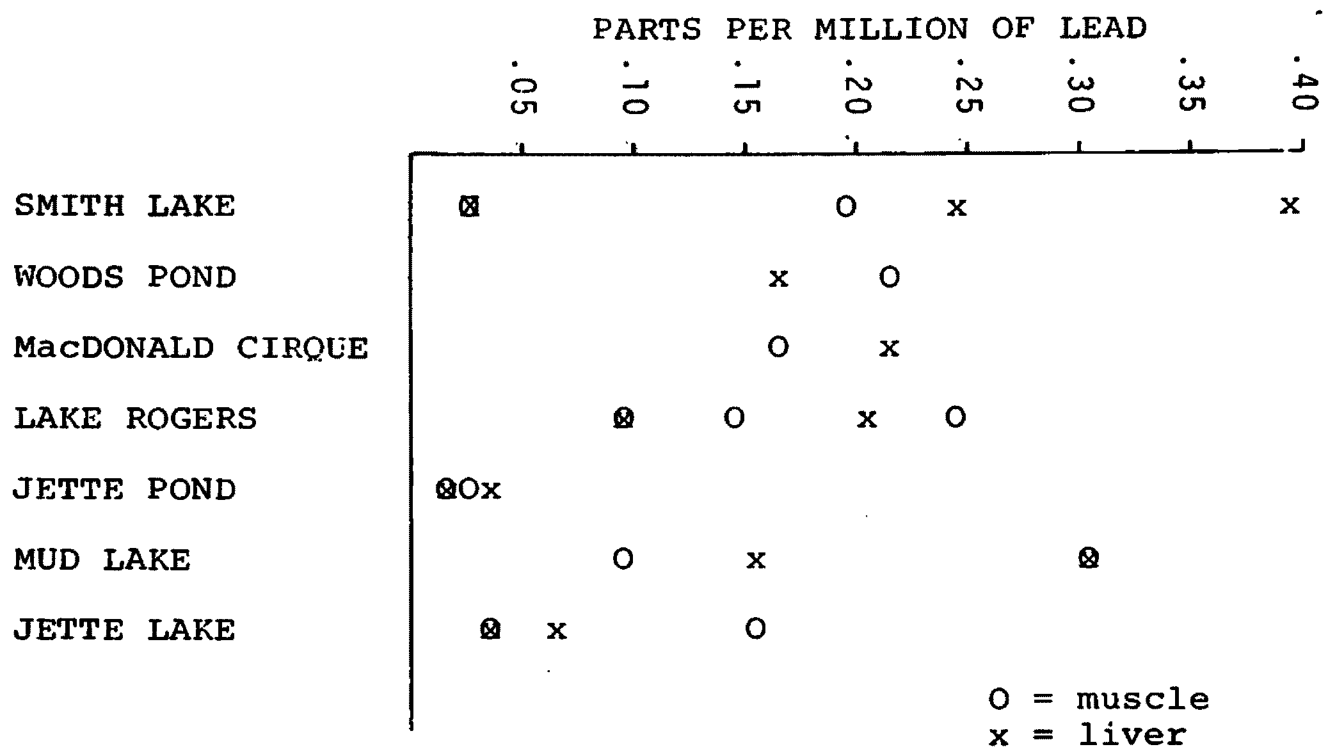
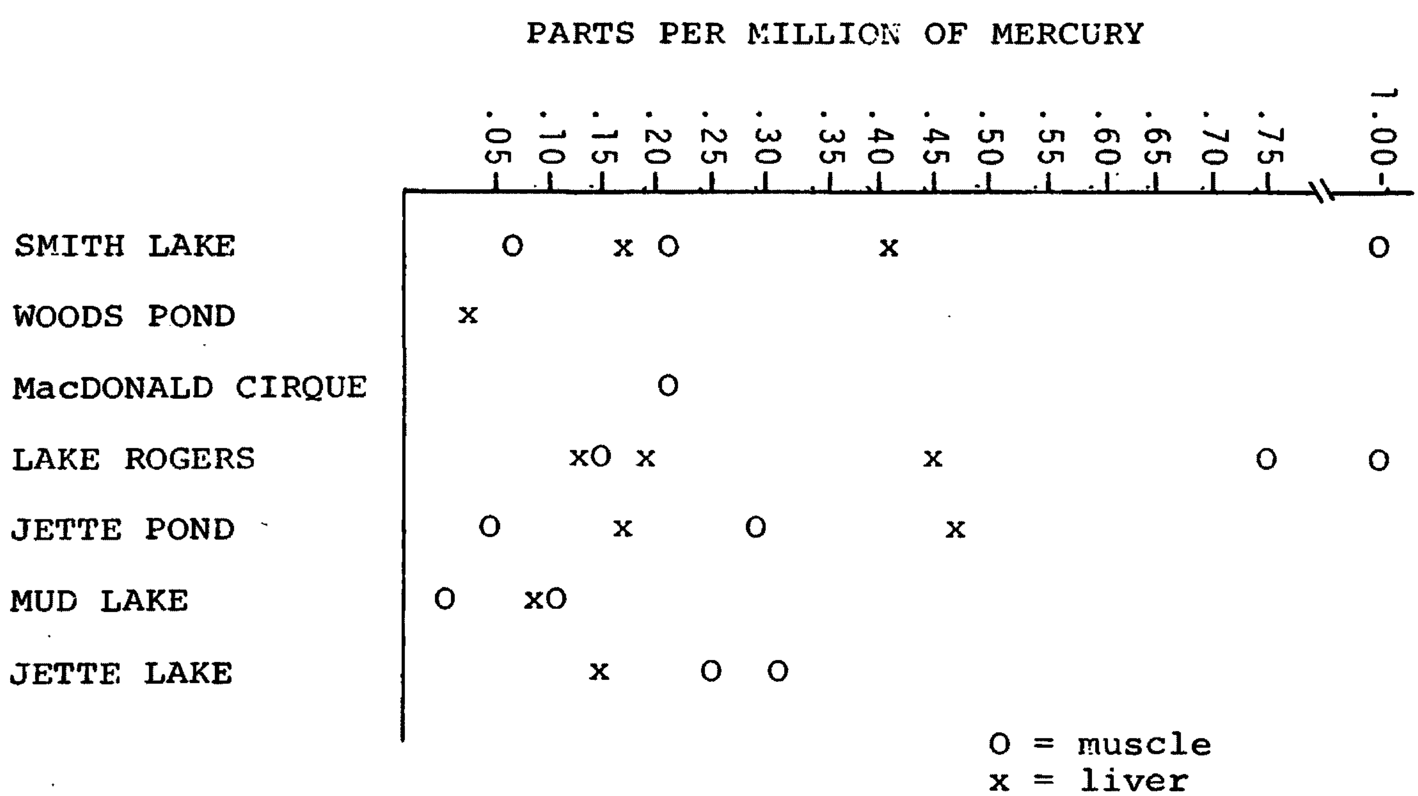


Figure A. Heavy Metals in the Tissues of <u>Thamnophis</u> from Collection Sites.



(The uneven number of samples on the graphs was influenced by the loss of samples by explosions.) Jeffries and French (1972) conducted a similar study in England concerning three species of rodents (i.e. field mice, bank voles, and field voles). They were concerned with the difference in concentration of lead in these mammals of woodland and field areas and those of roadside verges. The habitat most likely to contain the highest concentration was roadside verges, which were directly subjected to exhaust containing lead from passing cars. They found a large difference in the vegetation of the sites, but the mammals from the verges had a mean lead concentration of 2.26 w/w ppm and from the field 1.32 w/w ppm (Jeffries and French 1972). The small differential was surprising; and the field contamination was attributed to rainfall.

"The national average contamination of rain in the United States was 0.36 μg/l and reached as much as 300 μg/l in some localities" (Jeffries and French 1972). This variable should be considered in future studies in northwestern Montana, which experienced more precipitation than other sections of the state. It was unknown whether rain was one of the causes of the occurrence of heavy metals in relatively undisturbed sites.

In the results of the lead analyses, the muscle samples from specimens from Smith Lake had the largest range in ppm (.03-.40), and it also had the highest content observed for muscle. The occurrence of relatively high amounts in the snakes of Mud Lake, which was not affected by motorized vehicles directly, may be a reflection in the drainage of waters from Lake Five, a popular recreation site. Lake Rogers, which did not have motorized boat disturbances, had a similar average in muscle content with Mud Lake (that is, .25 ppm to .20 ppm).

In descending order, the lower average concentrations were found at Woods Pond, McDonald Reservoir, Jette Lake, and Jette Pond. The explanation as to why Jette Pond was the site of the lowest concentrations can only be hypothesized. Although the logging disturbance has been major in the area, the actual lead content was less than expected, even though empty gas tanks and caterpillar tractors were sighted on occasion at the pond site.

A juvenile from McDonald Reservoir was analyzed in total and had a .54 ppm body content of lead. It was interesting to speculate as to whether or not this juvenile had obtained this amount from a subsequent concentration of metal through the placenta of its mother, or from other sources. It was stated in former studies that uptake by animals was not related to trophic levels, but to such factors as metabolic rate and feeding habits (Krenkel _et al_. 1973, Jeffries and French 1972). The observation that recently both snakes tended to increase their feeding period, thereby maximizing their growth rate (Gregory and Stewart 1974) indicated further that in this study a juvenile could have experienced longer exposure through diet to metals in the environment than other "hibernating" members of the population.

In respect to the mercury concentrations, Smith Lake again was the area at which the highest concentration (1.0 w/w ppm) was found in the muscle tissue (Figure A). There was a wide difference in concentrations (between individual snakes and between different tissues of each snake) found at Lake Rogers and Jette Pond; and a much less deviation in the samples at Mud Lake and Jette Lake (Table 3.1).

As referred to in the introduction of this chapter, many sources existed to cause the presence of mercury; but even in a recent study on background levels of mercury at a remote sector of the Great Smoky Mountains National Park in 1973, mercury was found in the range of .017 to .098 ppm in fish (Fulkerson et al. 1974). Therefore, the probable natural occurrence of mercury in the soils in this incidence, as in the other study, had an inestimable influence at these seven sites. The environment in which the organism lived, such as aquatic or terrestrial, has been concluded to have much bearing in the case of mercury from a study of Fred Burr Creek. (See next section.)

Fred Burr

Two snakes and four frogs from this area had specific tissue samples from their bodies analyzed for mercury content (Table 3.2). The T. e. vagrans was analyzed in entirety because it was a juvenile snake; however, four samples were dissected from the gravid T. s. parietalis. These samples included a muscle section, the liver, the anterior positioned egg and the posterior positioned egg. These two eggs were selected from the 17 eggs in order to show any difference in the least developed egg (anteriad) and the most developed egg. A tissue assortment consisting of the head (including the brain) and the liver was dissected from each ranid.

The samples from the snakes contained a high percentage of mercury. (The two eggs and liver of the gravid T. s. parietalis were not analyzed because the samples exploded during the digestion process in the furnace.) The carcass of the juvenile snake had .77 ppm Hg, and the muscle of the

Table 3.2—Data from Herpetofauna at Fred Burr Creek

Species	Length (cm)	Wet Weight (g)	Tissue Samples (ppm Hg)
Thamnophis sirtalis	72.9 (s.v.)*	270.4	.39
Thamnophis elegans	23.1 (s.v.)	5.0	.77
Rana pretiosa	9.9 (t.)+	3.8	2.58
	14.5 (t.)	12.7	2.02
	14.2 (t.)	16.5	2.88
	16.1 (t.)	22.7	1.46

*s.v. = snout-vent

+t. = total

gravid female had .39 ppm. The samples, which were conglomerates of liver and brain tissues, from the frogs were much greater in content; they were as follows: 1.46, 2.02, 2.58, 2.88 ppm. The mean of the mercury content from the ranids was 2.27 ppm.

Comparisons were made graphically between these samples and samples from areas of less contamination (Figure B). The comparisons were based upon similar lengths, and if possible, between the same species. One frozen and two recently preserved juveniles, which were from McDonald Reservoir and Lake Rogers, were all lower in mercury content than the juvenile from Fred Burr. This juvenile had 8.5 times more mercury than the mean ($\bar{X}$ = .09 w/w ppm) of the other three juveniles.

The three adults, which were from Smith Lake, Jette Pond, and Jette Lake, were chosen because they were the largest snakes which were analyzed for mercury. Only the individual from Jette Lake, however, was

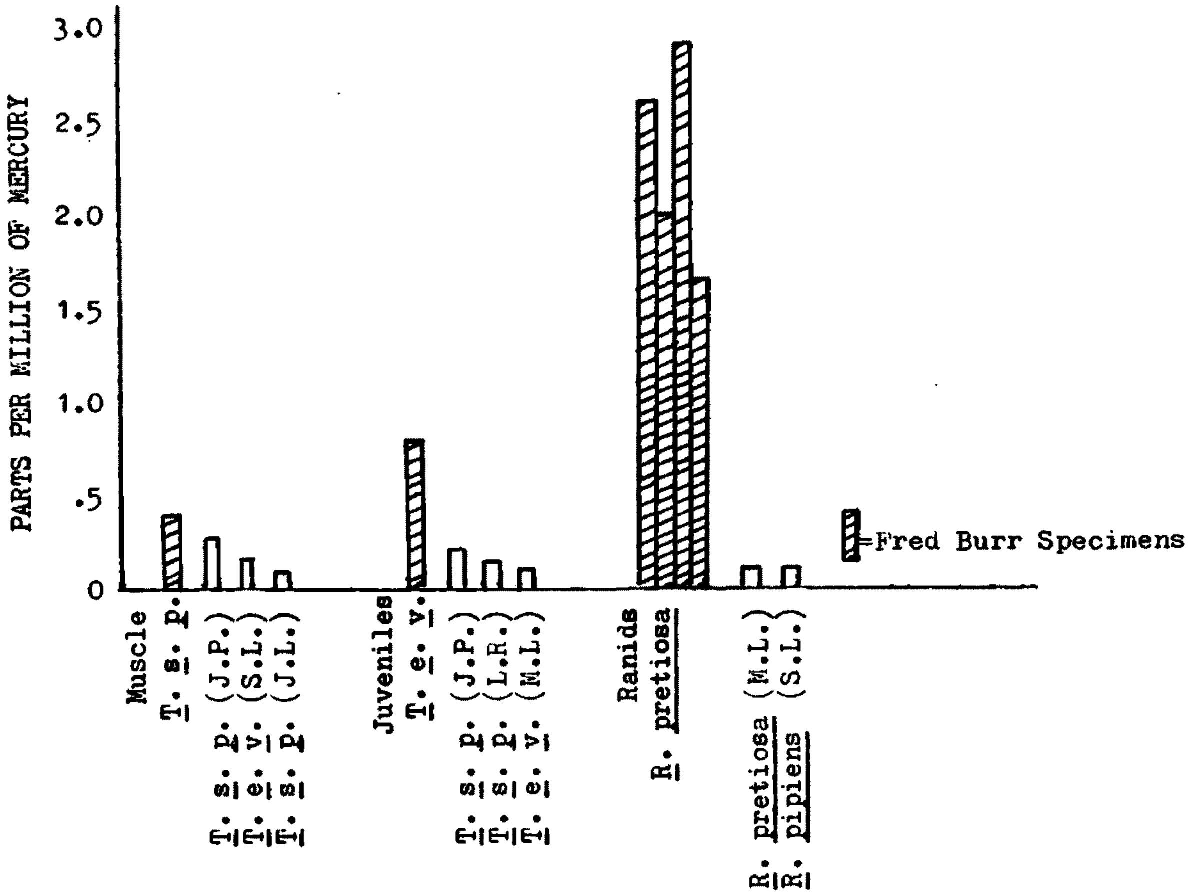


Figure B. Mercuric Concentration in Fred Burr Samples and in Other Similar Samples

Types of Tissue	Site	Species	Length	Wet Weight (g)	PPM (Hg)
MUSCLE	Fred Burr	T. s. p.	72.9 (s.v.)	270.4	.39
	Jette Pond	T. s. p.	52.6 (s.v.)	62.17	.28
	Smith Lake	T. e. v.	55.3 (s.v.)	----	.09
	Jette Lake	T. s. p.	64.0 (s.v.)	91.36	.05*
CARCASS OF JUVENILE	Fred Burr	T. e. v.	23.1 (s.v.)	5.0	.77
	Lake Rogers	T. s. p.	20.2 (s.v.)	5.72	.23*
		T. s. p.	23.0 (s.v.)	4.73	.04*
	MacDonald Cirque	T. e. v.	24.1 (s.v.)	----	.01
RANIDS	Fred Burr	R. pretiosa	9.9 (t.)	3.8	2.58
		R. pretiosa	14.5 (t.)	12.7	2.02
		R. pretiosa	14.2 (t.)	16.5	2.88
		R. pretiosa	16.1 (t.)	22.7	1.46
	Mud Lake	R. pretiosa	15.0 (t.)	18.5	.04*
	Smith Lake	R. pipiens	10.3 (t.)	6.9	.04*

*preserved specimens

the closest in size to the gravid T. s. parietalis. The range of the muscle content of the three snakes was .05-.28 ppm, and the mean was .14 ppm. The muscle from the T. s. parietalis from Fred Burr Creek was greater in concentration by a factor of 2.7.

Two ranids, one from each species, were compared to the ranids of Fred Burr Creek. Both frogs had the same amount of mercury (.04 ppm), although one was from Mud Lake and one from Smith Lake. The mean (2.27 ppm) of the four Fred Burr frogs was 57 times more than the mean of these two frogs. One note of interest was that the first two of the Fred Burr frogs, which were collected about one-half mile below the mill, had a mean concentration that was less than that of the two frogs from the mill's vicinity. These means were, respectively, 2.17 ppm and 2.30 ppm.

Van Meter (1974) analyzed trout muscle, and water samples at four different places along Fred Burr Creek. His results have been correlated with the samples collected in this study (Table 3.3). A distinct trend was demonstrated from his data. As the samples of water and rainbow trout were collected closer to the origin of mercury dissemination (i.e. the stamp mill), the mercury content became higher. This may be the partial cause for the difference in averages bewteen the four frogs, that were collected at two places.

The range of the mercury in the rainbow trout from Fred Burr Creek was .27-1.92; the mean was .56 ppm. These data were compared to two studies that were establishing background data for the rainbow trout, Salmo gairdneri. In Lake Powell, which was influenced by the natural mercury in the surrounding soil, rainbow and brook trout were considered in the same category. The mercury content ranged .07-.10 ppm, the mean

Table 3.3—Mercury Gradations at Fred Burr Creek (in ppm)

Number of Samples	Water Concentration*	*Salmo gairdneri** N	Range	Mean	*Rana pretiosa* N	Range	Mean	*Thamnophis sirtalis* N	Range	Mean	*Thamnophis elegans*+ N	Range	Mean
1[a]	.0022	9	.25-.45	(.34)	-	—	—	-	—	—	-	—	—
1[a]	.0044	7	.32-.67	(.43)	-	—	—	-	—	—	-	—	—
-[a]	—	-	—	—	2	1.46-2.88	(2.17)	-	—	—	-	—	—
1[b]	.1200	4	.67-1.92	(1.26)	2	2.02-2.58	(2.30)	1		.39	1		.77
1[c]	.0031	-	—	—	-	—	—	-	—	—	-	—	—

*Data from Van Meter (1974).

+*T. e. v.* carcass of juvenile analyzed in total.

[a] ½ mile in distance, approaching the mill.

[b] At the mill.

[c] Above the mill.

was .08 ppm (Potter et al. 1975). In the Great Smoky Mountains National Park, S. gairdneri had a range of .029-.098, the mean was .045 (Fulkerson et al. 1974). The difference between the fish from Fred Burr Creek and these two studies were not as great in porportion as the difference between the ranids in this present study. Permeability of amphibian skin to water and the elements in the water could have caused this distinction between fish and frogs.

Another contrasting phenomenon was observed in the samples from Fred Burr Creek. The aquatic vertebrates were higher in concentration than the terrestrial forms at the mill site; therefore, it was evident that there existed a connection between this occurrence and the high concentration in the water at this site. Fulkerson et al. (1974) concluded from food chain studies that there were three types of exposure for a fish. One was the adsorption of mercury on the mucous layer on the skin, another from the ingestion of food, and lastly, the assimilation of mercury into the bloodstream as a consequence of digestion. These exposures were probably valid for amphibians, in particular the spotted frog at Fred Burr Creek.

East Helena

Tissues from three wandering garter snakes, four spotted frogs, and one sucker were analyzed for lead. The range and mean of the wet-weight concentration of lead in parts per million were as follows: garter snakes, muscle: .36-.8 (mean: .55), liver: 0.1-.7 (mean: .39); frogs, tissue: .43-4.8 (mean: 1.58), liver: 0.1; sucker, tissue: .94. (It must be noted that the largest ranid had a liver with the weight of approximately 1.0 gram, which was the minimal weight necessary for analysis. Therefore, this ranid was sampled in two parts.) The muscle samples of the *Thamnophis* displayed a greater concentration of lead than the liver samples at this site (Table 3.4).

The fish and the two frogs which were collected on a ranch about a mile southeast of the stack were dissimilar in content. However, the frog with the highest concentration (4.8 ppm) was found at this site. The other frog which was similar in size was less than one-tenth (.46 ppm) of the concentration of the other. The relevance of this difference can not be analyzed without more samples; the sucker, however, with the concentration of .94 ppm was probably exemplary of other suckers in its size range. Suckers, which were on the lower trophic level in an aquatic food chain, have been found to consistently contain a lower amount of heavy metals than other species, such as trout, which were at the highest trophic level (Potter *et al*. 1975, Fulkerson *et al*. 1974).

No obvious correlation was obtained from the analysis of ranids in the concentration of lead and the distance from the ASARCO stack. The two frogs from the site furthest away (i.e. Quarry Lake) were relatively high in tissue, .43 and .66 ppm. (It should be noted that the liver [0.1 ppm] of the larger of the two frogs was significantly less in concentration than the tissue [.66 ppm].) However, the one snake, which was collected at this lake, was the lowest in concentration of both liver and muscle samples among the snakes.

A survey which was done on another food item of garter snakes, that of mice (i.e. conglomerate term including *Peromyscus*, *Microtus* and *Mus*), showed a definite accumulation in the kidney of the mice (Gordon 1972).

Table 3.4—Data from Specimens Collected at East Helena

Locus	Species	Length (cm)	Wet Weight (g)	Tissues (ppm Pb)		
				Liver	Muscle	Piece
T10N, R3W, S36 Lewis & Clark County	*Thamnophis elegans*	47.5 (s.v.)*	57.7	.36	.5	—
	Thamnophis elegans	38.2 (s.v.)	19.3	.7	.8	—
T9N, R3W, S1 Jefferson County	*Castastomus macrochelus*	12.0 (t.)+	19.1	—	—	.94
	Rana pretiosa	11.5 (t.)	7.8	—	—	4.8
	Rana pretiosa	10.5 (t.)	7.1	—	—	.46
T9N, R2W, S7 Jefferson County	*Thamnophis elegans*	41.1 (s.v.)	25.9	.1	.36	—
	Rana pretiosa	17.8 (t.)	33.9	.1	—	.66
	Rana pretiosa	13.0 (t.)	11.2	—	—	.43

*s.v. = snout-vent

+t. = total

The mice, which were collected within a radius of a few miles of the stack, had a range of lead in their kidneys of 0.5-110.0 ppm (mean: 42.5 ppm). Their kidneys were several times higher in concentration than the livers, which ranged 0.5-23.0 ppm (mean: 7.6 ppm).

The difference of these mice from the control specimens was substantial. The mean concentration of lead in the twelve control specimens was 0.5 ppm for kidneys, and 2.0 ppm for livers (Gordon 1972). The reason for the occurrence of greater concentrations of lead in the liver rather than the kidneys of the control mice was not resolved. However, the analysis of a large quantity (i.e. 81 specimens) of mice from the smelter clearly demonstrated the tendency for kidneys to concentrate lead in a stress environment.

The contrast between the means of the tissue concentrations of the mice and their predators, garter snakes, could be partially explained by the food study of _T. e. vagrans_ (see Chapter IV, Table 4.6). The rodent played a minor role in the percentage of food ingested by the wandering garter snake; amphibians and fish, contrastingly, played more important roles.

Therefore, in environments such as from the two contaminated areas in this study, the indigenous animals through their diets have been exposed to a sub-lethal, and, in many cases, non-affecting levels (Gordon 1973). In the predator, _Thamnophis_, the rate at which the ingestion of the metal occurred and subsequent losses through the deterioration of the metal itself (half-life) in the animal and fecal elimination, must be directly related to the seasonal emphasis of prey.

Preserved Specimens

Seven garter snakes and two ranids were analyzed for the concentration of heavy metals within their tissues. Care was taken to correlate approximate sizes between the recently preserved specimens and those collected in 1949 by Brunson (Table 3.5). This initial investigation into the effect that time and preservatives had upon the heavy metal concentrations in herpetofauna was influenced because of two reasons. These were that (1) a large amount of data existed in major repositories for biological specimens which were collected from known localities and which had not been sufficiently utilized, and (2) the need to minimize the collecting from areas with relative little pollution disturbance, if background data could be obtained from museum samples.

Feathers and eggshells have been used successfully in the analysis of polychlorinated biphenyls and mercury in Sweden (Rick 1975). These, however, were not kept in preservatives. The controversy of the effects of alcohol and formalin upon museum specimens had its origins in studies of fish tissues.

Evans _et al_. (1972) were the first Americans to make an extensive use of museum fish (i.e. 57 specimens) for determining the total mercury levels in the muscle tissue of fish from the Lake Erie-Lake St. Clair region of Ontario. Seventy percent of the preserved fish collected between 1920 and 1965 contained total mercury levels of less than 0.20 ppm; these figures correlated directly with current data taken of fish from uncontaminated areas of the Great Lakes.

Table 3.5--Concentrations of Heavy Metals in Preserved Specimens

Metal	Site	Date	Species	Total Length (cm)	Tissue Analyzed	Wet Weight (ppm)
MERCURY	Jette Lake	1-8-49	T. s. p.	76.0	muscle	.05
		12-7-74	T. e. v.	59.3	muscle	.07
	Lake Rogers	20-6-74	T. s. p.	30.5	entire body	.04
		16-7-49	T. s. p.	26.3	entire body	.23
	Smith Lake	3-8-74	R. pipiens	10.3	anteriad portion	.04
	Mud Lake	15-7-74	R. pretiosa	15.0	anteriad portion	.04
LEAD	Jette Lake	1-8-49	T. s. p.	76.0	liver	.25
		19-6-74	T. s. p.	60.0	liver	.30
	Lake Rogers	16-7-49	T. s. p.	28.5	entire body	exploded
		20-6-74	T. s. p.	26.2	entire body	.40

Another optomistic study on recent and 90-year-old benthopelagic fish by Barber and Cross (1972) indicated that although the mercury content of the water in the deep-ocean habitat had changed, the mercury concentrations in the two species had not changed. Contrastingly, a well-organized study specifically on the effects of preservatives and time by Gibbs et al. (1974) caused considerable doubt to exist on any study based on preserved material.

Specimens of fish which were preserved for one month in formalin, ethyl alcohol, and isopropyl alcohol had several various changes occur in their heavy metal content (Gibbs et al. 1974). These preserved specimens had higher cadmium, copper, zinc, and sometimes lead, and lower concentrations of mercury and sometimes lead, than did unpreserved frozen fish. Metal tags (i.e. tin) were the suspected cause of higher maximum concentrations than that caused by the preservatives.

With particular reference to lead and mercury, Gibbs et al. (1974) found that in specimens, which had been preserved for two to more than eighty-five years, lead did not appear to increase after a month in preservative; mercury increased for at least thirteen years. An anomalously high lead content that was found in the two oldest samples was attributed to the fact that the old specimens were fixed in alcohol. Alcohol did not harden the tissue to the extent that formalin did, therefore a more complete digestion of the sample occurred before analysis by the AAS.

In the present study, the recent and old museum specimens were hardened in formalin, subsequently rinsed in water, and preserved in alcohol. Aluminum tags were used with the old specimens. The analyses of the recent specimens were performed approximately 22 months after the collections were made. The specimens were measured for their wet weights, and thoroughly air-dried before analyses. The explosive nature was enhanced by the presence of preservatives, and although precautions were taken, one sample was lost (Table 3.5).

The results were compared between preserved samples and unpreserved frozen samples. The mercury content in the muscles of the preserved snakes from Jette Lake did not differ to a large extent; but the content of the preserved juveniles, which were analyzed in entirety, from Lake Rogers were quite different (Table 3.5). The mercury concentrations of samples from Jette Lake were less than that of the two muscle samples from unpreserved specimens (.3, .33 w/w ppm Hg). The relatively low concentrations were accepted to be correct in this case because of the presence of other samples with concentrations of less than .10 w/w ppm and the lack of an obvious increment of mercury in the 27-year-old specimen.

The concentration in both preserved juveniles was questionable, because neither displayed a considerably high differential between their content and that of the unpreserved adults of the population. (The juveniles from McDonald Reservoir and Fred Burr Creek were much higher in concentration of mercury than the adult specimens.) The fact that the 1949 specimen, in this case, had a greater concentration, which was not to be expected from an unpolluted era, was probably due to the phenomenon of concentration observed by Gibbs et al. (1974).

The low concentrations of the two ranids must be looked upon with some skepticism, also. At both sites, especially Smith Lake, unnatural sources for mercury were present from man's recreational activities. And, because the frogs at Fred Burr demonstrated a tendency to concentrate mercury, these frogs were expected to be higher than the snakes from the corresponding sites. The only consistent corollary was the fact that Mud Lake had concentrations of mercury in the unpreserved snakes ranging from .01-.11 w/w ppm Hg, and the concentration of the frog would be predicted as on the lower end of the stratum also.

In the lead analyses, the two specimens from Jette Lake (circa 1949—.25 w/w ppm; circa 1974—.30 w/w ppm) were considerably greater in concentration than the unpreserved frozen specimens (.02, and .07 w/w ppm). The effect of time in the preservatives was not obvious between 1949 and 1974 specimens. The range in the difference between the preserved and unpreserved specimens was inexplainable.

The juvenile from Lake Rogers was relatively high in its body content of lead, .40 w/w ppm. It was higher than any of the muscle or liver readings of the unpreserved snakes (Table 3.1). Because the other sample from 1949 was irreparably lost, no more theorizing was done on juveniles from Lake Rogers.

Various interpretations from this survey conveyed the fact that the validity in the use of preserved specimens was questionable. If a larger usage of these specimens was encouraged, a better understanding about their usage as a resource for data on heavy metals would be obtained.

CHAPTER IV

ECOLOGICAL ISOLATION MECHANISMS

INTRODUCTION

Many aspects of the ecology of T. sirtalis parietalis and T. elegans vagrans have caused the coexistence of these species in the same habitat. The selection of specific prey and the tolerance to ambient temperature levels have been the mechanisms that demonstrated two facets of ecological isolation in other sympatric populations of Thamnophis (Carpenter 1952, Fleharty 1967). These two categories were used to display differences between the red-sided and wandering garter snakes in this study. Specific collection sites were chosen to investigate the interspecific competition; these sites were chosen from a preliminary survey that was done in the early summer of 1974. The sites were the following: Woods Pond, Jette Pond, Jette Lake, Mud Lake, Smith Lake, and Lake Rogers.

The selection of food items by each species was based on samples which were procured in the field. Each snake was examined for its stomach content in order to obtain the frequency of occurrence of specific food types. The snakes were forced to regurgitate their stomach contents by a manual method of palpitation. This method has been used in other ecological studies (Carpenter 1952, Gregory and Stewart 1975); and in this study, care was taken in particular with garter snakes, which either were gravid or had swallowed a large food item.

In the analysis of temperature relationships, three parameters were measured. These were the cloacal temperature of the snake, the substrate on which it was found, and the air temperature. Cloacal temperatures have been found to demonstrate a direct relationship to the ground temperatures, which, in turn, were affected by the air temperatures. In order to prevent the false reading of cloacal temperatures, which was observed to occur by handling a snake for a duration of time, and thereby raising its temperature (Benedict 1932), these temperatures were recorded immediately upon capture. An aneroid barometer was used as a supplementary instrument to analyze the presence or absence of specific activities of the snakes, under prevailing weather conditions.

The analyses and results of the study of the two facets of garter snake ecology have been treated in the following order: (1) food study, and (2) temperature relationships. Tables have been included with the discussion of each site concerning food items; and in the conclusion, a supplementary table as well as those considering the species as a whole have been constructed. In the temperature section, one table with temperature and barometric measurements, individual site graphs, and a graph concerning each species as a whole have been incorporated with the discussion.

WOODS POND

Twenty-eight red-sided garter snakes and two wandering garter snakes were examined in the field for stomach content. Wandering garter snakes were seen on the first visit to the site, and both had empty stomachs. Consequently, the periodic feeding of the other species was studied, although only seven (25 percent) of these snakes that were captured had eaten any prey during the summer. (Seasons of the summer were arbitrarily broken into three categories: early summer (June 22-July 22), mid-summer (July 22-August 22), and late summer (August 22-September 23).)

In the early summer, five snakes were found in the process of ecdysis. All had empty stomachs and were emerging from the same brushy area. It was concluded that this area was a hibernaculum, and that the snakes had not yet begun to feed. A correlation of emptiness in the stomach, ecdysis, and a similar source of emergence was noted only at this site.

Leeches (Erpobdella punctata) occasionally were eaten in the summer; the metamorphosing long-toed salamanders (Ambystoma macrodactylum) were the food items which were eaten by the greatest number of snakes in mid-summer. Three of the larvae were eaten by a juvenile snake. The other resource was the earthworm (Lumbricus sp.), which was consumed by a juvenile (Table 4.1).

In the study of ecology, a predator species that was not subjected to competition became a generalist (i.e. it had a wide range of food types). At this site, interspecific competition was not observed; but the expected broad utilization of food resources was not noticed. The utilization of other foods (i.e. frogs, rodents, birds, slugs, et cetera) was not discovered. Therefore, the conclusion that several members of the population had migrated elsewhere for food was made because of this low diversity. The census that was conducted on the resident population and was reported in the following chapter reaffirmed this conclusion.

Table 4.1—Correlation of Prey Items of *Thamnophis* and Specific Habitats (Woods Pond, Mud Lake, and Smith Lake)

Site	Species	Number of Individuals	Snout-Vent Size Range (cm)	Prey Items
Woods Pond	*T. s. parietalis*	2	37.7-53.6	*Erpobdella punctata*
		4	26.9-54.6	*Ambystema macrodactylum*
		1	18.5	*Lumbricus sp.*
Mud Lake	*T. e. vagrans*	2	48.3-48.9	*Salmo sp.*
		11	27.4-56.9	*Prophysaon sp.*
		1	60.0	*Rana pretiosa*
Smith Lake	*T. e. vagrans*	1	29.4	*Richardsonius balteatus*
		1	48.1	*Haemopis grandis*
		1	48.3	*H. grandis* and *Rana pipiens*
		3	24.7-55.3	*Bufo boreas*
	T. s. parietalis	2	41.8-53.1	*R. pipiens* and *B. boreas*
		1	44.3	*H. grandis* and *B. boreas*
		3	48.5-64.7	*B. boreas*
		1	44.8	amphibian parts

No significant difference in diet was noted in the diets of the juveniles and adults.

MUD LAKE

Nineteen wandering garter snakes were examined; 14 (74 percent) had food in their stomachs. No red-sided garter snakes were sighted at the stream or lake; this site was comparable to Woods Pond in the scarcity of the second species. Three food resources were exploited as in the above case, although these snakes were more selective (Table 4.1). Eleven of the snakes that had eaten contained the slug, Prophysaon sp. The slugs typically were eaten in quantity; three individuals had two or less, and eight ranged from 5-39 slugs each. No size correlation was noted; large and small individuals ate this food resource.

The snakes were found under the same logs as the slugs. Thus, the possibility that the garter snakes were opportunistic also existed. This advantage of having food in such close proximity could have become a selective process by this species over many generations.

Salmonids were eaten in the early summer. On one occasion, at the southwestern end of the lake, a large female snake, which had ingested a spotted frog from the posterior end, was found. This was unusual because spotted frogs inhabited the area of the freshwater spring, which was a considerable distance from the southwestern end of the lake. The ranid, that was common in the lake proper, was the leopard frog. Larval forms were the usual amphibian source consumed at other sites.

The relatively small size of the population could have prevented specialization to exist between the adult and juvenile snakes.

SMITH LAKE

At this site four prey items were utilized by both species; however, combinations of three of these prey items were common in both species (Table 4.1). Three (33 percent) of the T. e. vagrans were empty, as were seven (50 percent) of the T. s. parietalis. The most consumed prey in both species were the newly metamorphosed toadlets (B. boreas); one red-sided garter snake was found so bloated that it was suspected of being gravid. However, upon examination, it contained 38 toadlets.

Both species of snakes were riparian in their feeding behavior; R. pipiens tadpoles were eaten in the early and mid-summer periods. The large leech, H. grandis, was not a major part of the diet; but it was found singly as well as with other food items in the stomachs of both species.

The T. e. vagrans demonstrated specialization in one resource area; a shiner (Richardsonius balteatus) was found in one member of this species. At Jette Lake, the selection of fish by this species was most pronounced. The occurrence of fish in T. e. vagrans and its absence in T. s. parietalis at Jette Lake and Smith Lake, and its consumption at Mud Lake, indicated that this was a potential resource that could be exploited by the wandering garter snake, if the species was placed under stress.

An absence of the slug, Deroceras, from the diet of either species was observed at this site. The slug was collected under logs, which had sheltered garter snakes. This resource was used by the wandering garter snake at Mud Lake and at Lake Rogers to a large degree; however, it also was eaten by the red-sided garter snake at Lake Rogers.

The food ingested by both species was indicative of the same microhabitats, the riparian environment. This region was well used by anglers hunting for earthworms and frogs; and the terrestrial habitat (i.e. areas with trees and brush) was more severely disturbed, on the level of the garter snakes, by vehicular traffic and cattle. The degree to which the disturbances affected the reptilian use of the riparian versus the terrestrial habitat was difficult to assess.

The effect of the disturbance on the size of the population at this site will be discussed in the following chapter; it has been speculated that the aftermath of these disturbances has been reflected in the food selectivity of the two species. The disturbances could have created an atmosphere in which both species were scarce enough in numbers that they have existed well on the same diet without need of specialization.

JETTE POND

Thirty-eight specimens in total were examined at this site; in *T. s. parietalis*, five (27 percent) of the 18 snakes had empty stomachs. In the other species, six (30 percent) of the 20 snakes had not eaten recently at the time of capture. Seven food items were consumed by *Thamnophis*, four of these foods were common to both species (Table 4.2).

Although a high percentage (31 percent) of *T. s. parietalis* with stomach contents had eaten the long-toed salamander, this category was closely followed in percentage (23 percent) by two other items, toadlets and the small leech species. Toadlets were the only terrestrial form consumed by this species, and the red-sided garter snake had a well-balanced diet of all four types of amphibians that were present at the site.

Table 4.2—Correlation of Prey Items of Thamnophis and Specific Habitats (Jette Pond and Jette Lake)

Site	Species	Number of Individuals	Snout-Vent Size Range (cm)	Prey Items
Jette Pond	T. e. vagrans	2	28.2–31.7	B. boreas
		4	27.6–36.6	E. punctata
		2	24.3–29.8	A. macrodactylum
		1	38.3	Hyla regilla, E. punctata, A. macrodactylum
		1	47.7	E. punctata, A. macrodactylum, H. grandis
		1	29.4	E. punctata and B. boreas
		1	28.6	E. punctata, H. regilla and slug parts
		1	45.4	E. punctata, B. boreas, A. macrodactylum
	T. s. parietalis	3	17.7–23.1	B. boreas
		3	21.8–49.8	E. punctata
		4	18.8–51.6	A. macrodactylum
		1	30.9	H. regilla
		1	28.6	R. pretiosa
		1	60.3	E. punctata and B. boreas
Jette Lake	T. e. vagrans	8	26.8–49.4	H. grandis
		3	20.7–22.9	E. punctata
		2	19.5–24.7	Deroceras sp.
		6	23.2–56.9	R. balteatus
		1	25.1	B. boreas
	T. s. parietalis	2	27.4–30.3	H. grandis
		6	15.2–52.4	E. punctata
		2	22.2–37.1	Lumbricus sp.

The wandering garter snake displayed a large distribution of food resources, also. However, the small leech was eaten in the highest percentage (29 percent); it was followed by toadlets and salamanders both at the same percentage (14 percent). This correlated with the red-sided garter snake which also preyed most heavily on these three forms.

This fact would have indicated direct competition; however, data on the wandering garter snake indicated that a ecological compromise was occurring on the part of this species. It was becoming an opportunistic predator; it ate the most available prey items. This was demonstrated by the presence of five (36 percent) of these snakes with different combinations of two or three prey items (Table 4.2). (Only one [8 percent] of the red-sided garter snakes had eaten a combination of two prey items.)

In two of the combinations of three prey items, food resources were present that were absent in the red-sided garter snake (Table 4.2). These two items were two slugs and the large leech, _Haemopis grandis_. (The slugs were represented by the shells only and, therefore, were not classified to species.) The occurrence of these in two _T. e. vagrans_ and the occurrence of _R. pretiosa_ in one _T. s. parietalis_ indicated that specialization in habitat niches may also have occurred. The questionable aspect of this phenomenon of specialization was that only a small number of each species exhibited the select food items.

The high percentage of amphibians in the categories of food items of both species was a product of the immediate environment. Fifty-seven percent of the food categories selected by wandering garter snakes were amphibians; and 80 percent were selected by the red-sided garter snake. This immediate environment was unusual in the fact that it was a breeding site for treefrogs, salamanders, toads, and spotted frogs. The presence of adult members of all these amphibians suggested that these populations of amphibians were stable. Therefore, the combined effect of continual presence of amphibians, and the relative small size of the pond (perimeter: 40 meters) were instrumental in causing the high frequency of these food resources in Thamnophis.

The smaller snakes of both species were selective in the size of prey. Toadlets, metamorphosing salamanders, and the small leech species were eaten in the red-sided garter snakes, with a snout-vent (s.v.) length of 22 cm and less. Fewer individuals of the wandering garter snake species were found in the same size category; however, those that were found contained metamorphosing salamanders and plant material. The plant material must have been ingested instead of the prey item that had escaped.

The three food categories mentioned in the preceding paragraph were not unique to juveniles. In fact, a combination of two types of small prey, toadlets and small leech species, were present in one individual of each species. The larger snake, 60.3 cm in s.v. length, had seven toadlets and one leech; the small snake, 29.4 cm in s.v. length, had one toadlet and two leeches. Thus, the large snake, which had a larger capacity, ate a larger quantity of the same prey items. However, a wide range of sizes of snake were present for those that ate the other combinations or individual foods (Table 4.2).

Therefore, the coexistence of the two species of snakes was feasible in this environment regardless of the overlap of specific food items because of three tendencies of food selectivity: 1) the juveniles ate small items, 2) the red-sided garter snakes ate predominantly amphibians, and 3) the wandering garter snakes ate a larger variety and combination of prey than the red-sided garter snakes.

JETTE LAKE

At this site, six food resources were utilized by the two species of garter snakes; two of these resources were common to both species (Table 4.2). Leeches, Haemopis and Erpobdella, were found in over 50 percent of the individuals which had eaten; although in the red-sided garter snake, the percentage (73 percent) was larger than the percentage (55 percent) in the wandering garter snake. The percentage of examined snakes which had empty stomachs was coincidentally similar, such that 50 percent of the T. s. parietalis and 44 percent of the T. e. vagrans were empty.

This habitat was unusually different from all the other sites which T. s. parietalis inhabited; there was no utilization by the species of the amphibians, Rana pipiens and Bufo boreas. The frogs were scattered along the entire shoreline, and a particular aggregation at the northeastern side was located in the vicinity of many felled trees. These trees had harbored a large portion of the captured snakes of both species. Similarly, T. e. vagrans, which has not relied heavily on amphibians, except under the special circumstances at Jette Pond, was not observed to have eaten any ranids; and only one individual was found with a toadlet.

It was difficult to understand the absence of amphibians in the diet of T. s. parietalis; however, it was observed that the diet of T. s. parietalis was markedly different in emphasis than T. e. vagrans.

Thus, it was thought that coexistence was possible because the red-sided garter snake ate predominantly Erpobdella, and supplemented its diet with earthworms, Lumbricus. The small leech species did not have the same frequency of occurrence (15 percent versus 55 percent) in the wandering garter snake; and no earthworms occurred in food resources utilized by this species.

Thamnophis elegans vagrans displayed a larger variety of food items, and there were two noticeable areas of emphasis in the diet (Table 4.2). Haemopis, which was common to both species, had a higher frequency of occurrence (40 percent versus 18 percent) in the wandering garter snake; and this food item was followed closely in frequency (30 percent) by the red-sided shiner, Richardsonius b. balteatus.

The slug, Deroceras, and a toadlet were the two food items which appeared to be supplementary in the diet of the wandering garter snake. Another value of the slug was evident, however, because only juvenile snakes (under 25 cm in snout-vent length) were observed to consume this prey. Most of the juveniles of the red-sided garter snake were observed to consume earthworms. This divergence among the food emphasis of juveniles lessened the competition for other small prey items, such as Erpobdella, which was a food item that was common to juveniles and adults of both species.

The fact that six (30 percent) of the 20 T. e. vagrans, which contained stomach samples, contained fish indicated an interesting individualistic character in this species. In the two other sites at which fish were consumed, there was a lower frequency of occurrence: one (17 percent) out of six snakes which contained stomach samples at

Smith Lake, and two (14 percent) out of 14 snakes at Mud Lake. The presence of fish in the diet of a population, in which only T. e. vagrans were found at Mud Lake and in the two sympatric species at Smith and Jette Lakes, indicated a habitat niche that was specifically utilized by this species rather than T. s. parietalis. It may have served ultimately as a potential resource on which the species could rely, if other foods were unavailable. If this assumption was correct, the high frequency of utilization of this food item at this site was caused possibly by another environmental stress.

LAKE ROGERS

Lake Rogers contained the largest sympatric population that was studied. Because of this large number of snakes, it was an excellent site for the study of interspecific competition. Eleven food resources were utilized in total by the two species and only four categories were common to each species. Eighteen individuals (53 percent) of the 34 red-sided garter snakes did not contain any food; 17 of these snakes were found in the early summer period.

Fifteen individuals (31 percent) of the 42 wandering garter snakes had empty stomachs; and similarly, the majority, 13, of these snakes were caught in the same period.

The food items of the two species overlapped in four categories; these were Haemopis grandis, Erpobdella punctata, Bufo boreas (toadlets), and Deroceras (Table 4.3). Ingested Haemopis were occasionally accompanied by their cocoons. This suggested that the garter snakes hunted for this food item at the breeding site of the leeches at the northeast

Table 4.3—Correlation of Prey Items of *Thamnophis* and Specific Habitats (Lake Rogers)

Site	Species	Number of Individuals	Snout-Vent Size Range (cm)	Prey Item
Lake Rogers	*T. e. vagrans*	9	16.8–25.3	*Deroceras sp.*
		15	23.9–54.9	*H. grandis*
		1	32.8	*E. punctata*
		1	31.5	*B. boreas* (toadlet)
		1	42.7	*Peromyscus sp.*
		1	55.3	*Sturnella neglecta*
	T. s. parietalis	1	49.3	*Deroceras sp.*
		4	20.8–54.4	*H. grandis*
		2	41.9–49.7	*H. grandis* and cocoons
		1	47.2	*H. grandis* and *E. punctata*
		2	21.0–42.3	*B. boreas* (toadlet)
		1	52.8	*B. boreas* (tadpoles)
		1	18.5	*E. punctata*
		1	47.5	*R. pipiens* (tadpole)
		1	23.6	*R. pretiosa* (bones)
		1	55.5	*Microtus sp.*
		1	44.1	*Spizella sp.*

sector of the lake; however, they predominantly hunted along the entire shoreline. Haemopis was the main prey for T. sirtalis parietalis and T. elegans vagrans and was ingested throughout the summer; the percentage of the snakes which had ingested this food for each species was respectively, 43 percent and 55 percent.

The large species of leech was more prolific in the lake than the smaller species, Erpobdella punctatum, which was also less conspicuous in the water. Consequently, the snakes that were observed to be searching for food along the water's edge probably selected the larger prey because of the proximity of the large leech rather than because of diet preference.

An interesting observation about the B. boreas that were ingested and the slugs, Deroceras, indicated a divergence of habitat use by the two species. All life history stages of foods were present at the site, and both species ate the newly metamorphosed toadlets; however, only in the red-sided garter snake did the ingestion of tadpoles of the boreal toad occur. The slug, which was a terrestrial food, was more commonly eaten by the wandering garter snake.

These differences suggested that the two snakes did eat the same items but that the red-sided garter snake extended its food habitat range into the aquatic realm. This was reaffirmed by the ingestion of tadpoles and adult forms of both species of ranids in the red-sided garter snakes.

Both species of garter snake had eaten different species of rodents and birds. The occurrence in each species of the same types, but different species, of prey was considered to be incidental. Rodents, Microtus and Peromyscus, were ingested in the early and late summer periods; the fringillids, sparrow and meadowlark, were found ingested, correspondingly, by the red-sided garter snake and wandering garter snake on August 3, 1974.

This inclusion of birds among the type of prey of these snakes indicated a seasonal change in diet. It was observed that the amphibians, aquatic and terrestrial, became less abundant, and that young birds, 4-6 cm in length, were more abundant in late summer. These seasonal fluctuations in types of prey and the fact that Carpenter (1952) observed that during July and August garter snakes exhibited their greatest arboreal tendencies probably attributed to the ingestion of the birds. However, it was difficult to ascertain whether or not broadening the range of the available food resources would occur in only the large adult snakes (i.e. with snout-vent lengths 44 cm and over) because Haemopis and Deroceras continued to be eaten in late summer by other adults (i.e. with snout-vent lengths of 35 cm and over).

The food selectivity by the snake species indicated different dependencies upon the same habitat niches. Although the red-sided garter snake ate members of the terrestrial niche of the wandering garter snake, 69 percent of the food items were from the riparian and aquatic niches. The T. elegans ate no aquatic amphibians, and only one individual had eaten a toadlet; however, 62 percent of the food items (leeches) also came from the riparian niche (Table 4.3).

This fact seemed to demonstrate that T. sirtalis was a specialist in the aquatic systems and had a broad range within that system. T. elegans, contrastingly, was a generalist, which depended on the aquatic and terrestrial systems, even though there was an obvious continual dependence on the large leech, *Haemopis grandis*.

Another selection was observed among juveniles, approximately between 17-25 cm. These small individuals consistently had the smaller prey items; *T. sirtalis* ate a small spotted frog, the small leech (*Erpobdella punctata*), and toadlets; *T. elegans* ate slugs, small leeches, and toadlets. The large leech, *Haemopis*, was the mutual category used by juveniles and adults of *T. elegans*; and in *T. sirtalis*, *Haemopis* and *B. boreas* toadlets were mutually eaten (Table 4.3).

In respect to *Haemopis*, the juvenile snakes contained small ones (1.0 cm in size); the adults generally contained a range of sizes, 4.0-8.0 cm. Thus, the food selection by the small and large snakes was correlated partially with size of the predator. In the case of toadlets (2.0-2.5 cm), this size correlation was not observed because the newly metamorphosed toads were readily abundant.

CONCLUSION

In the area of food selectivity the populations at each site demonstrated a uniqueness, which was the result of the particular environmental stresses placed upon the specific populations. However, in the sampling of each population, a constant similarity prevailed; a certain percentage of the individuals of each population had empty stomachs. In other studies, the occurrence of a high percentage of *Thamnophis* within a population without any stomach contents was considered common (Carpenter 1952, Fouquett 1954, Gregory and Stewart 1975).

Two factors have been suggested as the causes of this effect. Fouquette (1954) attributed the phenomenon to the fact that the snakes most likely to be caught were those foraging for food. Gregory (1973) indicated that climate affected the activity cycle of the population of Thamnophis sirtalis parietalis in the Interlake region of Manitoba (50°50'N, 97°30'W).

In Manitoba, which was close to the northern limit of the range of the red-sided garter snake, six months were spent in the hibernaculum. During the remaining part of the year, one-and-a-half months each spring and autumn were spent in an active non-feeding state at the hibernaculum. This was physiologically connected with the mating of the snakes (Gregory 1973). The significant months for feeding were June, July, and August; it was most intense in early and mid-summer (Gregory and Stewart 1975).

A comparison of the feeding activity of the snakes during the early, mid, and late summer season was made between the six sites in this study (Table 4.4). The early summer period (June 22-July 22) was the time of the greatest occurrence of empty stomachs in the garter snakes. The range in the percentages of the populations without any stomach content was from a low of 25 percent in the wandering garter snakes at Mud Lake to a high of 88 percent in the red-sided garter snakes at Woods Pond. The following were the specific percentages, in early summer, in each species at each site: Woods Pond, T. s. parietalis, 88 percent; Mud Lake, T. e. vagrans, 25 percent; Smith Lake, T. s. parietalis, 75 percent, T. e. vagrans, 40 percent; Jette Pond, T. s. parietalis, 25 percent, T. e. vagrans, 40 percent; Jette Lake, T. s. parietalis, 43

Table 4.4—Seasonal Occurrence of Food Items in Thamnophis

Site	Season	T. s. parietalis: No. Empty Stomachs	T. s. parietalis: No. of Individuals w/Stomach Contents: Adults	T. s. parietalis: No. of Individuals w/Stomach Contents: Juveniles	T. e. vagrans: No. Empty Stomachs	T. e. vagrans: No. of Individuals w/Stomach Contents: Adults	T. e. vagrans: No. of Individuals w/Stomach Contents: Juveniles
Woods Pond	Early Summer	15	2	0	3	0	0
	Mid-Summer	4	3	1	0	0	0
	Late Summer	2	0	1	0	0	0
Mud Lake	Early Summer	0	0	0	4	8	4
	Mid-Summer	0	0	0	0	1	0
	Late Summer	0	0	0	1	1	0
Smith Lake	Early Summer	6	2	0	2	2	1
	Mid-Summer	1	5	0	0	2	1
Jette Pond	Early Summer	2	2	4	4	1	5
	Mid-Summer	3	0	2	1	3	3
	Late Summer	0	3	2	1	1	1
Jette Lake	Early Summer	6	1	7	12	9	5
	Mid-Summer	4	1	1	4	3	3
Lake Rogers	Early Summer	17	7	4	13	9	11
	Mid-Summer	1	4	1	2	7	2

percent, T. e. vagrans, 46 percent; Lake Rogers, T. s. parietalis, 61 percent, T. e. vagrans, 39 percent.

As Gregory (1973) observed the period of abstinence from feeding occurred during the breeding season; it was assumed that this was the partial reason for the presence of at least 25 percent of the snakes in each population with empty stomachs in early summer. Collections at each site were made within a specific temperature range, so that there was a greater probability of finding the snakes either with recent food samples or ostensibly hunting for prey. The hypothesis of Fouquette (1954), therefore, was supported by these data.

The snakes with stomach contents were compared with regard to each species, regardless of the site; and it was found that the extent of utilization of the habitat niches of the two species were comparable (Tables 4.5 and 4.6). The frequency of the items, Erpobdella and Bufo, was the same as in the list of prey of T. s. parietalis; however, it must be remembered that Bufo was observed to be consumed in great numbers only, in the late summer, when the toadlets had just metamorphosed.

The next most commonly consumed prey by T. s. parietalis, in descending order, were Haemopis and Ambystoma. The fact that Ambystoma was also high in percentage indicated that the snakes were opportunistic in respect to metamorphosing amphibians. Thirty-six percent of its food was B. boreas and A. macrodactylum, and 41 percent of its food was the two species of leeches, which were present continuously throughout the summer.

If a list of combined food items was made for the species, T. s. parietalis, such that all the amphibian species were grouped together, a

Table 4.5—Summary of Food Items of *Thamnophis sirtalis parietalis*

Prey Species	Number of Occurrences	Frequency of Occurrence Singly (%)	Frequency of Occurrence Categorized (%)
Haemopis grandis	11	18	41
Erpobdella punctata	14	23	
Deroceras sp.	1	2	2
Rana pretiosa	2	3	46
Rana pipiens	3	5	
Bufo boreas	14	23	
Ambystoma macrodactylum	8	13	
Hyla regilla	1	2	
Microtus sp.	1	2	2
Spizella sp.	1	2	2
Lumbricus sp.	4	7	7

Table 4.6—Summary of Food Items of Thamnophis elegans vagrans

Prey Species	Number of Occurrences	Frequency of Occurrence Singly (%)	Frequency of Occurrence Categorized (%)
Haemopis grandis	26	29	44
Erpobdella punctata	13	15	
Prophysaon sp.	11	12	24
Deroceras sp.	11	12	
Richardsonius balteatus	7	8	10
Salmo sp.	2	2	
Rana pretiosa	1	1	20
Rana pipiens	1	1	
Bufo boreas	9	10	
Ambystoma macrodactylum	5	6	
Hyla regilla	2	1	
Peromyscus sp.	1	1	1
Sturnella neglecta	1	1	1

principal dependency of the snake species upon leeches and amphibians appeared. The leeches and amphibians, respectively, comprised 41 percent and 46 percent of the food items frequently found in red-sided garter snakes. Earthworms, slugs, mice, and birds were occasionally found and comprised the remainder, 13 percent.

In T. e. vagrans, the most frequently found prey items were Haemopis (29 percent) and Erpobdella (15 percent) (Table 4.5). The two genera of slug, Prophysaon and Deroceras, were equally frequent in the tertiary food category at 12 percent; and B. boreas comprised 10 percent. As in T. s. parietalis, the two leech species and toadlets were among the commonly found food items in T. e. vagrans.

An obvious difference, however, was the emphasis on slugs in T. e. vagrans (Table 4.6). If one combined the food types, the two most common food items appeared to be leeches (44 percent) and slugs (24 percent). A rather large portion of the remaining food items was comprised of amphibians (20 percent) and fish (10 percent); the minor portion (2 percent) was comprised of mice and birds.

The relative frequencies of leeches in both species indicated that this food type was equally important to each species. However, the high frequency of amphibians in T. s. parietalis, and the high frequency of slugs, amphibians, and fish in T. e. vagrans demonstrated the specialization of the red-sided garter snake and the usage of a wide range of habitats by the wandering garter snake. Therefore, the presence of both species at one site, such as a pond, was most likely a permanent relationship because of both the common and different preferences of each species and the existence of a variety of foods at the site.

TEMPERATURE RELATIONSHIPS

It was expected that a wide range of cloacal temperatures would occur because of the activity of the snake and the prevailing weather conditions. Fleharty (1967) noted the activity of a population of Thamnophis in relation to weather conditions and discovered that one species, T. rufipunctatus, was more active on overcast days than two other species. Thamnophis, as a genus, had a tolerance for cold, and its activity range was from 9°-36°C, with a mean of 26°C (Brattstrom 1965). Its lethal temperature limits were 0°C and 40-42°C (Leuth 1941).

Collections were made under many diverse conditions; therefore, temperatures were taken when the snakes were dormant under logs or active on the ground, and during cool, overcast days as well as warm, clear days. Cloacal temperatures were measured from all snakes except those whose size precluded the entrance of the thermometer; these snakes usually fell within the juvenile size class.

The cloacal temperatures ranged from 12°-32°C; the arithmetical means of the temperatures demonstrated the direct relationships which existed at each site (Table 4.7). The graphs of the cloacal temperature versus ground-surface temperature, which were composites of both species, demonstrated the ranges of temperatures that individual snakes maintained at a specific ground temperature, as well as the means. Commonly, the cooler ground temperatures at Jette Lake and Lake Rogers influenced the activity temperature of the snake, which was also cool. However, the graphs also demonstrated that the phenomenon of primitive endothermy was present in the garter snakes (Figures C, D, and E). This phenomenon was

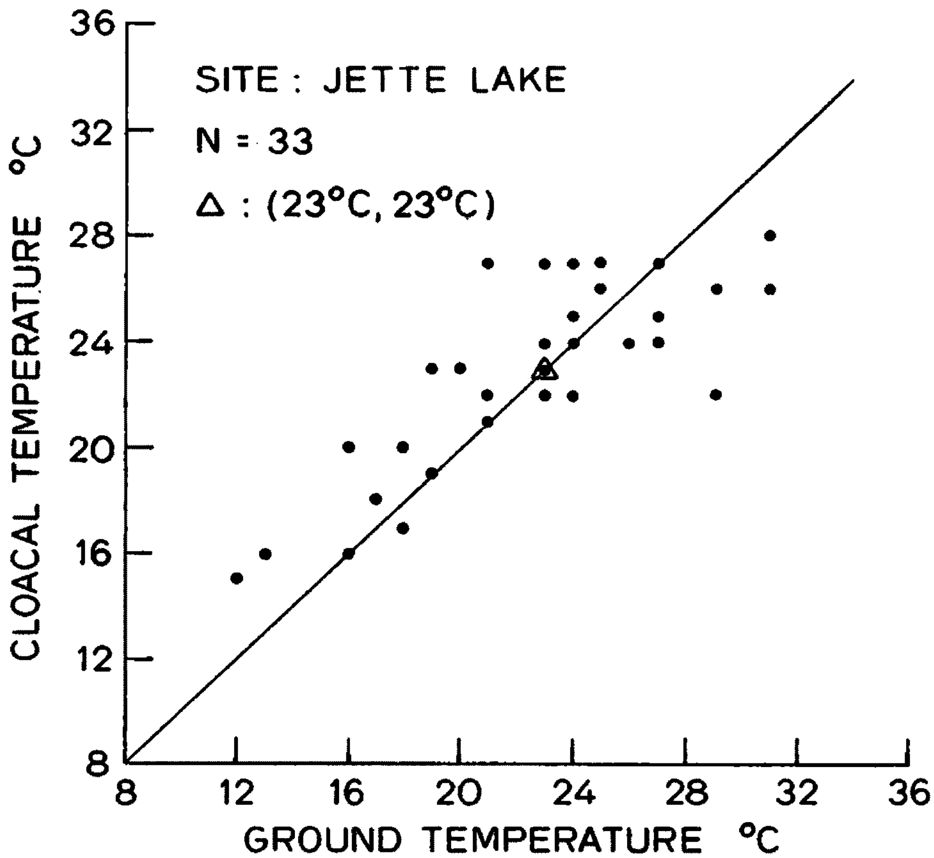
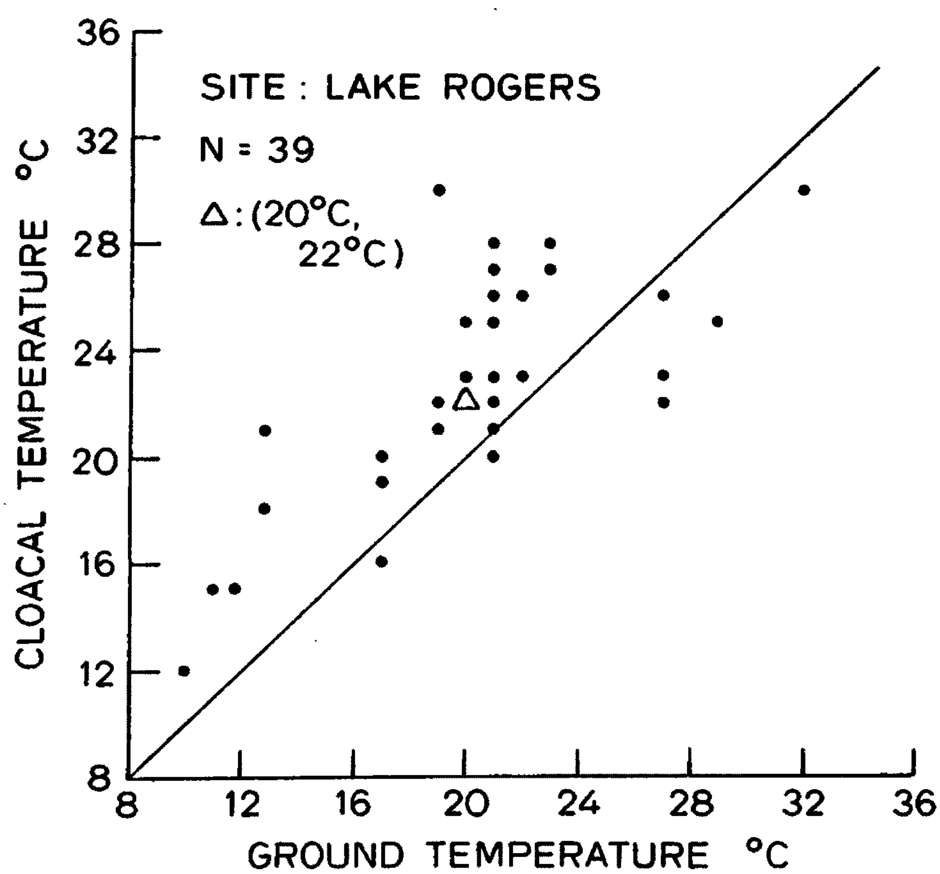


Figure C—Relationship of Cloacal and Ground Surface Temperatures in Genus *Thamnophis*.



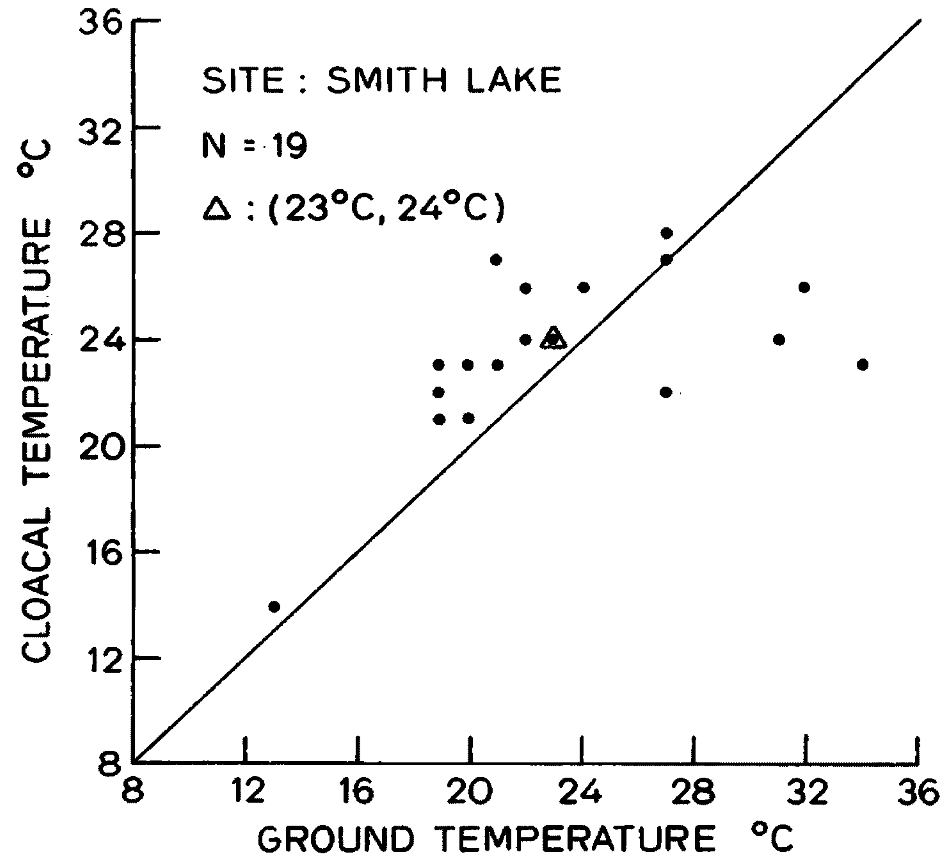
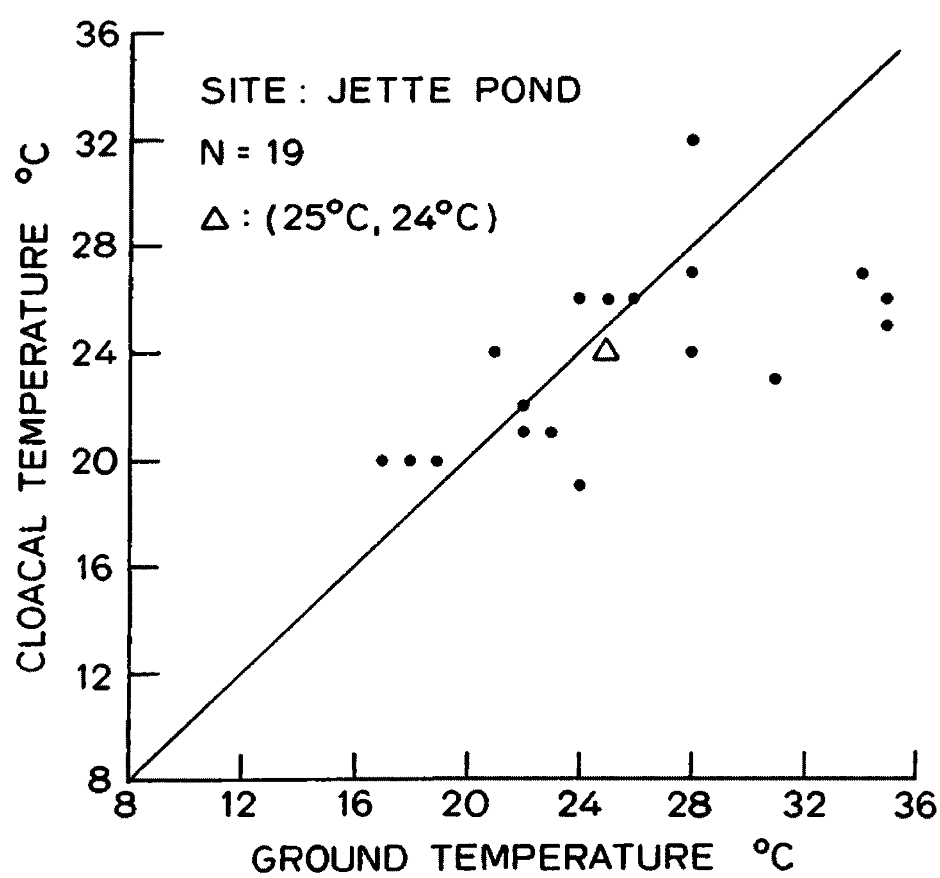


Figure D—Relationship of Cloacal and Ground Surface Temperatures in Genus *Thamnophis*.



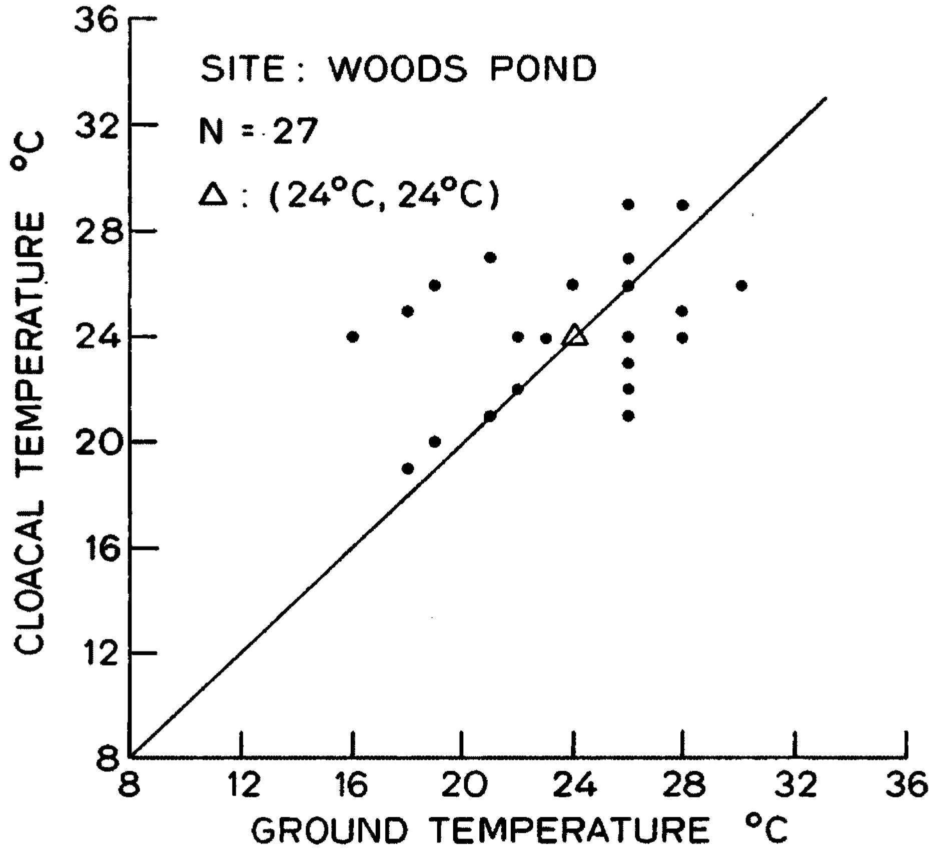


Figure E—Relationship of Cloacal and Ground Surface Temperatures in Genus _Thamnophis_.

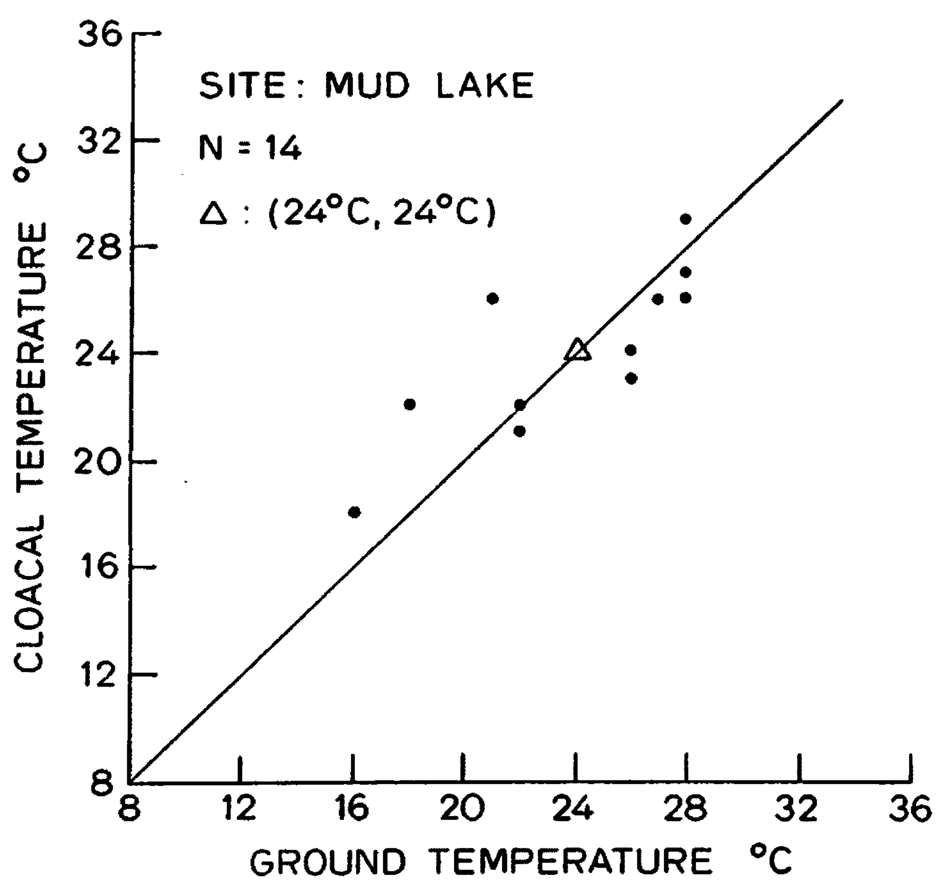


Table 4.7—Correlation of Ambient Temperatures and Atmospheric Pressure with the Cloacal Temperatures of *Thamnophis*

Site	N	Cloacal (°C)	Grand-Surface (°C)	Air (°C)	Atmospheric Pressure (mmHg)
Woods Pond	28	R 19–29 M 24	R 16–30 M 24	R 19–32 M 26	R 26.5–30.0
Mud Lake	14	R 18–29 M 24	R 16–28 M 24	R 21–29 M 24	R 29.3–30.0
Smith Lake	19	R 14–28 M 24	R 13–34 M 23	R 17–28 M 23	R 26.6–30.7
Jette Pond	19	R 19–32 M 24	R 17–35 M 25	R 19–27 M 24	R 26.6–30.3
Jette Lake	33	R 15–28 M 23	R 12–31 M 23	R 12–27 M 23	R 28.3–30.1
Lake Rogers	39	R 12–30 M 22	R 10–32 M 20	R 8–29 M 20	R 26.9–30.2

evident by visible and constant occurrence of most of the snakes with a higher cloacal temperature than the ground. (The exception of this observation was at Jette Pond and was most likely due to the activity which was constantly happening between the shore and water environments.)

Carpenter (1956) recorded the occurrence of endothermy in other species of garter snakes. Sometimes one species can show a tolerance to lower temperatures; Fleharty (1967) proved this tendency by graphing the activity temperatures of individual species. In this study, no significant difference was noted between T. sirtalis and T. elegans. The mean activity temperature was the same for both species, 23°C; the respective ranges were 14°-32°C and 12°-30°C (Figure F). Therefore, it was concluded that these two species were in direct competition in their daily periods of activity and that the distinguishable differences in diet compensated for the same acticity cycles.

If the air temperatures became 30°C or more, garter snakes have been known to maintain their temperature below 30°C (Leuth 1941). In this study, snakes, which were lower in temperature than the ground-surface temperature when it was 30°C, were thought to be exhibiting this behavior. However, other snakes with temperatures below that of the substrate could have recently moved from one environment to another. Without constant monitoring, the snakes may have just emerged from underground or water before capture; and their temperatures, therefore, corresponded with that of the previous environment.

An aneroid barometer, which was set by and compared to the U.S. Weather Bureau readings at Missoula and Kalispell, was used to measure the parameter of weather and atmospheric pressure. Collection periods

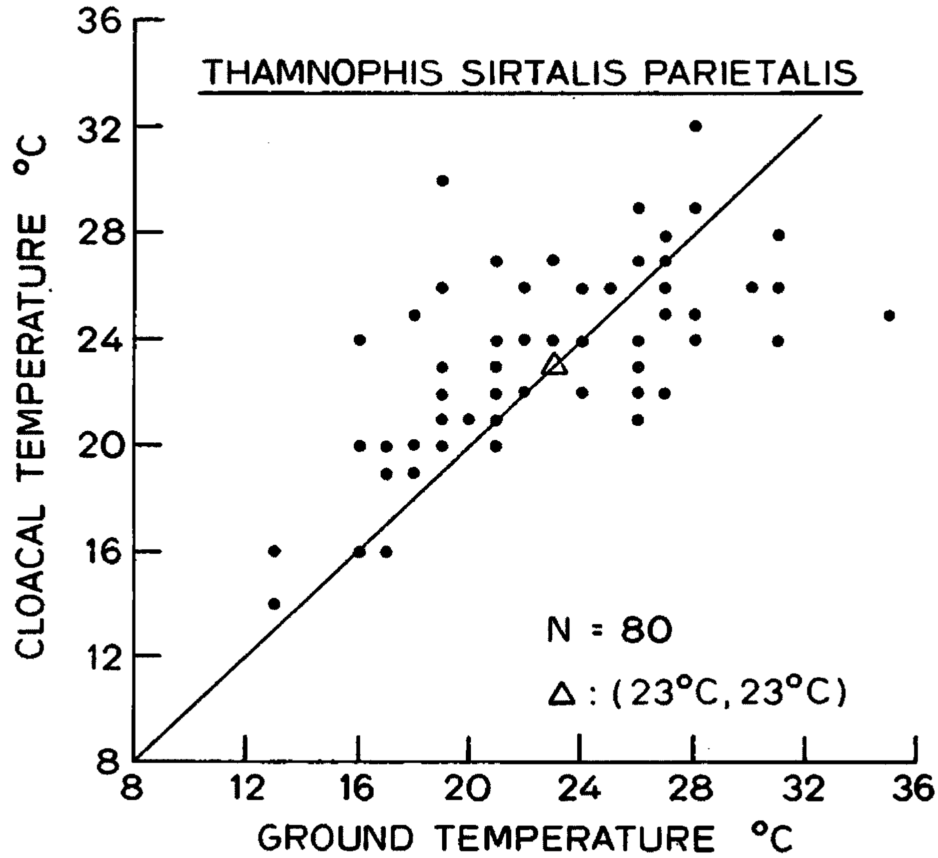
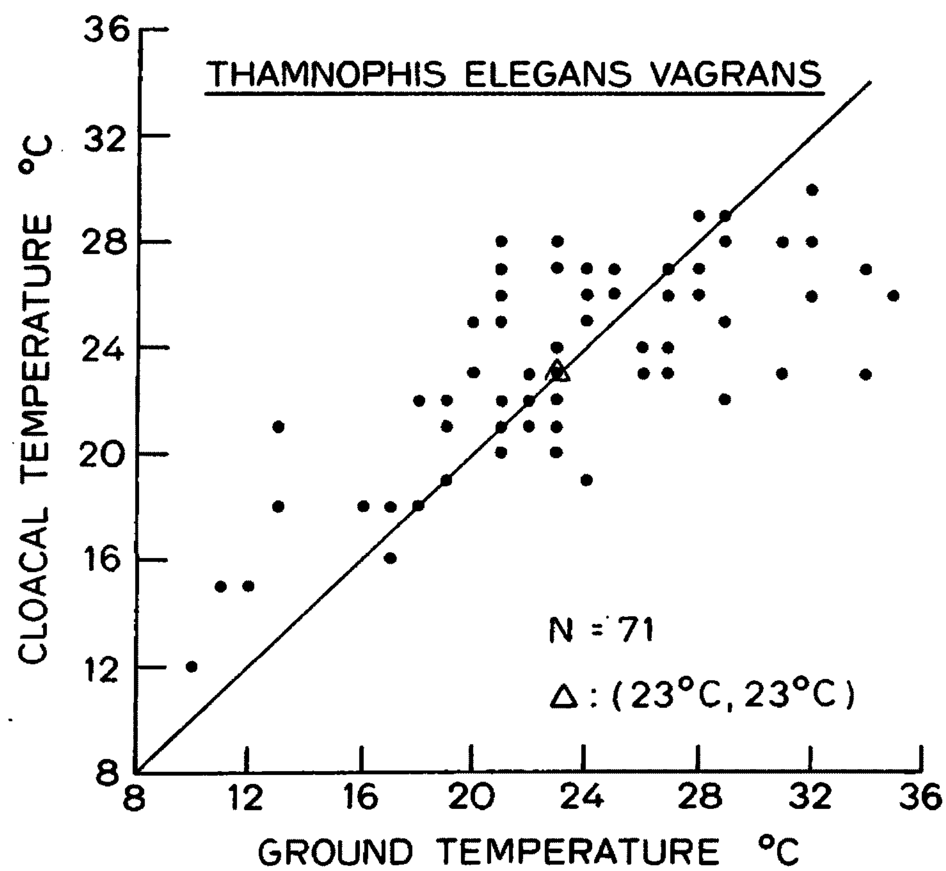


Figure F—Relationship of Cloacal and Ground Surface Temperatures for the species of *Thamnophis*.



were selected to increase the probability of finding snakes active. Therefore, warm days under high pressure systems were preferred. However, overcast and stormy days were inevitable.

The range of barometric readings at each site were recorded (Table 4.7). All the activities (i.e. sunning, moving, swimming, or resting under a log) were observed in both species within these sites during the range of barometric pressures. No trends were noted in the area of activities versus specific pressures; one noticeable absence of an activity, that of swimming, was observed at Smith Lake and Woods Pond. (This was thought to be incidental because the presence of fish from snakes at Smith Lake and Erpobdella at Woods Pond was indicative of aquatic activity.) This also reaffirmed the conclusion that other parameters, such as food, were the isolating mechanisms that allowed the coexistence of these species.

CHAPTER V

POPULATION SIZE AS AN INDICATOR OF DISTURBANCE

INTRODUCTION

The use of the size of a population of garter snakes as an indicator of disturbance was investigated in this study. A survey which entailed the recording of length and sex, and the marking of each snake for future recognition, was undertaken at each of the six collection sites (i.e. Woods Pond, Jette Pond, Jette Lake, Smith Lake, Lake Rogers, and Mud Lake). Snakes, that were small in body length and were unable to be sexed without laboratory dissection, were placed in the juvenile category. An overlap occurred in body lengths of juveniles and adults because of these field methods of sexing individuals (Tables 5.1 and 5.2).

The question as to whether or not a snake was mature has been difficult to determine. It has been done in the laboratory by observing the sexual receptivity of female T. s. parietalis (Hawley and Aleksiuk 1976). Females which were over 50 cm in snout-vent length were observed to have a larger number of ovarian follicles than smaller females; and in a group of females with a snout-vent range of 40-70 cm, courtships by males increased in number per female as the snout-vent length increased above 45 cm. White and Kolb (1974) did a similar study, in which dead Thamnophis were placed in specific reproductive categories, by the examination of the gonads. Generally, an index by which to determine the maturity of male snakes has not been as easily perceived.

Table 5.1

Means and Ranges of the Lengths of *T. sirtalis parietalis* by Category Vs. Locality[1]

Sites	Juveniles					Males					Females				
	N	Total		Snout-Vent		N	Total		Snout-Vent		N	Total		Snout-Vent	
		(Mean)	(Range)	(Mean)	(Range)		(Mean)	(Range)	(Mean)	(Range)		(Mean)	(Range)	(Mean)	(Range)
+*Woods Pond	6 (5)	33.2	24.8-41.6	28.1	18.5-31.1	9	60.4	46.5-73.0	44.9	32.7-53.6	12	66.0	49.9-81.0	51.8	37.7-72.0
Jette Pond	10	31.0	23.5-41.9	23.1	17.7-30.9	5	68.8	59.6-80.8	51.4	44.1-60.3	3	60.8	56.5-64.7	46.3	43.0-49.8
Jette Lake	11	33.3	23.7-39.8	24.8	15.2-37.1	5	64.5	41.5-80.3	50.0	30.6-62.5	4	57.3	41.5-69.8	43.4	31.3-55.7
Smith Lake	—	—	—	—	—	8	59.6	49.7-68.5	45.6	41.8-53.1	6	71.1	60.0-88.0	56.7	47.2-66.0
Lake Rogers	14	30.5	24.7-44.8	22.3	18.5-29.3	12	61.8	55.2-67.3	47.3	42.3-51.7	8	62.3	29.5-74.8	49.7	21.9-61.9
Mud Lake	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—

[1]All measurements to the nearest 0.1 cm.

+Five individuals were measured in snout-vent; six were measured in total length.

*One individual could not be sexed in field; it exceeded juvenile category (64.0, 47.6).

Table 5.2

Means and Ranges of the Lengths of *T. elegans vagrans* by Category Vs. Locality[1]

Sites	N	Juveniles Total (Mean)	Juveniles Total (Range)	Juveniles Snout-Vent (Mean)	Juveniles Snout-Vent (Range)	N	Males Total (Mean)	Males Total (Range)	Males Snout-Vent (Mean)	Males Snout-Vent (Range)	N	Females Total (Mean)	Females Total (Range)	Females Snout-Vent (Mean)	Females Snout-Vent (Range)
+Woods Pond	1	35.0	—	—	—	1	47.3	—	32.9	—	1	45.0	—	35.0	—
Jette Pond	11	36.7	28.0-38.3	28.0	22.1-34.9	3	56.4	44.5-62.7	40.5	28.5-47.7	6	51.3	41.4-74.0	38.2	28.0-58.0
Jette Lake	14	29.8	25.7-39.6	22.7	19.3-29.5	10	57.4	48.0-73.0	44.2	35.5-55.6	12	54.7	43.2-74.3	42.5	33.7-59.2
Smith Lake	4	37.5	30.0-47.1	28.2	23.0-36.0	1	62.0	—	48.1	—	3	55.5	43.8-72.6	45.7	33.5-55.3
*Lake Rogers	21	32.3	22.2-43.7	24.8	16.8-33.2	9	57.5	42.4-70.3	42.1	31.5-55.5	10	58.2	44.1-69.8	46.8	32.8-55.3
Mud Lake	4	32.2	20.2-36.4	24.1	15.4-28.1	10	58.9	52.7-65.4	46.9	40.9-56.9	5	58.5	47.3-77.0	44.8	34.4-60.0

[1]All measurements to the nearest 0.1 cm.

+One individual was not measured in snout-vent, only total length.

*Two individuals were omitted; they could not be sexed in field and exceeded the juvenile category (50.0, 38.8; 53.2, 41.2).

The reason for the presence of differing proportions among the juveniles and adults and among adult males and females in each population was multi-faceted. In this study, the densities of the juveniles and adults changed continually as seasonal changes occurred. It has been observed in a population of T. s. parietalis from Manitoba that males emerged from the hibernaculum in mid-May or early June before the females emerged; this behavior had obviously increased the probability of a greater percentage of females being mated (Gregory 1974). Contrastingly, during July and late August, in the same population, the tendency of female red-sided garter snakes to bask in open areas increased the opportunity of collecting this sex (Aleksiuk and Stewart 1971). This behavior has also been noted in three other species of Thamnophis (Carpenter 1952).

In a study of a sympatric population of Thamnophis, the population size was observed to more than double in one season because of the young snakes (10–39 cm in snout-vent length), which made up more than half of the individuals (Carpenter 1952). If half of these young snakes had survived the first year of life, the expected result of having the juveniles outnumber the adults would have occurred the next year. However, in the study of Carpenter (1952), the low ratio of juveniles to the total population in the following year suggested a high mortality of juveniles by "winter kill."

Winter kill was death by the inability to bury below the frost level in the hibernaculum (Carpenter 1952); this loss of juveniles has been explained because recently born snakes have been found to feed late in their first fall of life (Gregory and Stewart 1975). Another factor that could have caused a lower number of juveniles in the immediate vicinity of the adult population was the behavioral factor of young snakes selecting hibernating sites, other than the communal den of the large snakes (Gregory 1974).

The period of aphagia, or lack of feeding, and the low winter temperatures have been noted to cause the emergent red-sided garter snakes to be near the starvation point in their physical condition (Aleksiuk 1976). The poor nutritional condition of these snakes in the spring made them vulnerable to predation at the site of the hibernacula (Aleksiuk and Stewart 1971). The further predation on juveniles because of their relative abundance and, on large individuals, because of their greater visibility to many predators, has been attributed to the reason for the bulk of a population to fall with the middle size range (Carpenter 1952). The range of the dominant size class and the mean size of the red-sided and the wandering garter snake species in the current study were respectively 61.0-70.9 cm (53.3 cm) and 41.0-50.9 cm (46.5 cm).

Without more years of observation of the ecology of these individual populations, specific reasons for the proportions of the number of adults to the number of juveniles cannot be formulated. Although the inclusion of the parameters mentioned in the above discussion, and the investigation of other parameters would be necessary in subsequent studies.

METHODS AND MATERIALS

Each site was visited during specific times of the day so that the period at which most of the snakes were active would be encountered. Trips to the specific sites were various in number; special attention was paid to the sites that had low concentrations of snakes in order to increase the validity of the estimate of the sizes of the populations. The populations were sampled in the field by a mark and recapture procedure.

Several methods of marking reptiles have been devised; the tatooing of scales (Woodbury 1956), the implanting of miniature radio transmitters into the body cavity (McGinnis 1967), and the fastening of a visible plastic plug to the caudals by the use of the "Buttoneer" (Pough 1970) were three of the common methods. The standard method, that has been traditionally used by such authors as Carpenter (1952) and Fleharty (1967), was initiated by Blanchard and Finster (1933). It was this method that was used in this current study (Table 5.3).

The procedure of Blanchard and Finster (1933) entailed the removal of certain caudal scales or a combination of scales by the use of fingernail clippers. The removal of these scales was specifically patterned for each site. Neo-Polycin ointment was applied for the prevention of infection. The scar tissue, which subsequently formed, was easily recognizable over a year later in this study; Conant (1948) expected that four to five years would be the time limit for the recognition of snakes which were marked by this method.

A routine of inspection was established at each site; however, care was taken to lessen the actual disturbance that this study caused upon each site's ecosystem. No grounded logs were ever overturned, especially those which were probable hibernacula. And, no traps were used in the collection of the snakes, all captures were done by hand.

The data from the mark-recapture survey of each site were tabulated (Table 5.3), and a statistical test was performed on these data.

Table 5.3

Population Survey: Individuals Marked, Recaptured

Sites	*Thamnophis sirtalis parietalis*		*Thamnophis elegans vagrans*	
	Marked	Recaptured	Marked	Recaptured
Woods Pond	28	3	3	0
Jette Pond	18	0	20	2
Jette Lake	20	3	35	5
Smith Lake	14	0	8	0
Lake Rogers	34	4	42	4
Mud Lake	—	—	19	1

Table 5.4

Population Estimate (after Schnabel [1938])

Sites	*Thamnophis sirtalis parietalis*		*Thamnophis elegans vagrans*	
	Number of Individuals	Standard Error	Number of Individuals	Standard Error
Woods Pond	108	± 62	infinite	—
Jette Pond	infinite	—	80	± 57
Jette Lake	52	± 30	106	± 47
Smith Lake	infinite	—	infinite	—
Lake Rogers	110	± 55	212	±106
Mud Lake	—	—	86	± 86

The statistical method, which was adopted for this particular study, was patterned after Schnabel (1938). Standard error was included for each estimate (Table 5.4).

RESULTS

In the following discussion of the populations at each site, the null hypothesis was the same. The null hypothesis was from the model of Schnabel (1938); it stated that each sample was drawn at random and that the population of the site remained constant throughout the period of consideration. Therefore, it assumed that there was no removal or recruitment of individuals within a population.

At Woods Pond, 28 red-sided garter snakes were marked; three of these marked snakes were recaptured. The recaptures occurred on two separate dates; in each case the recaptured snakes had been marked on the previous visitation, which was seven to eight days earlier. The amount of movement of these three snakes averaged 13 meters.

The population estimate was formulated as 108 $\pm$ 62 for the red-sided garter snakes. No estimate could be made for the wandering garter snakes, because only three snakes of this species were found and no recaptures were made. Two wandering garter snakes were found on the first visit on June 19, 1974, and the other was found on September 14, 1975.

At Jette Pond, 18 red-sided garter snakes were marked without any recaptures; 20 wandering garter snakes were marked with two recaptures. One of the recaptured individuals was found actively swimming in the pond, presumably hunting because it regurgitated live leeches. The other was originally found hunting along the south shore; and on its recapture, it was found dormant under a log along the northwestern shore. The population estimate which the statistical analysis produced was one of infinity for the T. sirtalis parietalis and one of 80 ± 57 individuals for the T. elegans vagrans.

Unlike Jette Pond, recaptures occurred for both species at Jette Lake. Twenty red-sided garter snakes were marked and three were recaptured; 35 wandering garter snakes were marked and five were recaptured. There was an unusual occurrence of the same individual red-sided garter snake being recaptured five days later and, again, in June, 1975, approximately ten months later.

The two other recaptured red-sided garter snakes were juveniles; one of these juveniles was marked on the last visit in August, 1974 and recaptured in June, 1975. Two juvenile wandering garter snakes were likewise marked and recaptured. Three adults of this species were also recaptured; one male was captured with an interim of a month and another male was "recaptured" as a dead individual. The snake was originally caught around pilings, and it was found dead under a large, water soaked log. There were 19 days intervening, but, by the condition of the snake, it had not been dead for this length of time. The cause of death was suspected as by cattle.

The formulated size of the population of T. sirtalis parietalis was 52 ± 30, and size of the T. elegans vagrans was 106 ± 47 individuals. This estimate was commensurate with the actual marking of more T. e. vagrans (i.e. 35) than T. s. parietalis (i.e. 20).

At Smith Lake, 14 red-sided garter snakes and eight wandering garter snakes were marked. None of these marked individuals of either species was recaptured. The estimate of these populations was, therefore, formulated as infinite.

At Lake Rogers, the greatest number of individuals of both species were marked. Thirty-four red-sided garter snakes were marked and four more recaptured; 42 wandering garter snakes were also marked, and four were recaptured. In _T. s. parietalis_, the span of time was fairly short from the time of marking to recapture; the interval time range was from one to three days for three of the snakes. The fourth was recaptured exactly a year minus a day after its marking.

In _T. e. vagrans_, two recaptures occurred during 1974; the time intervals were one to seventeen days. Two snakes, which were marked on the same initial trip of June 29, 1974, were found again, coincidentally, a year later on June 29, 1975. The population estimate of the wandering garter snake was 212 $\pm$ 106 individuals; that of the red-sided garter snake was 110 $\pm$ 55 individuals.

At Mud Lake, 19 wandering garter snakes were marked; one wandering garter snake was recaptured twice. Representatives of the other species, _T. sirtalis parietalis_, were never seen. The region of greatest concentration of wandering garter snakes were found at the southeastern shore, where planks of wood provided shelter for the snakes. It was in this vicinity that the recaptured individual was found each time; the time interval between the recaptures ranged from one day to sixteen days. A visitation was made in 1975, but no marked snakes were found. The estimated size of the population was 86 $\pm$ 86.

DISCUSSION

Futility in the use of statistical methods to estimate the size of a population was experienced. All the available objective methods would have given the same result of infinity for populations in which no recaptures were made. In this discussion, the results from the statistical test have been either verified or nullified by the field observations made by the author during this period of study.

At Woods Pond, the estimate of _T. s. parietalis_ was considered fairly high and the estimate of infinity for _T. e. vagrans_ was rejected (Table 5.4). The alternative reason to explain the phenomena was that, for both species, this locus was but a temporary feeding area for a large amount of transient snakes. The relative geographical area that surrounded Woods Pond was plentiful in temporary and permanent potholes and ponds. And, the numerous red-sided snakes which were observed as having "hibernated," or rested, under select shrubs, must have emigrated to other suitable habitats.

Originally, on a preliminary trip, the site was chosen as a relatively undisturbed area by man, however the existence of natural predatory disturbance was exemplified by the presence of a large, emaciated red-sided garter snake. Carpenter (1952) attributed the paucity of large snakes in a population to their visibility to predators. The main disturbance was noise and dust from horses, farm machinery and cars that passed the pond on the dirt road. Only on one specific occasion alongside the pond were reptiles, specifically western painted turtles, found dead by a .22 caliber gun. This threat was not applicable for garter snakes because the scarcity of snakes found in this study indicated that the possibility of finding three or more snakes at a visit was minimal.

Because the site was originally assumed as undisturbed, it was also assumed to demonstrate the stability of a population of garter snakes. During the course of the study, the use of this site for the study of the relationship of a population's size to the degree of disturbance upon the site was decided to be inapplicable.

At Jette Pond, approximately an equal number of snakes were marked for both species (i.e. 18 T. s. parietalis, 20 T. e. vagrans), however, an estimate was attainable only for T. elegans vagrans (Table 5.4). This was because two recaptures occurred in this species. The capture of the T. elegans vagrans was over a range of eight visits, with an average of 2.5 snakes per visit; the capture of T. sirtalis parietalis was over a range of six visits, with an average of 3.0 snakes per visit. It was concluded, therefore, that both species were equally abundant. The high availability of felled logs, which the snakes could use for shelter, contributed to the success of the environment; and because the grounded logs were not disturbed, it can be hypothesized that many of the marked snakes could have been using these as a shelter or as a hibernaculum.

Disturbances were continually occurring at Jette Pond by the diurnal visits of a herd of cattle, by the passing of random cars and logging equipment on the dirt road, and by sporadic visits of caterpillar tractors to the vicinity of the pond proper. Surprisingly, the logging operations, in conjunction with the abundance of amphibians, provided an ideal situation for the maintenance of a sympatric population. Thusly, the felled trees from the logging provided ample shelter for both species;

and the abundance of food items in the pond provided the staples for the support of a resident population of garter snakes. In such a situation as this, it was beneficial to have man-made disturbance; however, if the logs and brush were ever burned in clearing operations, both species in this population would be severely affected.

Jette Lake was a private lake that was in the process of changing ownership; thereby, it was being changed from a one-family lake to a lake surrounded by subdivisions for many families. Sampling of the population took place during the transition; land was being cleared and piles of brush and branches were being placed along the margin of the lake.

Members of both species were common; however, there was an obviously greater number of wandering garter snakes than red-sided garter snakes. This was demonstrated by the difference between the number of *T. elegans vagrans* (i.e. 35) that were marked and that of the *T. sirtalis parietalis* (i.e. 20). Both species had high recapture numbers; the highest number of recaptured snakes occurred at this site (Table 5.3). Five wandering garter snakes were recaptured. Although one recaptured individual had recently died, the number of recaptures and the frequency of marking which was 4.5 wandering garter snakes per visit indicated that this was a stable, healthy population.

The high number of recaptures increased the credibility of the model and reduced the standard error; the estimate of 106 ± 47 for the population size of *T. elegans vagrans* was, therefore, accepted as a valid description of this species. The frequency of marking red-sided garter snakes was 2.5 individuals per visit; and among the three recaptured snakes, one individual was captured twice. The occurrence of the same individual being caught on dates a year apart was indicative of the relative dependence of the red-sided garter snakes on the aquatic ecosystem of Jette Lake. Thusly, the fact that red-sided garter snakes were not as frequently found was recognized not as an indicator of a transient species, but that this ecosystem was inhabited by a smaller population of red-sided garter snakes.

From field observations solely, the sympatric population of garter snakes appeared to be in good condition. The magnitude of disturbance upon the population by the clearing operations was unknown. The fact that the owner was concerned with having no major disturbance within a 61-meter periphery of the lake by the workers or by the families was beneficial to the snakes. However, the home range of the garter snakes has been demonstrated to be at least two kilometers in area; and seasonal changes in diet would probably cause the snakes to hunt in the forested areas around the subdivided plots. A follow-up study would be of importance in this area in particular to estimate the impact upon the population after the completion of the subdivisions.

Smith Lake was the area of most usage by people. The major impact was by vacationers from out of state. They, plus the local populus, collected frogs, toadlets, and earthworms in happy abandon for fishing. The impact of overturned logs, which were not rolled back to their original position by these fishermen, was unknown. The log disturbance and the removal of possible food items of garter snakes compounded the encroachment of men into the lake's ecosystem.

This site was visited literally by more people per unit of land than any other sites. The mere effect of people walking along a shoreline has been recognized as a principal threat to the stability of organisms which live along the beaches along the eastern seaboard. It could only be speculated that this, other than the other two disturbances, was the cause of an extremely small number of snakes of both species being found (i.e. eight wandering and 14 red-sided garter snakes in an area of 800 meters by 90 meters).

The statistical model was ineffective in supplying an estimate of the population (Table 5.4), and the only valid description that could be made about the sympatric population was attained by field observation. In the field, hundreds of frogs were observed to be present at each visit; and, because amphibians were found to be of major importance to the red-sided garter snake as a species (Chapter IV, Table 4.5), a small population could be supported in this habitat. Thusly, the higher frequency of red-sided garter snakes than wandering garter snakes at Smith Lake was concluded as indicative of a habitat that favored red-sided garter snakes.

Eleven visitations were made to this site, and less than one snake of either species was found per visit. The rate at which the site has increased in use by people each year was not estimated. However, it has been concluded by this survey on the garter snakes by a mark-recapture method that the site has not supported a stable population of either species for a period of time. If the Montana Department of Fish and Game ever regulated the amount of usage by the people, it has been hypothesized that the red-sided garter snake would readily reinvade the ecosystem.

Lake Rogers was unlike Smith Lake in its usage by people and abundance of garter snakes, however it was similar in area and in its attraction for waterfowl. Seventy-six garter snakes were marked; these included 34 red-sided garter snakes and 42 wandering garter snakes. There was a minor difference in number between the number of snakes marked in each species; but the statistical estimate indicated an abundance of wandering garter snakes. The difference was on a two-fold scale (Table 5.4).

The statistical estimate did not procure an accurate answer on the proportions of individuals of each species. The attributing factors, that caused the capture of more T. e. vagrans than T. s. parietalis, were the aquatic orientation of T. s. parietalis and the inaccessibility of the mats of aquatic vegetation upon which this species rested. Therefore, it was probable that there was as large or larger populations of T. s. parietalis than T. e. vagrans at this site.

Regardless of the estimate's accuracy, this site had the greatest concentration of garter snakes. The Montana Department of Fish and Game had allowed only non-motorized boating and no fishing; these stipulations, plus the fact that the lake was used as a breeding site for grayling, attributed to the slight use by people of the lake. Not more than 15 to 20 people were seen along the margin of the lake which was studied in this survey. Similar to Jette Lake, this lake's margin also has been subdivided into plots, with 90-year leases from the U.S. Forest Service. A prediction of the effect that these subdivisions will have upon the diversity of organisms was inestimatable.

With consideration to the current status of the lake, the presence of disturbances has not been as critical a factor in the existence of a sympatric population of garter snakes as has the kind of disturbances. The state's limitations on its use, the random visits by local people and the occasional traffic together caused the natural equilibrium of the aquatic system to be maintained. The only noticeable disturbances that have affected the immediate area were the herd of cattle which caused the packing down of the soil on the shoreline, and the thinning operations by the U.S. Forest Service. However, these have not been proven to cause adverse effects in the situation of Jette Pond; therefore, it was concluded that the prevention of recreation usage on the lake was directly related to the plentiful number of animals, especially garter snakes.

Mud Lake presented a unique problem because the physical disturbances to the lake and its periphery were minimal; the emphasis on recreational activities was centered at Lake Five, which drained into Mud Lake. The presence of specific levels of heavy metals in the biota was attributed to the drainage of waters from Lake Five. This was the single disturbance that was observed to have affected the background concentrations of lead and mercury which were present in the garter snakes and frogs.

The absence of usage by people similar to that of Smith Lake, and the occurrence of a small population of wandering garter snakes, both suggested that the site was capable of being inhabited by the red-sided garter snake. However, after the first few visitations, the lack of _T. s. parietalis_ was apparent and puzzling. The food items were present to support the species; but there must have been some barriers, natural and/or man-made, that prevented the inhabitation by red-sided garter snake species.

The estimation from the statistical model was equally as puzzling in its answer for the population of wandering garter snakes. The lower estimate of zero was rejected because at least 19 snakes, which were marked, resided there. The fact that the same snake was caught twice in the same area of the lake also certified a dependence of this species upon the area. The upper estimate of 172 individuals was rejected because of the relative size of the area examined and the low number of individuals which were found in that area.

The different coloration of the individuals versus others of the same species from other sites indicated a distinct gene pool in the area of Mud Lake. The answer to the question of how large the population was and why no red-sided garter snakes were found has to be answered by further research. Therefore, a correlation between the disturbances and the use of a population size as an indicator of disturbance was not able to be made at this site.

CHAPTER VI

GENETIC VARIATION

This study of genetic variation within these two species was based on scale counts and body length measurements. In order to obtain a comprehensive view of the variation that exists in western Montana, this survey was broken into sections which concentrated on different aspects. The sections consisted of the following: 1) an investigation of three litters of garter snakes, which were produced from gravid females within the laboratory; 2) an investigation of the variation within each study site; 3) a statistical analysis of a study completed by Todd (1965) for correlation with data from study sites; and 4) a review of preserved specimens collected by Brunson from 1947 to 1954 for comparative purposes. Each section has been treated individually for the purpose of clarity; however, referrals have been made to others within each section.

GENETIC VARIATION WITHIN LITTERS

Although scale counts and scale patterns have been a standard mode to identify a species (Schmidt and Davis 1941), sexual dimorphic traits have also been included in specific descriptions. Length is one such trait. In T. e. vagrans, adult males have averaged smaller in size than females (Fleharty 1967); in T. s. sirtalis, a close relative of T. s. parietalis, males, at birth, were significantly larger (Blanchard and Blanchard 1942, Martof 1954). This relationship changed as the females matured (Blanchard and Blanchard 1942); they became longer than the males.

This sexual dimorphic trait was analyzed in the litters from three gravid females which were collected from different sites in the early summer season. At the time of capture, all the females were very large and obviously gravid; however, the time of gestation of the ova was difficult to estimate. This was because the species Thamnophis had been observed to mate in spring and/or fall in different populations (Blanchard and Blanchard 1942, Gregory 1974). Once copulation has occurred, the female can store living spermatozoa in its oviducts, until ovulation is completed; in this manner, a female can delay fertilization during hibernation (Gregory 1974).

The females were maintained in the laboratory for various durations of time. The lives of the females were terminated when the stage of development of embryos was estimated to be close to maturation. In two of the garter snakes, the embryos were mature and, upon release from the uterus, were able to move and breathe. In each female, a range of embryonic development existed; the embryos that were in the most advanced stages of development were nearest to the cloaca (Table 6.1).

The snakes within each litter were measured, and all their scale counts were made, excepting in the case of Brood I. The young in this litter were the least developed, and the ventral scales were not readily visible for counts. Length was measured in two parts: snout to vent, and tail. A method of comparison, which was adopted from Martof (1954), was used. The ratio of the tail to the snout-vent measurement was formulated; this has been proven to show the most significant difference in the sexual dimorphic trait for the snakes studied by Martof (1954). The

Table 6.1—Comparison of Embryonic Snakes and Female Parents

Species	Brood Number	Number of Young (sex)	Scale Counts				Body Lengths by Sex (cm)				
			Ventral		Caudal		Sex	Snout-Vent		Tail	
			Range	Mean	Range	Mean		Range	Mean	Range	Mean
T. e. v.	I	12 M, 6 F and 1 egg	—	—	67–84	75	M F	4.0*–10.5 7.9–10.5	9.2 9.5	2.0–3.7 2.6–3.5	3.2 3.1
	Female Parent		164		70			57.8		15.7	
T. e. v.	II	4 M, 8 F and 2 eggs and 1 half-developed egg	156–178	167	57–84	74	M F	14.7–16.0 11.4–16.9	15.4 15.4	4.2–4.9 3.2–5.4	4.5 4.7
	Female Parent		166		55			61.5		14.5	
T. s. p.	III	6 M, 7 F	147–166	155	76–89	82	M F	16.0–17.0 16.2–16.8	16.6 16.4	5.1–5.8 5.1–5.9	5.5 5.5
	Female Parent		155		26+			58.8		7.8+	

*Extremely immature male embryo.

+Broken tail.

comparison of the tail length to the total body length has been used more commonly (Conant 1951); however, the tail was compared in part to itself by this method.

In the case of the litters in this survey no significant difference of lengths was observed between the two sexes. Within Brood I, the ratio of tail length to snout-vent length for males averaged 34 percent (range: 30 to 38), and females averaged 33 percent (range: 30 to 36). (The very small, partially developed male was omitted in the average.) Within Brood II, the ratio for males averaged 30 percent (range: 26 to 31); females averaged 31 percent (range: 28 to 33). Within Brood III, the males averaged 34 percent (range: 31 to 35); the females averaged 34 percent (range: 31 to 36). Although the results were not significant the value of this ratio has been used as an alternative to the method of Conant (1951). And in further studies, this ratio would be of more value to the investigator than total length comparisons. It would be of interest to investigate whether this ratio within individual snakes was constant throughout their life span.

The typical scale count of each species was favored within an individual litter (Table 6.2). In _T. e. vagrans_ (Brood I and II), the litters had a majority of the individuals with the normal labial count: (8-8) (10-10). The percentages were 44 and 58 percent, respectively. In _T. s. parietalis_ (Brood III), 92 percent of the litter had the normal labial count: (7-7) (10-10).

Within the litter of _T. s. parietalis_, it was noticed that females had more irregular labial counts than males. In this example, 29.0 percent of the females were compared to 0.0 percent of the males. In _T. e._

Table 6.2—Labial Count Within the Litters

Species	Brood Number	Labial Count: Upper (Right-Left)	Labial Count: Lower (Right-Left)	Number of Individuals: Male	Number of Individuals: Female
T. e. v.	I	(8-8)	(10-10)	4	4
		(8-8)	(10- 9)	2	0
		(8-8)	(10- 8)	0	1
		(8-8)	(9- 9)	1	0
		(8-8)	(8- 9)	1	0
		(8-6)	(9- 9)	1	0
		(8-9)	(9- 9)	0	1
		(7-8)	(9-10)	1	0
		(7-8)	(10- 9)	1	0
		(9-8)	(9- 9)	1	0
T. e. v.	II	(8-8)	(10-10)	1	5
		(8-8)	(10- 9)	0	1
		(8-8)	(10- 8)	0	1
		(8-7)	(10-10)	1	0
		(8-7)	(10- 9)	1	0
		(8-7)	(9- 9)	1	0
		(7-8)	(9- 9)	0	1
T. s. p.	III	(7-7)	(10-10)	6	5
		(7-7)	(10- 9)	0	1
		(8-8)	(10-10)	0	1

vagrans, the males had more abnormal counts: in Brood I (67.0 to 33 percent of females), and in Brood II (75.0 to 38 percent of females).

The means of the ventral and caudal scale counts of the young did not vary much from that of the mother. In Brood I, the ventrals were not compared, the caudals averaged 75; the mother had a count of 70 caudals. In Brood II, the ventrals averaged 167 and the caudals 74; the mother had a count of 166 ventrals and 55 caudals. In Brood III, the ventrals averaged 155 and the caudals 82; the mother had a count of 155 ventrals and 26 caudals. The tail was broken on the T. s. parietalis female.

In litters of both species, two anomalies were present. These abnormalities were more prevalent in the litters than observed in the field or in the preserved specimens. In Brood III, the juveniles had extra scales between the base of the upper labial scales. (See Figure G and Table 6.3.) In this litter of the female T. s. parietalis, 62 percent had extra labial scales; in all the study sites, except Mud Lake, it was observed. However, the percent of the population ranged from 10 to 27. In the preserved specimens, it was present in 25 percent of the individual snakes.

This gravid T. s. parietalis, however, did have an extra scale, which was incomplete and divided two normal scales, on its right labium, and probably was partially the cause for the high occurrence in the litter. It should be noted that this female was chosen from Smith Lake at random; in other words, the snake was not chosen because it possessed the trait.

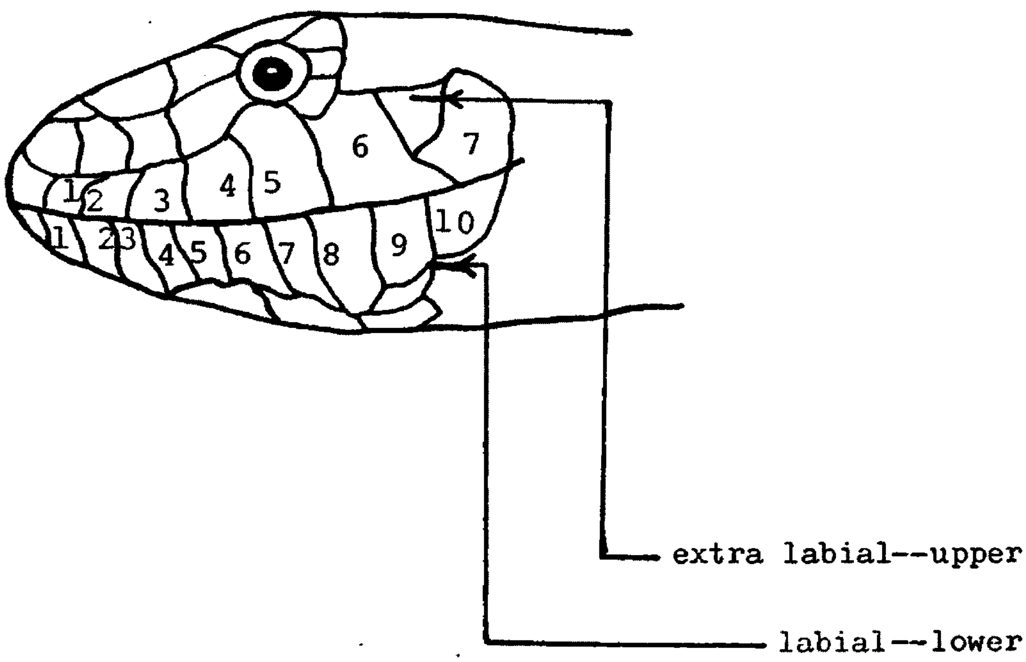


Figure G. Extra Labial Phenomenon in Thamnophis sirtalis parietalis

Table 6.3—Labial Anomaly in Thamnophis sirtalis parietalis

Species	Age Class	Labial Count			
		N	Upper (Right to Left)	N	Lower (Right to Left)
T. s. p.	Female Parent	—	7'–8	—	10–10
	Juvenile	4	7 –7	12	10–10
		1	7'–7	1	10– 9
		2	7'–7'		
		1	7'–7°		
		1	7 –7'		
		2	7°–7'		
		1	7 –7°		
		1	8 –8		

' = 7th scale had extra scale beside it.

° = 6th scale had extra scale beside it.

Table 6.4—Caudal Anomalies in the Young of Both Broods of T. elegans vagrans and Their Female Parents

Species	N (Sex)	Age Class	Number of Undivided Caudals
T. e. v.	2F	Parents	0
	9F, 11M	Juvenile	0
	1F, —	Juvenile	1
	1F, 1M	Juvenile	2
	—, 1M	Juvenile	3
	—, 1M	Juvenile	4
	2F, 1M	Juvenile	5
	1F, —	Juvenile	11
	—, 1M	Juvenile	14

The second anomaly was in the other species (Table 6.4). The presence of undivided caudals is frequently found usually near the base of the tail in sets of two or more consecutive scales. In Brood I, 22 percent had the anomaly; in Brood II, 50 percent had it. Neither of the gravid females had undivided caudals. In the study sites, the individuals in each population that had undivided caudals ranged from 0 to 14 percent of the separate populations; and in the preserved specimens, 20 percent possessed the anomaly.

GENETIC VARIATIONS AT THE STUDY SITES

In each site the length of animals was compared among the adult males and females in each population. Each population of sympatric species was separated into sub-populations which consisted of one species, and then the sub-population was separated into categories. These categories were juvenile, male, and female (Tables 5.1 and 5.2). Juveniles were snakes which were visibly sexually immature by observation in the field; and consequently, their size precluded the entrance of the thermometer into their cloacae. Their sexual identity could have been learned only by dissection in a laboratory.

Although a similar population study has been done by a creek in California in which all the snakes found during the survey were preserved and analyzed (White and Kolb 1974), the emphasis on this current study was not placed in this area of concentration. Furthermore, the capture-recapture data would have been biased; and more importantly, the damage that would have been done to the dynamics of the individual populations would have been undeterminable.

Thusly, juveniles ranged 20.2-47.1 cm in body length, and adult males and females ranged 41.0-77.0 cm. The overlap could be explained by the field methods mentioned in the previous paragraph in regards to recognition of sexual identity. Carpenter (1952) used the range 10-39 cm as the juvenile body, or snout-vent, range, 40-60 cm as the common adult body range, and above 60 cm in body length as a rare occurrence.

Snout-vent length was measured ot the nearest millimeter, and the mean of the snout-vent length of each sex was compared at each site (Tables 5.1 and 5.2). In three of the five sites, adult males were smaller than the females in the species, _T._ _sirtalis_ _parietalis_. In four of the six sites with _T._ _elegans_ _vagrans_, adult males were larger than the females. The general hypothesis on size for garter snakes stated that adult males were usually considerably smaller than females (Carpenter 1955, Fleharty 1967, White and Kolb 1974) and have a slower rate of growth (Fitch 1965). The reason that some of these snakes differed from the hypothesis could be explained by many factors.

Length has been used as a reflection of age, but growth rate in an individual has to be a reflection of its environment. Each population which was studied had a different locus (i.e. altitudinal and latitudinal coordinates); however, the general habitat was the same. Therefore, each population of snakes was a separate ecotype. The adaptations under which the snakes have gone was not investigated in this study, but it was highly probable that variations in length of the seasonal feeding period were present at each locus. The average length, growth rate, and maturation of the individuals in an ecotype would reflect the stresses from their particular environment. This might have been one reason for the variation found in the lengths of both sexes of snakes.

Labial counts of each species at each site were made (Appendix); and from this, two lists were made of the total variation found in each species (Tables 6.5-6.8). The counts that have been used as standards for both species were consistently the majority; although, it was not always over half of the population (Table 6.9). Because of this occurrence, the count which made up the next highest percentage was compared among sites in order to discover if a trend existed.

In two cases, the next most common counts of the red-sided garter snakes were shared. The results were as follows: Smith Lake and Jette Pond both had (7-8) (10-10) as second high; Lake Rogers had (7-9) (10-10); Woods Pond and Jette Pond had (8-10) (10-10); and Jette Lake had (8-8) (10-10).

In the wandering garter snake no obvious trend was seen among the second most high counts at the sites. The results were as follows: Smith Lake and Jette Pond had (7-8) (10-10); Lake Rogers and Jette Pond had (8-8) (9-10); Woods Pond had (8-8) (8-10); and Jette Lake had (8-7) (10-10).

One trend was noticed, however, which involved the regularity of the labial counts. The red-sided garter snake had a larger percentage of individuals with irregular counts in the upper labials than the wandering garter snake; and the exact opposite was true for the wandering garter snakes' lower labials. At the sites, the percentage of variation in the labials of the red-sided garter were: upper, 29-56 percent;

Table 6.5—Labial Scale Variation in Thamnophis elegans vagrans

Lower Labials (Right–Left)	Upper Labials (Right–Left)	Number of Individuals	Total Number of Individuals (percent)
(10–10)	(8–8)	72	55
(10–10)	(8–7)	5	4
(10–10)	(7–8)	8	6
(10–10)	(7–7)	2	1
(10–10)	(9–8)	2	1
(10– 9)	(8–8)	6	5
(9–10)	(8–8)	11	9
(9–10)	(7–8)	3	2
(9–10)	(7–7)	2	1
(9– 9)	(7–8)	1	1
(9– 9)	(7–7)	4	3
(9– 9)	(8–8)	6	5
(9– 8)	(7–7)	1	1
(8–10)	(8–8)	3	2
(8– 9)	(8–7)	1	1
(8– 8)	(8–8)	1	1
(8– 8)	(7–7)	1	1
(8– 8)	(6–8)	1	1

Table 6.6—Labial Scale Variation in Thamnophis sirtalis parietalis

Lower Labials (Right-Left)	Upper Labials (Right-Left)	Number of Individuals	Total Number of Individuals (percent)
(10-10)	(7-7)	37	35
(10-10)	(7-8)	11	10
(10-10)	(8-7)	14	13
(10-10)	(8-8)	10	9.5
(10-10)	(8-9)	1	1
(10- 9)	(7-7)	4	4
(10- 9)	(7-8)	1	1
(10- 9)	(8-7)	3	3
(11-10)	(8-8)	1	1
(11-10)	(7-8)	1	1
(9-10)	(8-8)	1	1
(9-10)	(7-8)	3	3
(9-10)	(7-7)	10	9.5
(9- 9)	(8-8)	2	2
(9- 9)	(7-8)	2	2
(9- 9)	(7-7)	4	4

Table 6.7—Distribution of Lower and Upper Scale Count in *Thamnophis elegans vagrans*

Lower (Right-Left)	N	Percentage	Upper (Right-Left)	N	Percentage
(10-10)	89	67	(8-8)	99	76
(9-10)	16	12	(8-7)	6	5
(10- 9)	6	6	(7-8)	12	9
(9- 9)	11	9	(7-7)	10	7
(8-10)	3	2	(9-8)	2	2
(8- 8)	3	2	(6-8)	1	1
(9- 8)	1	1			
(8- 9)	1	1			

Table 6.8—Distribution of Lower and Upper Scale Count in *Thamnophis sirtalis parietalis*

Lower (Right-Left)	N	Percentage	Upper (Right-Left)	N	Percentage
(10-10)	73	68.5	(7-7)	55	52.5
(9-10)	14	13.5	(7-8)	18	17
(10- 9)	8	8	(8-7)	17	16
(9- 9)	8	8	(8-8)	14	13.5
(11-10)	2	2	(8-9)	1	1

Table 6.9—Percentage of *Thamnophis* with Typical Labial Counts at Each Site

Typical Species Scale Count	Species	Percentages at Sites					
		Woods Pond	Jette Pond	Jette Lake	Lake Rogers	Smith Lake	Mud Lake
(7-7) (10-10)	*T. s. p.*	32	46	35	33	57	—
(8-8) (10-10)	*T. e. v.*	100	55	68	55	36	50

lower, 14-42 percent. The percentage in the labials of the wandering garter were: upper, 14-31 percent; lower, 23-34 percent.

Ventrals and caudals were counted in the field, and the first 40 snakes of both species were counted. It was not pursued further because of the ample data available in Todd (1965) and the study of preserved specimens, which combined would give an adequate description of the common counts in western Montana (Tables 6.10 and 6.11). (The discussion of the total of this study's data, Todd (1965), and the museum specimens has been placed in the final section.)

The range for the T. e. vagrans from the sites was from 151-180 in ventral count, and 56-100 in the caudal count. The most frequently found counts, respectively, were between 166-170 and 71-75. The range for T. s. parietalis from the sites was from 146-190 in ventrals and 61-90 in caudals. The most frequent counts were between 161-165 and 76-80, respectively. In both circumstances the counts were in agreement with the standard description.

The dorsal row count was also done in the field; however, it served as a corrolary for data that already had been substantiated by such authorities as Stebbins (1966). It was not, therefore, counted on all the snakes. The count proved to be quite a stable characteristic as compared to the labial count. The most common count, 21-21-17, was exactly in agreement with the species description for T. elegans vagrans; and the common count, 19-19-17, described T. sirtalis parietalis correctly. However, in both species quite a variation in count was noted (Tables 6.12 and 6.13).

Table 6.10—Variation in Ventrals and Caudals in *T. elegans vagrans*

Present Study	N	Todd's Study	Preserved	All
—Ventrals—				
181–185	0	4	0	4
176–180	6	12	3	21
171–175	6	14	2	22
166–170	18	52	7	78
161–165	6	33	6	45
156–160	8	1	0	9
151–155	1	0	1	2
	45	116	19	181
—Caudals—				
96–100	1	0	0	1
91–95	1	0	0	1
86–90	0	6	1	7
81–85	9	8	4	21
76–80	11	11	3	25
71–75	13	19	6	38
66–70	11	40	2	53
61–65	3	25	1	29
56–60	5	0	0	5
	54	109*	17*	

*Difference in number of snakes examined due to broken tails omitted.

Table 6.11—Variation in Ventrals and Caudals in T. sirtalis parietalis

Present Study	N	Todd's Study	Preserved	All
—Ventrals—				
186–190	1	0	0	1
181–185	2	3	0	5
176–180	1	8	1	10
171–175	0	19	1	20
166–170	7	38	3	48
161–165	10	18	7	35
156–160	9	11	3	23
151–155	6	0	1	7
146–150	3	0	0	3
	39	97	16	152
—Caudals—				
91–95	0	3	0	3
86–90	6	6	2	14
81–85	9	7	3	19
76–80	11	9	4	24
71–75	8	13	3	24
66–70	1	37	2	40
61–65	2	16	1	19
56–60	0	1	0	1
	37*	92*	15*	

*Difference in number of snakes examined due to broken tails omitted.

Table 6.12—Dorsal Row Variation in *T. elegans vagrans*

Todd's Study	N	Present Study	N	Preserved	N
20–20–17	17	20–21–17	1	20–20–17	1
20–20–16	5	21–21–17	10	20–21–17	2
20–20–19	2	21–20–17	1	21–21–17	4
20–19–17	13	19–19–17	1	20–21–16	1
20–19–16	3	21–21–16	9	20–19–16	1
20–19–18	1	21–19–17	2	21–21–16	3
20–21–17	9	21–20–15	1	21–20–16	2
20–21–16	5	21–19–16	3	21–20–15	1
20–21–18	3	19–21–18	1	22–17–17	1
21–21–17	7	19–17–18	1	22–21–17	1
19–20–17	10	21–17–16	1	20–21–15	1
19–20–16	5			21–18–16	1
19–20–18	2				
19–20–19	1				
19–20–15	1				
19–21–17	7				
19–21–19	2				
19–21–16	2				
21–20–17	5				
21–20–16	1				
19–19–17	5				
19–19–16	3				
21–19–17	3				
21–19–16	1				
20–22–18	1				
20–18–17	1				
21–22–17	1				

Table 6.13—Dorsal Row Variation in *T. sirtalis parietalis*

Todd's Study	N	Present Study	N	Preserved	N
19–19–17	13	19–19–17	12	19–19–17	5
19–19–16	7	19–19–16	7	19–18–17	3
19–19–18	4	19–17–16	1	20–19–17	2
19–19–16	3	19–17–18	1	19–17–17	2
19–19–15	2	19–18–16	1	20–18–17	1
19–20–17	16	20–20–19	1	18–19–17	1
19–20–16	7	20–19–17	1	18–17–17	1
19–20–18	2	21–17–17	1	19–16–17	1
19–20–15	1				
20–19–17	7				
20–19–16	5				
19–17–17	6				
20–20–17	5				
20–20–18	2				
18–19–17	3				
18–19–16	2				
19–18–17	2				
19–21–17	2				
19–21–16	2				
20–10–16	2				
21–19–17	2				
18–20–15	1				
20–21–17	1				

The variation in both this study and Todd (1965) could have been caused by the very nature of counting the dorsal row. In this study, a count of the scales was made across the neck, approximately 5-7 rows back from the base of the head, and a second at mid-body of the snake, and a third approximately 10 rows anteriad to the vent. This method was not as exacting as the formula described by Schmidt and Davis (1941).

In a truly taxonomic review of Thamnophis, Schmidt and Davis (1941) would have to be followed. Their formula entailed obtaining the mean of all the rows in each area of the snake and noting the place where the specific change in count occurs. Because only one place was counted, three times repeatedly, in each section of the snake, a higher probability exists of finding differing scale row counts per individual specimen. In regards to the variety that was present in the labials, surely the genetic nature (i.e. gene pool) of the study sites must have influenced the dorsal row count also.

The two anomalies, extra incomplete scales between the upper labials in the red-sided garter snake and the undivided caudal scales in the wandering garter snake, were seen within the study sites. The occurrence of extra incomplete labials was noted as a percentage of population. The range among study sites was from 10 to 27 percent (Table 6.14).

The occurrence of undivided caudals ranged from 0 to 14 percent of the populations at the sites. The number of undivided caudals found on an individual was less than the ones found in the young of the litters; a row of two and three were frequent. The range in the number of undivided caudals was 1-6, and no sexual preference was noted (Table 6.15).

Table 6.14—The Range of Occurrence of Upper Labial Anomalies in *T. sirtalis parietalis* at the Sites

Sites	N	Range	Percentage of Population
Jette Lake	8	7 -7	
	1	7°-7	10
	1	8°-8	
	2	8 -7	
	4	8 -8	
	4	7 -8	
Smith Lake	9	7 -7	
	1	7'-7	
	1	7'-8	21
	1	7'-8'	
	1	7 -8	
Woods Pond	9	7 -7	
	2	7'-7	
	1	7 -7'	
	1	8'-7°	22
	1	8'-8'	
	1	8'-8	
	5	8 -7	
	3	8 -8	
	4	7 -8	
Lake Rogers	16	7 -7	
	2	7 -7'	
	1	7'-7'	
	1	8'-7'	
	1	8 -7'	23
	1	7 -8'	
	1	7'-8'	
	1	7 -8	
	4	8 -7	
	3	8 -8	
Jette Pond	5	7 -7	
	4	7'-7'	27
	1	7'-7	
	4	7 -8	
	2	8 -7	
	1	8 -8	
	1	8 -9	

'=7th scale had extra scale beside it.

°=6th scale had extra scale beside it.

Table 6.15—The Distribution of the Occurrence of Undivided Caudals in *T. elegans vagrans* at the Sites

Sites	Number of Undivided Caudals	Female	Male	Juvenile	Total	Percentage of Total
Woods Pond	0	1	1	1	3	0
Jette Lake	0	11	9	13	33	5
	2	0	0	1	1	
	4	0	1	0	1	
Lake Rogers	0	9	7	20	36	14
	1	0	2	0	2	
	2	0	0	1	1	
	3	2	0	1	3	
Mud Lake	0	6	6	4	16	11
	2	0	1	0	1	
	3	0	1	0	1	
Jette Pond	0	5	3	11	19	5
	3	1	0	0	1	
Smith Lake	0	2	1	4	7	12
	6	0	1	0	1	

Table 6.16—Color Variation in *Thamnophis sirtalis parietalis*

Ventral Color	Sites						Percentage
	Woods Pond	Jette Lake	Lake Rogers	Jette Pond	Smith Lake	Mud Lake	
Orange	8	1	0	2	2	0	12
Blue	0	0	0	0	1	0	1
Black	3	0	1	1	2	0	6
White, Yellow	16	19	33	15	9	0	81

Color has been used for standard identification of species; however, different color phases of the same species have been identified. This has been associated with the ability of an organism to adapt to its environment and blend in with the surrounding colors. Color was studied in both species. T. elegans did not differ in color to a great extent. Only at one site, Mud Lake, were the individuals distinctly brown with a very faint dorsal stripe.

T. sirtalis was consistent in dorsal color, but it had much variation in the coloration of its ventrals and caudals. In reptiles, the color on the underside has been correlated with sexual dimorphism in the males; or in the case of aquatic species, the underside to predators acts as a camouflage agent. The actual reason for the color difference seen in these populations was not attributed to either dimorphism or camouflage.

The color in the ventrals and caudals ranged from white to yellow, to orange, blue or black. White or yellow were most common. Of all the red-sided garter snakes, 81 percent were white and yellow on the ventral surface; 12 percent were orange; 6 percent were black; and 1 percent was blue. As an aside, the population at Woods Pond had the highest percentage (30) with orange coloration (Table 6.16).

GEOGRAPHIC VARIATION

The collection of data on the red-sided garter snake and the wandering garter snake centered at two locations respectively, Kicking Horse Reservoir (T20N, R20W, S25 and 36, Lake County) and Nine Pipe National Wildlife Refuge (T20N, R20W, S33, Lake County) in the study by Todd (1965). Because of the regional emphasis, 71 out of the 97 red-sided garter snakes were from one locus, and 89 out of the 116 wandering garter snakes were from the other locus.

Although Todd did not consider his sites separately, he listed the labial counts of both species in descending order of frequency. By comparing this to a similar list from this study, 24 different types of counts were found in the red-sided garter snake. In Todd's study, 21 different types were found; in this study, 16 different types were found. Thirteen types were common to both studies. As should be expected, both lists shared the most frequent counts in the first three positions (Tables 6.17 and 6.6).

There were also 24 different types in the wandering garter snake. In Todd's study, 16 different types were reported; in this study, 18 types were reported. Ten types overlapped in both studies. As in the red-sided garter snake, the three counts that made up the highest percentage in this study were the same in Todd's study (Tables 6.18 and 6.5).

In both species, the largest percentage had the typical counts for the species. In Todd's study, these percentages were 67 percent for the wandering garter snake and 44 percent for the red-sided garter snake; in this study, the corresponding figures were 55 percent and 35 percent. Even though both species exhibited the same number of categories of variation, a greater percentage of individuals in the red-sided garter snake deviated from the typical count.

The labial counts were also split into upper and lower categories in order to investigate the range of counts. The typical counts in _T. elegans vagrans_ comprised 76 percent of the upper labials and 67

Table 6.17—Labial Variation in *Thamnophis sirtalis parietalis* (Todd)

Count	Number of Specimens	Percentage
(10–10) (7–7)	43	44.0
(10–10) (7–8)	11	11.0
(10–10) (8–7)	6	6.0
(10–10) (8–8)	2	2.0
(10– 9) (7–9)	1	1.0
(9–10) (7–7)	6	6.0
(9–10) (7–8)	5	5.0
(9–10) (8–8)	1	1.0
(10– 9) (7–7)	5	5.0
(10– 9) (8–7)	1	1.0
(10– 9) (7–8)	2	2.0
(11–10) (7–7)	1	1.0
(10–11) (7–8)	3	3.0
(10–11) (8–8)	1	1.0
(10–11) (7–7)	2	2.0
(10–11) (8–7)	1	1.0
(9– 9) (7–8)	1	1.0
(9– 9) (8–8)	1	1.0
(9– 9) (7–7)	2	2.0
(9–11) (8–7)	1	1.0
(11–11) (8–8)	1	1.0

Table 6.18—Labial Variation in Thamnophis elegans vagrans (Todd)

Count	Number of Specimens	Percentage
(8–8) (10–10)	78	67.0
(8–7) (10–10)	3	3.0
(7–8) (10–10)	12	10.0
(7–7) (10–10)	2	2.0
(8–8) (9–10)	6	5.0
(8–7) (9–10)	1	.8
(8–8) (10– 9)	4	3.0
(8–7) (10– 9)	1	2.0
(8–8) (10–11)	3	3.0
(8–8) (11–11)	1	.8
(7–8) (10–11)	1	.8
(7–8) (9– 9)	1	.8
(7–7) (10– 9)	1	.8
(9–8) (10–10)	1	.8
(7–7) (9– 9)	1	.8

percent of the lower labials in this study; and 79 percent and 83 percent in Todd's study (Table 6.19). In _T. sirtalis parietalis_, the typical counts comprised 52.5 percent of the upper labials and 68.5 percent of the lower labials; and 59 percent and 65 percent in Todd's study (Table 6.20). Although the figures were higher when the labial counts were treated individually, the trend stated in the above paragraph was demonstrated.

The range in Todd's study for ventrals and caudals for _T. elegans vagrans_ were correspondingly 160-182 and 61-87. The greatest number of individuals fell within the category of 166-170 in ventrals and 66-70 in caudals. The majority in this study differed in the caudal range, i.e. between 71-75 (Table 6.10).

For _T. sirtalis parietalis_, the range of ventral scale count was 159-181 and the caudal range was 61-93. Most of the individuals were included in the ventral category of 166-170, and in the caudal category of 66-70. Both of these differed from the present study in which the range of 161-165 included the greatest majority for ventral count and 76-80 for the caudal count. These results may be attributed to the fact that Todd was working with a larger number of snakes; however, the sum total of all the snakes is covered in the following section as the primary indication of the genetic variation within scale counts (Table 6.11).

The dorsal row count of Todd's work differed for both species from that of this study, both in the variety found among the snakes and the count that was the most prevalent (Tables 6.12 and 6.13). In Todd's study the count, 19-20-17, was the most common in the red-sided garter snake, and 20-20-17 in the wandering garter snake. An interesting fact was that the counts, 19-19-17 and 19-19-16, which have been used to

Table 6.19—Distribution of Lower and Upper Scale Count in *T. elegans vagrans* (Todd)

Lower (Right-Left)	N	Percentage	Upper (Right-Left)	N	Percentage
(10-10)	96	83.00	(8-8)	92	79.00
(9-10)	7	7.40	(8-7)	4	3.50
(10- 9)	6	5.10	(7-8)	15	14.00
(9- 9)	2	1.40	(7-7)	4	2.90
(10-11)	4	2.90			
(11-11)	1	.74			

Table 6.20—Distribution of Lower and Upper Scale Count in *T. sirtalis parietalis* (Todd)

Lower (Right-Left)	N	Percentage	Upper (Right-Left)	N	Percentage
(10-10)	63	65.0	(7-7)	58	59.0
(10- 9)	9	9.0	(8-7)	11	11.0
(9-10)	11	12.0	(7-8)	21	22.0
(10-11)	7	7.0	(8-8)	6	6.0
(9- 9)	4	4.0	(7-9)	1	1.0
(9-11)	1	1.0			
(11-11)	1	1.0			
(11-10)	1	1.0			

describe the red-sided garter snake (Schmidt and Davis 1941), were quite frequent; but that a count, 19-20-17, was more frequent. The 20-20-17 count, however, followed the specific description of the other species.

Anomalies such as extra incomplete scales between the upper labials in T. sirtalis were not mentioned by Todd, and most of his snakes were not sexed, so that any further corollaries could not be made between the two studies. However, Todd's study did act as an invaluable reference because of the large samples of snakes analyzed. Therefore, any specific traits of garter snakes in western Montana could be more easily qualified.

ANALYSIS OF PRESERVED SPECIMENS AND SUMMARY OF ANALYSES

An analysis of some museum specimens, which were collected between 1947-1951 by Dr. R. B. Brunson, provided an opportunity to correlate the present genetic traits with the genetics of earlier generations of the same species (Tables 6.21 and 6.22). The regularity in the typical counts of both species was investigated. Sixty-three percent of the 19 wandering garter snakes had the typical count. This was also true for 44 percent of the 16 red-sided garter snakes.

Nine different categories of labial counts were found in the red-sided garter snake. Eight of the nine counts were common to Todd's and this study's lists; the odd count was rather unusual. It was (6-7) (10-10).

Five categories were found in the wandering garter snake, and three of the more common counts were present in the other two studies, also. The other two counts were present in either one or the other study.

Table 6.21—Data on Museum Specimens (*T. sirtalis parietalis*)

Collection Number	Sex	Total Length (cm)	Snout-Vent Length (cm)	Upper Labials	Lower Labials	Dorsal Row Count	Ventrals	Caudals
R-3050(2)	♀	110	87.0	(7 -7)	(10-10)	19-18-17	170	73
R-150(2)	♂	63	49.0	(7 -8)	(10-10)	20-19-17	160	67
	♂	61	44.0	(7'-7)	(10-10)	19-18-17	161	79
R-554(2)		59.5	—	(7 -7)	(9-10)	20-19-17	172	70
	♂	56	42.0	(7 -7)	(10- 9)	19-18-17	177	75
R-352(3)	♂	61.5	44.5	(8 -8)	(10-10)	19-17-17	163	77
	♂	46.5	35.5	(7'-7)	(10-10)	19-19-17	166	64
	gravid ♀	57	45.5	(8 -7)	(10-10)	19-19-17	165	46
R-249(4)	♂	72	53.5	(7 -7)	(10-10)	19-19-17	165	76
	♂	71	52.0	(7 -7)	(10-10)	20-18-17	166	87
	♀	45.5	31.5	(8 -7)	(10-10)	18-19-17	161	79
	gravid ♀	73	53.0	(7'-8')	(9-10)	19-17-17	158	81
R-1049(4)	♂	62	44.0	(7 -7)	(10-10)	19-19-17	160	83
R-1049	♀	32	23.0	(6 -7)	(10-10)	19-19-17	163	86
R-3150	♀	32	24.5	(8 -7)	(10- 9)	18-17-17	162	83
R-1149	♂	21	15.5	(7 -7)	(10-10)	19-16-17	155	71

Collection Number	Date Collected	Locus		
R-150	March 1950	Kicking Horse Reservoir	T20N-R19W-S36	Lake
R-249	June 1949	McDonald Cirque	T19N-R19W-S11	Lake
R-352	1952	Bison Range	T19N-R21W-S27	Lake
R-554	1954	Bonner	T14N-R17W-S13	Missoula
R-1049	July 1949	Lake Rogers	T27N-R23W-S$\frac{30/29}{31/32}$	Flathead
R-1149	1949	Jette Lake	T23N-R21W-S14	Lake
R-3050	July 1950	North Crow Cirque	T21N-R18W-S18	Lake
R-3150	July 1950	Lake Rogers	T27N-R23W-S30	Flathead

'=7th scale had extra scale beside it.

Table 6.22—Data on Museum Specimens (T. elegans vagrans)

Collection Number	Sex	Total Length (cm)	Upper Labials	Lower Labials	Dorsal Row Count	Ventrals	Caudals
R-851	♀	66.0	(8-8)	(9-10)	20-19-16	163	70
R-3350(2)	♀	55.0	(8-8)	(10-10)	21-21-16	155	23
	♀	72.0	(8-8)	(9-10)	21-20-15	169	74
R-2850(3)	♀	33.0	(8-8)	(10-10)	21-21-17	169	84
	♂	40.0	(8-8)	(10-10)	20-20-17	164	83
R-448	♀	56.5	(8-8)	(10-10)	21-20-16	168	87
R-1152	♀	43.0	(8-7)	(10- 9)	20-21-16	170	71
R-1047	♀	68.0	(8-8)	(10-10)	20-21-17	163	62
R-252(3)	♀	77.0	(8-8)	(10-10)	21-21-17	172	77
R-2250	♀	55.0	(7-8)	(10-10)	21-21-17	177	75
R-2650	♀	63.0	(7-8)	(10-10)	21-21-16	165	75
R-3450	♀	64.0	(8-8)	(10-10)	20-21-16	165	74
R-4350(2)	♂	55.0	(8-8)	(10-10)	22-17-17	179	85
R-3250	♂	62.0	(8-8)	(10-10)	20-21-17	172	84
R-3550	♀	58.5	(7-8)	(9-10)	22-21-17	169	46
R-4050(4)	♀	75.0	(8-8)	(10-10)	20-21-15	170	70
	♀	62.0	(8-8)	(10-10)	21-18-16	176	71
R-3950(2)	♂	57.0	(7-8)	(10-10)	21-21-17	168	79
R-4650	♂	49.0	(8-8)	(10-10)	21-21-16	163	76

Collection Number	Date Collected		Locus		
R-252		1952	——	——	——
R-448	May	1948	Kitchen Creek	T11N-R16W-S19	Missoula
R-851	May	1951	Pond	T12N-R20W-S22	Missoula
R-1047	May	1947	UM Biological Station	T24N-R19W-S4	Lake
R-1152		1952	——	——	——
R-2250	July	1950	Camas Creek	T33N-R19W-S23	Flathead
R-2650	Aug.	1950	St. Mary's Lake	T17N-R18W-S6	Lake
R-2850	June	1950	McDonald Cirque	T19N-R19W-S11	Lake
R-3250	June	1950	Cottonwood Creek	T15N-R13W-S29	Powell
R-3350	June	1950	Mud Lake	T21N-R19W-S2	Lake
R-3450	June	1950	Mud Lake	T21N-R19W-S2	Lake
R-3550	July	1950	Estes Lake Trail	T26N-R19W-S20	Lake
R-3950	Sept.	1950	Upper Holland Lake	T20N-R15W-S29	Missoula
R-4050	Sept.	1950	Gyp Mountain	T20N-R15W-S3	Missoula
R-4350		1950	Rock Creek	——	——
R-4650	June	1950	Whitefish Reservoir	T30N-R22W-	Flathead

As the percentages indicated in all the analyses, there had been a consistent regularity within the population of T. elegans vagrans and an almost predictable large variety of counts in the T. sirtalis parietalis.

The range in the number of ventrals of the red-sided garter snake was 155-177, and the range in the caudals was 64-87. The most regularly occurring counts fell within the same ranges as the snakes within this current study. These ranges were correspondingly, 161-165 and 76-80. The sum total of all these snakes indicated these count ranges had a high probability of being found; however, the ranges, 166-170 and 66-70, were the typical counts in this regional area of the state (Table 6.11).

The range in the number of ventrals of the wandering garter snake was 155-179, and the range in the caudals was 62-87. The most frequent ventral range, 166-170, was the same in all the studies. The caudal range, 71-75, contained the highest percentage of individuals in the museum specimens and in the ones from this study. By combining the number of snakes within each category from each study, the range, 66-70, for the caudal count became the most typical. The count 166-170 was uniformly the most typical one for ventrals (Table 6.10).

The most common dorsal row count, 21-21-17, agreed with the species description for the wandering garter snake. Four of 12 different categories were also in a list of dorsal row counts from this study; there were a total of 19 different categories that were noted from this study and in the museum specimens. (Todd's samples were not included in the sum total because his counting method was not described. Therefore, it may or may not have influenced an incorrect judgment concerning the variety of categories which occur in this species. For visual comparison, Table 6.12 has been constructed.)

The count, 19-19-17, which was used in the description of the red-sided garter snake, was prevalent in the preserved specimens. Two of the eight counts, which were observed, were mutually shared between this study and the preserved specimens. A total of 14 categories were found and have been listed with the data from Todd for cross-reference purposes (Table 6.12).

The occurrence of extra incomplete scales in the upper labials of the T. sirtalis parietalis seemed to infer with high probability that Todd may not have noticed their presence. Twenty-five percent of the 16 preserved snakes had this anomaly (Table 6.23). In a sample of 35 preserved wandering garter snakes, seven, or 20 percent, of the snakes had undivided caudals. The most common occurrence was one undivided caudal; the range, however, was from 1-4 consecutive scales (Table 6.24). No sexual preference was noted. It can be concluded that its absence in Todd's report was because it was not in the scope of his report.

Table 6.23—Museum Specimens with Extra Labials (*T. sirtalis parietalis*)

Upper Labial Count	Number of Individuals	Percentage
7 -7	7	44
8 -7	2	13
7 -8	1	6
8 -8	1	6
6 -7	1	6
7'-7	2	13
7'-8'	1	6
8'-7'	1	6

'=7th scale had extra scale beside it.

Table 6.24—Museum Specimens with Undivided Caudals (*T. elegans vagrans*)

Collection Number	Sex	Number of Undivided Caudals	Specific Location of Caudals
R-547	♂	2	4-5
R-747	♂	4	1-4
R-749	♂	1	2
R-1248	♀	1	2
R-1649	♀	1	2
R-1650	juvenile	1	4
R-2850(3)	♀	2	2-3

Collection Number	Date Collected	Locus		
R-547	May 1947	Gold Creek Pond	T14N-R17W-S13	Missoula
R-747	May 1947	Daphnia Pond	T26N-R19W-S6	Flathead
R-749	1949	Snowshoe Mine	——	——
R-1248	Aug. 1948	UM Biological Station	T24N-R19W-S4	Lake
R-1649	Aug. 1949	Trout Creek	T16N-R26W-S23	Mineral
R-1650	May 1950	River (Hamilton Mountain)	——	——
R-2850(3)	June 1950	McDonald Cirque	T19N-R19W-S11	Lake

CHAPTER VII

SUMMARY

A preliminary survey of the use of garter snakes as indicators of contaminated region was completed. This survey entailed the analysis for lead and mercury in the tissues of *Thamnophis sirtalis parietalis* and *Thamnophis elegans vagrans* from six select sites in western Montana for background data, from the vicinity of East Helena's ASARCO smelter, and from the area of the stamp mill along Fred Burr Creek at Phillipsburg. Analyses were done by the use of the Beckman Atomic Absorption attachment for the Model DB Spectrophotometer.

Both the areas of the ASARCO smelter and Fred Burr Creek's stamp mill have been areas of chronic pollution for the respective heavy metals, lead and mercury. These areas served as contamination sites. The snakes from East Helena ranged in lead concentration from .1–.7 w/w ppm (mean: .39) for liver and from .36–.8 w/w ppm (mean: .55) for muscle. The snakes from the six collection sites ranged from .02–.3 w/w ppm (mean: .12) for liver and from .02–.40 w/w ppm (mean: .18) for muscle. Two obvious trends were noted: (1) the *Thamnophis* at East Helena were ingesting and absorbing more lead from their environment than were the snakes from the other sites; (2) the muscle tissues concentrated the lead more than the livers.

The results from the mercury analyses of the garter snakes were for the liver, .01–1.0 w/w ppm (mean: .26) and for the muscle, .05–.49

w/w ppm (mean: .24). The samples from the snakes at Fred Burr Creek were higher in content; the carcass of a juvenile _Thamnophis elegans vagrans_ contained .77 w/w ppm Hg, and the muscle of the gravid _Thamnophis sirtalis parietalis_ contained .39 w/w ppm Hg. Complementary samples from the gravid snake were lost because they exploded during the digestion process in the furnace.

Although there were few snake samples, the auxiliary ranid samples analyzed from Fred Burr Creek indicated the potential of the herpetofauna as monitors for pollution. The ranids ranged in mercury from 1.46–2.88 w/w ppm (mean: 2.27); the concentration of mercury in frogs increased as one came closer to the stamp mill. This trend was reinforced by a study of Van Meter (1974) in which he found that the water and trout likewise became higher in mercury as one collected upstream toward the mill.

Frogs and one fish were used to supplement the East Helena survey. The frogs ranged in lead from .43–4.8 w/w ppm (mean: 1.58), and the fish contained .94 w/w ppm. There was no correlation in the concentration of lead in the samples and the distance from the ASARCO stack. Mice have been shown to contain higher concentrations of lead, i.e. in livers, 7.6 w/w ppm, and in kidneys, 42.5 w/w ppm (Gordon 1972).

The lower concentrations of heavy metals in the snakes as compared to known food items (frogs, fish, mice) was attributed to the type of exposure each animal had in its microhabitat, and the seasonal and regional emphasis of prey of _Thamnophis_. In the aquatic environment, the exposure was concluded as threefold by Fulkerson _et al_. (1974): (1) adsorption of mercury on the nucous layer of the skin, (2) ingestion of food, (3) assimilation of mercury into the bloodstream as a consequence of digestion.

Selection of food types by the species of garter snakes was demonstrated as similar but different at the six collection sites (Chapter IV); the multi-faceted reason for the diet selection of garter snakes at Fred Burr Creek and East Helena was not researched in this study. However, the extent to which heavy metals caused sub-lethal results was attributed to the rate of ingestion and the rate of deterioration of the metal within the snake. Therefore, the food selection of a snake directly influenced the toxicity of the heavy metal within the snake. A topic for future research was the interrelationship of heavy metals in garter snakes and their food items.

An investigation into the use of preserved specimens of herpetofauna for background levels of heavy metals was undertaken. Phenomena were encountered that indicated more research must be done in the area of chemical reactions between formalin and alcohol with heavy metals with a biological specimen. For instance, an unexpectedly high concentration of lead was found in one museum sample from Jette Lake; this snake collected in 1949 had .25 ppm Pb. The snakes which were collected and frozen in 1974 had .03 and .07 ppm Pb; the concentration of lead in the older sample must have been caused by chemical interactions similar to those found in another study by Gibbs et al. (1974).

The selection of food items and the activity temperatures of both species of garter snakes were investigated. Five of the six collection sites had sympatric populations of Thamnophis. No obvious activity temperature range was preferred by one species or the other; however, the ability to maintain a body temperature above the ambient temperature was noted among Thamnophis. Selection of food was an important ecological isolating mechanism.

Seasonal abstinence of eating was observed in early summer by a substantial percentage of each population; T. s. parietalis demonstrated a principal dependency on Haemopis grandis and amphibians, which were mainly Bufo boreas and Ambystoma macrodactylum. T. e. vagrans had an emphasis on leeches, which were H. grandis and Erpobdella punctatum, and slugs, which were Prophysaon and Deroceras. Leeches were the mainstay of both species throughout the summer season.

The wandering garter snake species acted as an opportunistic predator at Jette Pond; it consistently ate combinations of prey items. Larval salamanders and leeches were found together in the stomach; other combinations were found also. At Smith Lake and Jette Lake, the ingestion of red shiners by T. elegans vagrans, and not by T. sirtalis parietalis, was another food selection which created a situation for coexistence without dietary overlap.

The population sizes at the six collection sites were measured by a mark and recapture method. The results were analyzed statistically by using a method which was designed by Schnabel (1938). The estimates increased in accuracy as the numbers of recaptures increased; the model did not take into consideration an individual which was recaptured more than one time. The main assumption of the model was that there was no recruitment of individuals from the surrounding environment or emigration of individuals to the surrounding environment.

The model was ineffective in estimating the size of the most disturbed site such as Smith Lake. Biological observations were used to support or nullify answers of the statistical model for each site. However, the biological and intuitive observations of the author indicated that population size could be used as a valid monitor of environmental disturbances. As for further uses of the mark-recapture method, it could be used cooperatively with pollution studies in disturbed areas in order to procure a more complete picture of the effects of pollution on a species.

A cursory review of the genetical variation of scale counts and color pattern was completed. Both the red-sided garter snake and the wandering garter snake displayed a wide range of counts in labials; the red-sided garter snake had extra scales among the upper labials and the wandering garter snake had a frequent occurrence of undivided caudal scales.

The genetic study was a research project which incorporated data from the litters of three gravid Thamnophis, from the snakes from the six collection sites and from University of Montana museum samples. A statistical analysis of a former study by Todd (1965) was included in order that a more accurate picture of the variation in western Montana could be obtained. The data from the genetic study were included in toto in the Appendix so that further research in this region would be pursued.

Snakes of the genus Thamnophis have been used as biological indicators in many states. This study has created a basis for investigation in many areas of the biology of garter snakes. Most significant was their use in heavy metal analyses; it was hoped that more environmental studies in Montana will include herpetofauna, as a result of this thesis.

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APPENDIX

LABORATORY DATA ON RETAINED GRAVID *THAMNOPHIS*

and

VARIATIONS OF LABIAL COUNTS AT SITES

Laboratory Data on Retained Gravid Thamnophis

Species	Brood	Site	Dates Maintained	Number of Days	Number in Litter	Number of Live Snakes	Number Partially Developed Embryos	Number of Eggs
T. e. vagrans	I	Dahl Lake	June 20 - July 2, 1974	18	12 ♂, 6 ♀	None	All	1
T. e. vagrans	II	Breneman's Marsh	June 21 - Sept. 11, 1974	82	4 ♂, 8 ♀	4 ♂, 7 ♀	1 ♀, 1 (unknown)	2
T. s. parietalis	III	Smith Lake	June 20 - Aug. 26, 1974	67	6 ♂, 7 ♀	6 ♂, 7 ♀	None	None

Woods Pond

Thamnophis sirtalis parietalis			Thamnophis elegans vagrans		
Labial Count	N	Percent	Labial Count	N	Percent
(7-7) (10-10)	8	29	(8-8) (10-10)	3	100
(7-7) (9-10)	2	7			
(7-7) (9- 9)	1	4			
(7-8) (10-10)	2	7			
(7-8) (10- 9)	1	4			
(8-7) (10-10)	5	18			
(8-7) (10- 9)	1	4			
(8-8) (10-10)	3	11			
(7-7) (10- 9)	2	7			
(8-8) (11-10)	1	4			
(7-8) (9-10)	1	4			

Thamnophis sirtalis parietalis						Thamnophis elegans vagrans					
Upper Labial	N	Percent	Lower Labial	N	Percent	Upper Labial	N	Percent	Lower Labial	N	Percent
(7-7)	12	44	(10-10)	19	70	(8-8)	3	100	(10-10)	3	100
(7-8)	4	15	(9-10)	2	7						
(8-8)	5	19	(9- 9)	1	4						
(8-7)	6	22	(10- 9)	4	15						
			(11-10)	1	4						

Jette Pond

Thamnophis sirtalis parietalis

Labial Count	N	Percent
(7-8) (10-10)	2	12
(7-8) (11-10)	1	5
(7-8) (9- 9)	8	46
(7-7) (10-10)	1	5
(7-7) (10- 9)	2	12
(8-7) (10-10)	1	5
(8-8) (10-10)	1	5
(8-9) (10-10)	1	5
(7-7) (9- 9)	1	5

Thamnophis elegans vagrans

Labial Count	N	Percent
(8-8) (10-10)	11	55
(8-8) (9-10)	2	10
(8-8) (10- 9)	1	5
(8-8) (9- 9)	1	5
(7-7) (10-10)	1	5
(7-7) (9- 8)	1	5
(7-7) (9-10)	1	5
(7-8) (10-10)	2	10

Thamnophis sirtalis parietalis

Upper Labial	N	Percent	Lower Labial	N	Percent
(7-8)	4	22	(10-10)	14	78
(7-7)	10	56	(11-10)	1	5
(8-9)	1	5	(9- 9)	2	12
(8-7)	2	12	(10- 9)	1	5
(8-8)	1	5			

Thamnophis elegans vagrans

Upper Labial	N	Percent	Lower Labial	N	Percent
(8-8)	15	75	(10-10)	14	70
(7-7)	3	15	(9-10)	3	15
(7-8)	2	10	(9- 9)	1	5
			(10- 9)	1	5
			(9- 8)	1	5

Jette Lake

Thamnophis sirtalis parietalis

Labial Count	N	Percent
(7-7) (10-10)	7	35
(7-7) (10- 9)	1	5
(7-8) (9-10)	2	10
(7-8) (10-10)	2	10
(7-7) (9- 9)	1	5
(8-8) (9-10)	1	5
(8-8) (10-10)	3	15
(8-8) (9- 9)	1	5
(8-7) (10-10)	2	10

Thamnophis elegans vagrans

Labial Count	N	Percent
(8-8) (10-10)	24	68
(8-8) (9-10)	2	6
(8-8) (10- 9)	2	6
(8-8) (9- 9)	2	6
(8-7) (10-10)	3	8
(7-8) (9-10)	1	3
(7-7) (9-10)	1	3

Thamnophis sirtalis parietalis

Upper Labial	N	Percent	Lower Labial	N	Percent
(7-7)	9	45	(10-10)	14	70
(7-8)	4	20	(10- 9)	1	5
(8-7)	2	10	(9-10)	3	15
(8-8)	5	25	(9- 9)	2	10

Thamnophis elegans vagrans

Upper Labial	N	Percent	Lower Labial	N	Percent
(8-8)	30	86	(10-10)	27	77
(8-7)	3	8	(9-10)	4	11
(7-8)	1	3	(10- 9)	2	6
(7-7)	1	3	(9- 9)	2	6

Lake Rogers

Thamnophis sirtalis parietalis			Thamnophis elegans vagrans		
Labial Count	N	Percent	Labial Count	N	Percent
(7-7) (10-10)	10	33	(8-8) (10-10)	22	55
(7-7) (10- 9)	2	6	(8-8) (9-10)	4	9
(7-7) (9-10)	6	20	(8-8) (10- 9)	1	3
(7-7) (9- 9)	2	6	(8-7) (10-10)	1	3
(7-8) (10-10)	2	6	(8-7) (8- 9)	1	3
(8-8) (10-10)	2	6	(7-8) (9- 9)	1	3
(8-8) (9- 9)	1	3	(7-8) (10-10)	2	5
(8-7) (10-10)	5	17	(7-8) (9-10)	1	3
(8-7) (10- 9)	1	3	(7-7) (9- 9)	3	7
			(7-7) (8- 8)	1	3
			(6-8) (8- 8)	1	3
			(9-8) (10-10)	1	3

Thamnophis sirtalis parietalis						Thamnophis elegans vagrans					
Upper Labial	N	Percent	Lower Labial	N	Percent	Upper Labial	N	Percent	Lower Labial	N	Percent
(7-7)	19	61	(9- 9)	4	13	(8-8)	27	69	(10-10)	26	66
(7-8)	3	10	(10- 9)	3	10	(8-7)	2	5	(10- 9)	1	3
(8-7)	6	19	(10-10)	18	58	(7-8)	4	10	(9-10)	5	13
(8-8)	3	10	(9-10)	6	19	(7-7)	4	10	(9- 9)	4	10
						(6-8)	1	3	(8- 9)	1	3
						(9-8)	1	3	(8- 8)	2	5

Smith Lake

Thamnophis sirtalis parietalis

Labial Count	N	Percent
(7-8) (10-10)	4	29
(7-7) (10-10)	8	51
(7-7) (9-10)	2	14

Thamnophis elegans vagrans

Labial Count	N	Percent
(8-8) (10-10)	3	36
(8-8) (9-10)	1	13
(8-8) (9- 9)	1	13
(8-8) (10- 9)	1	13
(7-8) (10-10)	2	25

Thamnophis sirtalis parietalis

Upper Labial	N	Percent	Lower Labial	N	Percent
(7-7)	10	71	(10-10)	12	86
(7-8)	4	29	(9-10)	2	14

Thamnophis elegans vagrans

Upper Labial	N	Percent	Lower Labial	N	Percent
(8-8)	6	75	(10-10)	5	61
(7-8)	2	25	(9-10)	1	13
			(10- 9)	1	13
			(9- 9)	1	13

Mud Lake

Labial Counts	N	Percent
T. e. v.		
(8-8) (10-10)	9	50
(7-8) (9-10)	1	6
(7-8) (9- 9)	1	6
(7-7) (10-10)	1	6
(8-8) (8-10)	2	11
(8-8) (9- 9)	1	6
(8-8) (8- 8)	1	6
(9-8) (10-10)	1	6
(7-8) (10-10)	1	6

Upper Labial	N	Percent	Lower Labial	N	Percent
T. e. v.					
(8-8)	13	72	(10-10)	12	66
(7-8)	3	16	(9-10)	1	6
(7-7)	1	6	(9- 9)	2	11
(9-8)	1	6	(8- 8)	1	6
			(8-10)	2	11