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An Ecological Study of the Cumberland Plateau Salamander, <u>Plethodon</u> <u>kentucki</u> Mittleman, in West Virginia

A Thesis

Submitted to

the Faculty of the Graduate School

of Marshall University

In Partial Fulfillment

of the Requirements for the Degree

Master of Science

by

Jeffrey E. Bailey

This thesis was accepted on $\frac{4/23}{92}$ Year Month Day as meeting the researh requirements for the Master's Degree. Advisor: Thomas K. Canley () Department of Biological Sciences Lonard Deutsch

Dean of the Graduate School

487055

ii

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iii

Table of Contents

	Page
Title Page	i
Approval of Examining Committee	ii
Acknowledgements	iii
Table of Contents	iv
List of Figures	vi
List of Tables	ix
Abstract	1
Introduction	3
Description and Comparison of the Species	5
Description of the Study Area	14
Chapter I - Range and Distribution of <u>Plethodon</u> <u>kentucki</u>	
in West Virginia	20
Introduction	20
Methods and Materials	22
Results	22
Discussion	24
Summary	31
Chapter II - Aspects of the Natural History of <u>Plethodon</u>	
kentucki in West Virginia	34
Introduction	34
Methods and Materials	35
Results	41
Discussion	67
Summary	78

. .

iv

Chapter III - Competition as a Possible Factor Limiting

the Range of <u>Plethodon</u> <u>kentucki</u> in West

	Vi	irg	ini	a	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	81
Introductio	n .	•••	•	•	•	•	•	•	•	•	٠	•	•	•	•	•	•	•	•	•	•	•	81
Methods and	Mate	eria	als	1	•	•	•	•	•	•	•	•	•	•	•	٠	•	•	•	•	•	•	83
Results .	•••	• •	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•		84
Discussion	•••	•••	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	85
Summary .	•••	•••	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	90
Chapter IV	- A C	Comp	par	at	i	7e	Ar	na]	lys	is	c	f	Pı	:ey	7 5	Se]	.ec	:ti	or	ı			
	bet	wee	en	<u>P1</u>	et	:hc	odo	<u>n</u>	<u>ke</u>	ent	uc	ki	. e	inc	l I	<u>216</u>	eth	100	lor	ī			
	glu	itir	nos	us	j	n	Ŵe	st	: V	/ir	gi	ni	a	•	•	•	•	•	•	•	•	•	92
Introductio	n	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	92
Methods and	Mate	eria	ls		•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	93
Results .	•••	•	•	•	•	•	•		•	•	•	•	•	•	•	•	•	•	•	•	•	•	94
Discussion	•••	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	96
Summary .	•••	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	٠	•			98
Conclusion	•••	•	•	•	•	•	•	•	•	•	•		•	•	•	•	•	•	•	•		•	99
Literature (Cited	ι.	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	102
Vita	• • •	•	•	•	•		•			•	•	•	•	•	•	•			•	•			106

v

List of Figures

1

Figure		Page
1.	Distribution P. <u>kentucki</u> , P. <u>punctatus</u> , P.	
	cylindraceus, P. wehrlei, and P. glutinosus	
	in eastern United States	6
2.	Dorsal view of <u>P. kentucki</u> (top) and <u>P</u> . <u>glutinosus</u>	
	(bottom) showing differences in the size of white	
	spots	9
3.	Lateral view showing flattened head of <u>P. kentucki</u>	
	(left), and robust head of <u>P</u> . <u>glutinosus</u> (right)	10
4.	Ventral view of <u>P</u> . <u>glutinosus</u> (left) and <u>P</u> . <u>kentucki</u>	
	(right)	11
5.	Lateral view of <u>P</u> . <u>kentucki</u> specimen from study site	
	showing profuse white line between fore and hind	
	limb	12
6.	An outline of the study area at Beechfork State Park	
	in Wayne County, West Virginia	15
7.	An outline of the seven quadrats at the study site,	
	showing their locations on the horizontal and	
	vertical line transects	17
8.	An outline of quadrat 8 (1000 m²) showing west-	
	facing, northwest-facing, and southwest-facing	
	slopes	18
9.	The known range and distribution of <u>P. kentucki</u> in	
	West Virginia	23
10.	Geographic distribution of P. glutinosus in West	

vi

	vii
Virginia	25
Mean on-site air and soil temperatures, and number	
of <u>P. kentucki</u> observed for February, March, and	
April 1991	44
Diel activity profile for <u>P</u> . <u>kentucki</u> at Beech Fork	
State Park	46
Relationship between mean air temperature °C (bars	
indicate ± 1 S.E.) and surface density of <u>P</u> . <u>kentucki</u>	
for each month of the study	48
Relationship between mean soil temperature °C (bars	
indicate ± 1 S.E.) and surface density of <u>P</u> . <u>kentucki</u>	
for each month of the study	50
Relationship between mean % air relative humidity	
(bars indicate \pm 1 S.E.) and surface density of <u>P</u> .	
<u>kentucki</u> for each month of the study	52
Relationship between mean % soil moisture (bars	
indicate ± 1 S.E.) and surface density of <u>P. kentucki</u>	
for each month of the study	53
Relationship between mean soil pH (bars indicate ± 1	
S.E.) and surface density of <u>P. kentucki</u> for each	
month of the study	55
Comparison of on-site air and soil temperatures with	
quadrat temperatures for March and April 1991, when	
surface density of <u>P</u> . <u>kentucki</u> was highest	57
Movement of recaptured P. kentucki in quadrats 8 and	
5	62
	Mean on-site air and soil temperatures, and number of <u>P</u> . <u>kentucki</u> observed for February, March, and April 1991

ġ,

20.	Percentages of <u>P</u> . <u>kentucki</u> found under rocks, logs,	
	leaves, and roots	63
21.	Percentage distribution of P. kentucki on southwest-,	
	west-, and northwest-facing slopes	65
22.	Results of the seven behavioral states observed	
	during competition studies between <u>P</u> . <u>kentucki</u> and <u>P</u> .	
	glutinosus	86

. 196 viii

List of Tables

Table	Page
1.	Distinguishing morphological characteristics of
	<u>P. kentucki</u> and <u>P. glutinosus</u> \dots \dots 7
2.	Mean elevation for counties following the New-Kanawha
	River system
3.	Monthly surface abundance of <u>P. kentucki</u> at Beech Fork
	State Park in Wayne County, West Virginia 42
4.	Number of <u>P</u> . <u>kentucki</u> marked in each quadrat at Beech
	Fork State Park
5.	Mean air and soil temperatures °C, air RH %, and
	rainfall for a northeastern (Ritchie) and south-
	western (Wayne) county in West Virginia
6.	Mean CTM values (± 1 S.E.) for <u>P</u> . <u>kentucki</u> and
	<u>P</u> . <u>glutinosus</u>
ʻ7.	Mean dehydration values (± 1 S.E.) for <u>P</u> . <u>kentucki</u>
	and \underline{P} . <u>glutinosus</u>
8.	Percent frequency occurrence of plant species in canopy,
	understory, and shrub layers at Beech Fork State
	Park
9.	Amphibians and reptiles living sympatrically with
	P. <u>kentucki</u> at Beech Fork State Park 68
10.	Distribution of food items in stomachs of 70
	<u>P. kentucki</u> and 79 <u>P. glutinosus</u> \dots 95
11.	Comparison of snout-vent-length and head width for
	P. kentucki and P. glutinosus

Dec.

ABSTRACT

A study was conducted to determine various aspects of the ecology of <u>Plethodon kentucki</u> in West Virginia. Results of studies on range and distribution revealed that <u>P. kentucki</u> is limited to southwestern counties in West Virginia.

A population of <u>P</u>. <u>kentucki</u> at Beech Fork State Park was extremely seasonal with regard to activity. Seasonal activity was not significantly correlated with air temperature, soil temperature, air relative humidity, or soil pH. Seasonal activity was significantly correlated with soil moisture.

Critical Thermal Maxima and dehydration values were not significantly different between <u>P</u>. <u>kentucki</u> and its congener, <u>P</u>. <u>glutinosus</u>, thus each is equally well adapted to temperature and moisture conditions in West Virginia. Environmental conditions (temperature and moisture) do not explain the absence of <u>P</u>. <u>kentucki</u> in northern counties where <u>P</u>. <u>glutinosus</u> is abundant.

<u>Plethodon kentucki</u> was most active between 9:00 and 10:00 PM EST. Linear movements were measured at 1.32 m for a juvenile, and 1.81 m for an adult female. Rocks (47.9%) were utilized as cover objects more frequently than leaves (20.8%) and logs (4.2%). Crevices beneath exposed roots of living trees were important microhabitats for <u>P. kentucki</u> (27.1%). Individuals of <u>P. kentucki</u> were found more frequently on west-facing slopes (44.8%) than on southwest- (31.0%) and northwest-facing slopes (14.6%).

Based on results of laboratory studies, competition occurs between <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> in West Virginia. <u>Plethodon glutinosus</u> was more territorial in nature, and was considered the dominant competitor. Competition apparently plays a key role in limiting the range and distribution of <u>P</u>. <u>kentucki</u> in West Virginia.

The stomachs of 70 <u>P</u>. <u>kentucki</u> and 79 <u>P</u>. <u>glutionsus</u> were analyzed for food contents. Prey selection was significantly correlated, thus indicating a high degree of similarity in feeding habits for the two species. Hymenopterans were found in stomachs of both species more frequently than any other prey type (<u>P</u>. <u>k</u>. 71.4%; <u>P</u>. <u>g</u>. 54.4%), followed by coleopterans (<u>P</u>. <u>k</u>. 40.0%; <u>P</u>. <u>g</u>. 32.9%) and gastropods (<u>P</u>. <u>k</u>. 30.0%; <u>P</u>. <u>g</u>. 30.4%). Food appears to be a resource for which these two species compete.

Head width was not significantly different for the two species. Prey size probably does not partition feeding habits, and thus probably increases the intensity of competition for food between P. kentucki and P. glutinosus.

Introduction

The Cumberland Plateau salamander, <u>Plethodon kentucki</u> Mittleman (1951), is a woodland species inhabiting deciduous forests in the eastern United States. Although it is relatively common throughout its range, many aspects of its ecology are unknown, particularly when compared to several of its congeners. Numerous studies have focused on a closely related species, the slimy salamander, <u>Plethodon glutinosus</u> Green (1818). Semilitsch (1980) monitored populations of <u>P</u>. <u>glutinosus</u> in Maryland and Pennsylvania to determine population structure and density. Highton (1956) gave a detailed description of the life history of <u>P</u>. <u>glutinosus</u> in Florida. Wells and Wells (1976) reported on density and patterns of movement. These and other studies have added to the wealth of general information accumulated on the ecology of this species.

<u>Plethodon kentucki</u> has received little attention since its description in 1951. Studies prior focused on validating its distinctness as a true species by means of morphometric measurements and or electrophoretic analyses (Clay et al. 1955; Maha and Maxson 1983; Highton and MacGregor 1983). Since <u>P</u>. <u>kentucki</u> is similar in appearance and in habitat preference to <u>P</u>. <u>glutinosus</u>, it may have been unnoticed for years by salamander researchers who simply regarded it as the latter species. It is the purpose of this study to determine various ecological aspects of <u>P</u>. <u>kentucki</u> in West Virginia with emphasis on its interactions

with P. glutinosus. The objectives were to (1) determine the range and distribution of P. kentucki in West Virginia, (2) determine various aspects of the natural history of P. kentucki in order to define its ecology in West Virginia, (3) determine if competition exists between P. kentucki and P. glutinosus and, if so, how it might influence the range and distribution of P. kentucki in West Virginia, and (4) determine if resource partitioning is occurring by comparing the feeding habits of P. kentucki and

P. glutinosus.

Description and Comparison of the Species

The Cumberland Plateau salamander, Plethodon kentucki Mittleman (1951), is a lungless salamander and a member of the family Plethodontidae. It is one of five eastern large Plethodon known to occur in West Virginia and the only species known to be restricted to the southwestern portion of the state. The total range of P. kentucki extends from the New River in West Virginia southwest to western Virginia and eastern Kentucky. Two of the remaining four large Plethodon species, P. punctatus Highton (1972), white-spotted salamander, and P. cylindraceus Harlan (1825), white-spotted slimy salamander, occupy small areas in the eastern panhandle of West Virginia with the former extending into northwestern Virginia, and the latter farther south to the The fourth species, P. wehrlei Fowler and Dunn Carolinas. (1917), Wehrle's salamander, lives sympatrically with P. kentucki in West Virginia and extends from southwestern New York to northwestern North Carolina. The fifth species, P. glutinosus Green (1818), slimy salamander, also lives sympatrically with P. kentucki in West Virginia. It has a broader range than P. kentucki (Conant 1975), extending from central New York south to The total ranges central Florida and west to eastern Oklahoma. of each species are presented in Figure 1.

Although very similar morphologically, <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> can be distinguished by using several key characteristics (Table 1). Both are large salamanders that can

Figure 1. Geographic distribution of <u>P</u>. <u>kentucki</u>, <u>P</u>. <u>punctatus</u>, <u>P. cylindraceus</u>, <u>P. wehrlei</u>, and <u>P. glutinosus</u> in eastern United States (adopted from Conant and Collins 1991).

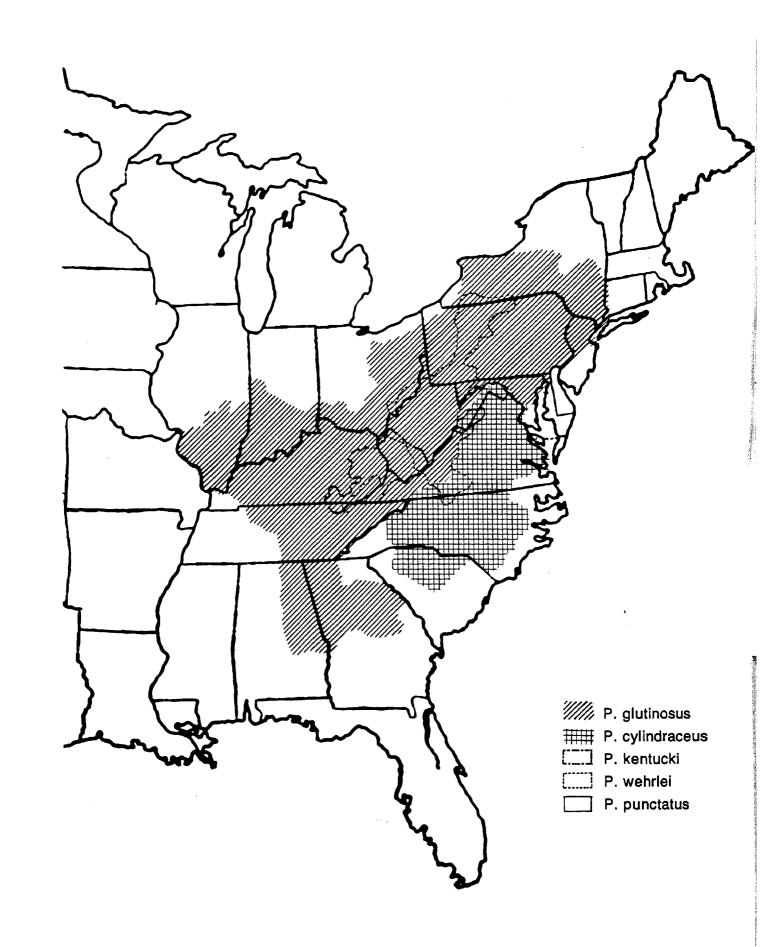


Table 1. Distinguishing morphological characteristics of <u>P. kentucki</u> and <u>P. glutinosus</u> .										
Species	white spots	chin color	hedonic gland	head shape						
<u>P. k</u> .	small	light	large	flattened						
<u>P</u> .g.	large	dark	small	robust						

exceed a SVL (snout to anterior angle of vent) of 80 mm; however, <u>P. kentucki</u> is typically smaller with a maximum SVL range of 60 mm to 77 mm (Highton and MacGregor 1983). The dorsal surface of each is shiny black with scattered small white spots which may be absent or greatly reduced on some individuals (Figure 2). The amount of brassy flecking in the white spots varies and in some cases may be characteristic of a local population. The belly and throat range in color from white to slate gray and black. Typically, both species have 16 costal grooves (Green and Pauley 1987).

Other than being somewhat smaller, <u>P</u>. <u>kentucki</u> is noticeably flatter. Although the entire body appears flattened dorsoventrally, the region exhibiting this characteristic best is the head (Figure 3). Highton and MacGregor (1983) reported that <u>Plethodon kentucki</u> has a lighter chin color than <u>P</u>. <u>glutinosus</u>, a larger mental hedonic gland (Figure 4), and typically has smaller white spots (Figure 2). In some local populations, the lateral white spots of <u>P</u>. <u>kentucki</u> are so numerous they form a profuse white line that extends from the fore to the hind limbs (Figure 5).

<u>Plethodon kentucki</u> and <u>P. glutinosus</u> are woodland species that occupy a wide variety of terrestrial habitats including moist ravines, shale banks, cave entrances, flood plains, and damp hillsides (Green and Pauley 1987). Cover objects utilized as microhabitats include large flat stones, rotting logs, rock crevices, leaf litter, and bases of large trees.

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Figure 2. Dorsal view of P. <u>kentucki</u> (top) and P. <u>glutinosus</u> (bottom) showing differences in the size of white spots.

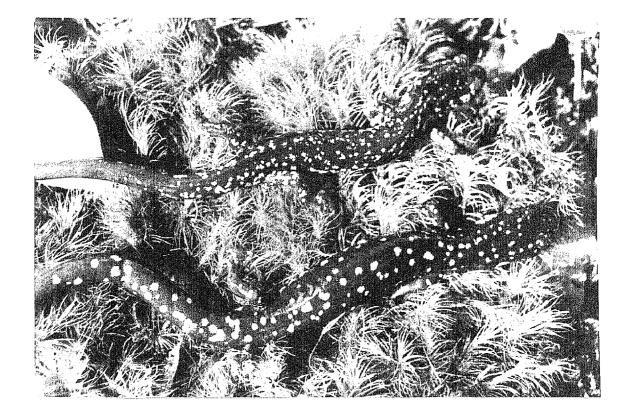


Figure 3. Lateral view showing flattened head of <u>P</u>. <u>kentucki</u> (left) and robust head of <u>P</u>. <u>glutinosus</u> (right).

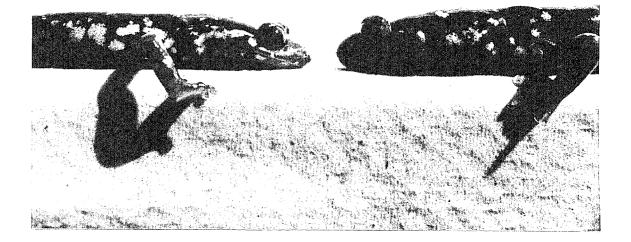
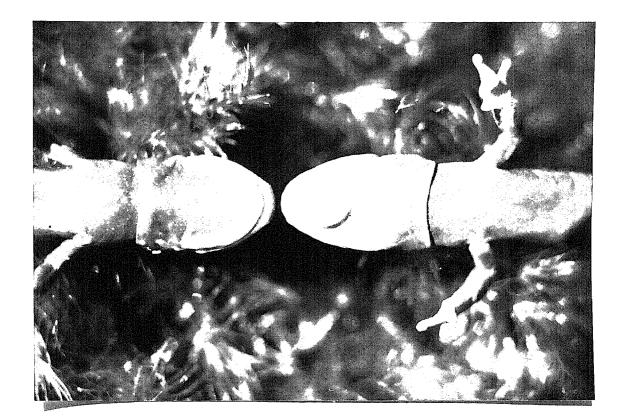


Figure 4. Ventral view of <u>P. glutinosus</u> (left) and <u>P. kentucki</u> (right). <u>Plethodon kentucki</u> has a lighter chin and larger mental hedonic gland (Highton and MacGregor 1983).

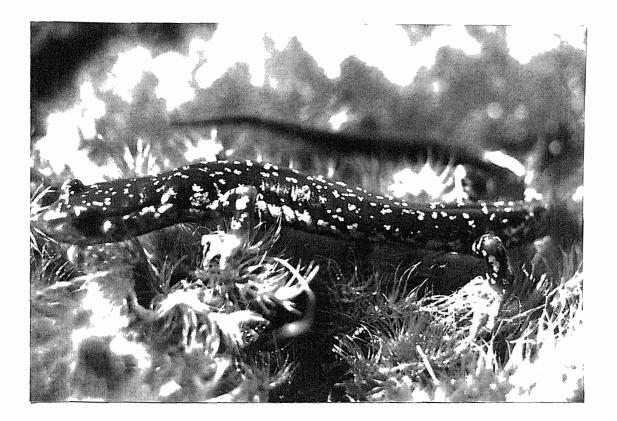


Statistics.

Figure 5. Lateral view of P. <u>kentucki</u> specimen from the study site showing profuse white line between fore and hind limbs.

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Mittleman described P. kentucki based on specimens collected from four localities in southeastern Kentucky (Harlan County on Pine Mountain, Big Black Mountain, near Cumberland, and in Pike County near Virgie) (Highton and MacGregor 1983). Because of the morphological similarity between P. kentucki and P. glutinosus, Mittleman based his description on comparisons of morphological characteristics, vomerine tooth counts, and pigmentation (Maha et al. 1983). Plethodon kentucki was thereafter considered distinct from its closely related congener. However, Clay et al. (1955) reevaluated the material Mittleman used and concluded that there was no evidence to support the contention that it came from two species. Furthermore, they suggested that P. kentucki was simply a local population of P. glutinosus. This idea was not challenged until Highton and MacGregor (1983), and Maha and Maxson (1983) validated P. kentucki as a distinct species. Highton and MacGregor made their distinction based on the results of an electrophoretic analysis of 22 presumptive genetic loci. Although they reported a great deal of geographic genetic variation among populations of \underline{P} . kentucki, there was a substantial amount of genetic variation between this species and other eastern large Plethodon, including P. glutinosus. Maha and Maxson (1983) utilized micro-complement fixation to report on immunological comparisons between albumins of the two species and confirmed that P. kentucki is distinct from P. glutinosus and all other species of the genus Plethodon.

More recent work conducted by Highton et al. (1989) has

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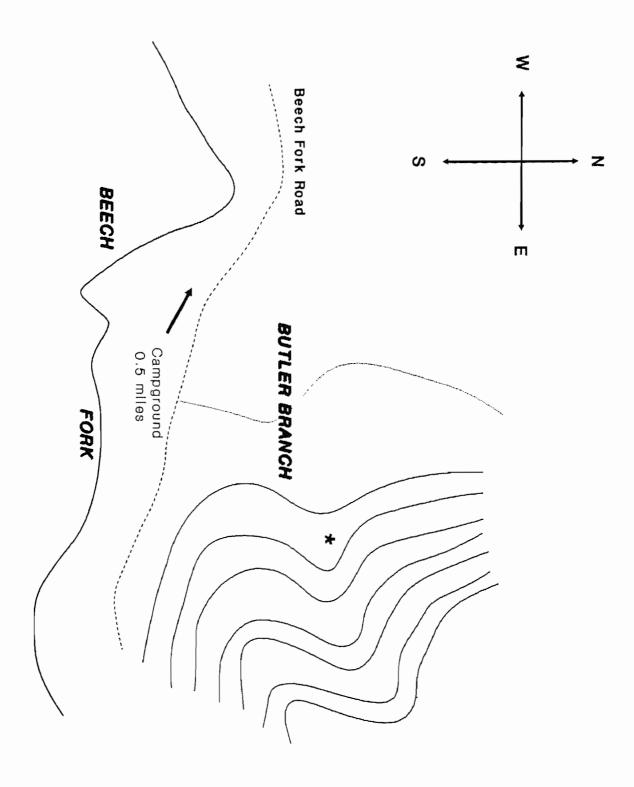
resulted in the placement of <u>P</u>. <u>kentucki</u> in what is known as the <u>P</u>. <u>glutinosus</u> complex. Through electrophoretic and immunological analyses of genetic variation within and among 135 populations of the complex, they suggested that it is comprised of 16 forms that have arrived at the species level of divergence. Among the 16 forms, <u>P</u>. <u>kentucki</u> differs most from <u>P</u>. <u>glutinosus</u>.

<u>Description of the Study Area</u> (Methods and Materials)

Field studies for <u>P</u>. <u>kentucki</u> were conducted at Beech Fork State Park (0.5 miles east on Beech Fork Road, and 0.3 miles north on Butler Branch Road) in Wayne County, West Virginia (Figure 6). This site was chosen because <u>P</u>. <u>kentucki</u> is the predominant species in the area, thus all ecological data collected would represent conditions most suitable for the success of this species.

The entire study site encompassed three aspects; westfacing, southwest-facing, and northwest-facing hillsides with ranges in elevation from 197 m to 247 m. The site was diverse in terms of topographical features and included steep-forested hillsides (inclination = 44°), narrow depressions, microreliefs, and exposed rock faces. An intermittent creek marked the lower boundary of the study area and created moist banks with loose rocky soil. The dominant vegetation of the site can be characterized as oak-beech.

Figure 6. An outline of the study area at Beech Fork State Park in Wayne County, West Virginia. Taken from U.S. Geological Survey topographic map, Winslow quadrangle (* indicates location of study site).



To obtain environmental data and study various ecological parameters, two line transects were constructed in the study area (Figure 7). A horizontal line transect 80 m long was positioned along a west-southwest hillside approximately 5-10 m above an intermittent creek. Four quadrats (25 m²) each 20 m apart were placed along the transect. Inclination varied from 32-38° at quadrats 1 and 2 to virtually no inclination 0-1° at quadrats 3 and 4. Quadrat 3 was located on a depression approximately 20 m x 60 m, while quadrat 4 approached a flat on the small floodplain of the intermittent creek. Mean elevation along the horizontal transect was 205 m, ranging from 197 m at quad 1 to 212 m at quad A vertical line transect 75 m long was positioned on the same 4. west-southwest hillside so that it bisected the horizontal line transect near quadrat 3. Three 25 m² quadrats (#'s 5,6,7) were placed along the vertical transect. Quadrat 5 was constructed near the intermittent creek, approximately three meters below quadrat 3, and thus was the lowest quadrat on the vertical line with an elevation of 208 m. Quadrat 6 was located 20 m upslope from quadrat 5 with an elevation of 225 m and inclination of 42°. Quadrat 7 was located 40 m upslope from quadrat 6 and approached the summit of the hillside with an elevation of 247 m and inclination of 38°.

A final quadrat (1000 m²) was constructed on the same hillside but was independent of the line transects and quadrats 1 through 7 (Figure 8). It was designated quadrat 8 and was positioned to obtain three aspects; southwest-, west-, and

Figure 7. An outline of the seven quadrats at the study site, showing their locations on horizontal and vertical line transects.

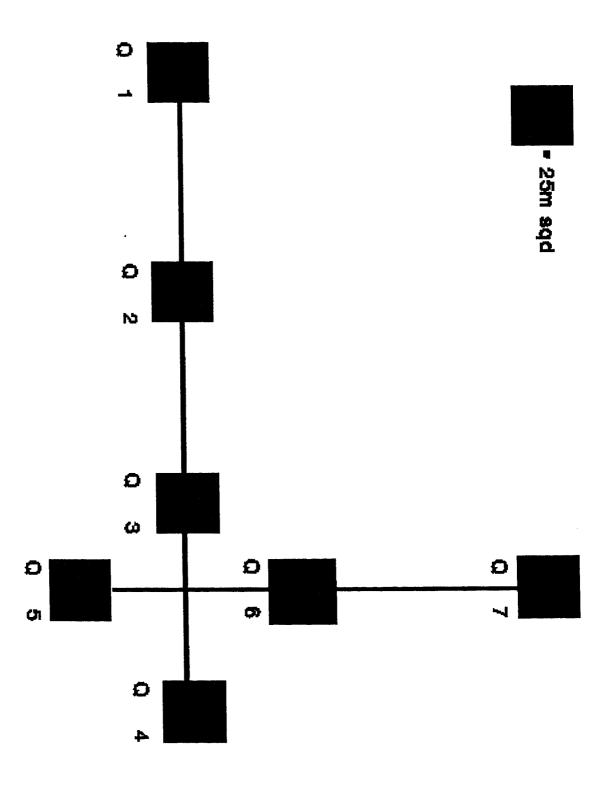
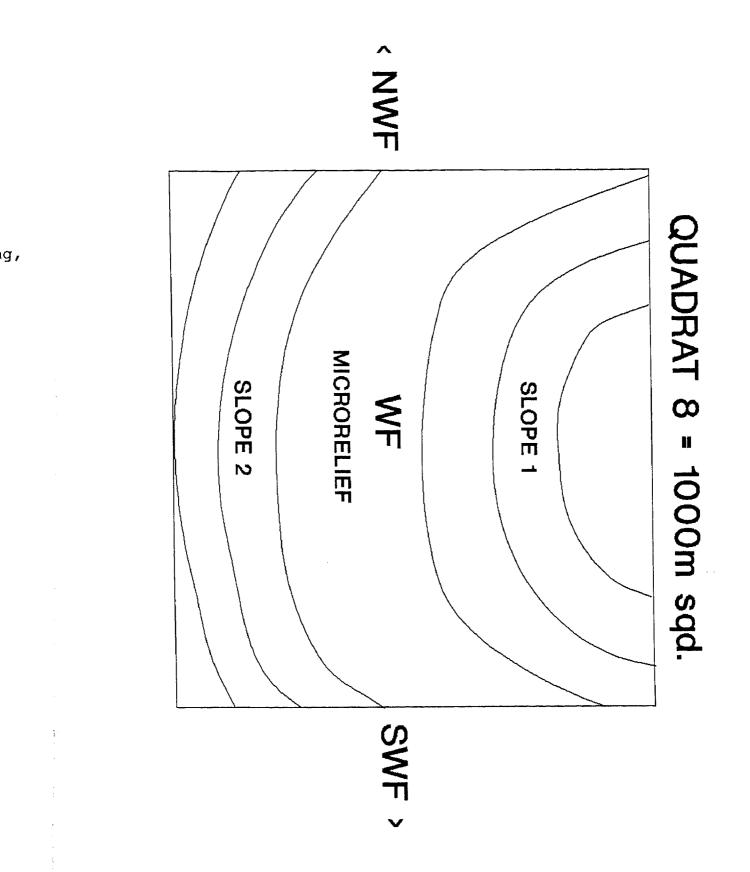


Figure 8. An outline of quadrat 8 (1000 m²) showing west-facing, northwest-facing, and southwest-facing slopes.



northwest-facing slopes. Topographical features of quadrat 8 were divided into three sections: a slope at the top with a 38° inclination, a microrelief near the midpoint of the quadrat with an inclination of 10°, and a second slope with an inclination of 44°. Elevation ranged from 246 m at the highest point of the quadrat to 200 m at the lowest point. The southwest side was part of a deep ravine ending in an intermittent creekbed. The west and northwest sides were bordered by a road that followed a wide ravine between adjacent hillsides.

CHAPTER I

Range and Distribution of <u>Plethodon kentucki</u> in West Virginia

Introduction

An important aspect concerning the ecology of a salamander species is spatial distribution. There are several factors (abiotic and biotic) involved in determining the distribution of salamanders. The abiotic component consists of environmental factors which include among others, temperature and moisture. In West Virginia, the Cheat Mountain salamander is restricted to the cooler and wetter Allegheny Mountains (Green and Pauley 1987). The biotic component includes interactions among and between species such as competition and predation. Jaeger (1972) concluded that Plethodon cinereus and Plethodon shenandoah maintain parapatric distributions in Virginia by competing for a limited supply of food, with P. cinereus excluding its congener from habitats preferred by both species. Both components are important, and in combination probably function in determining the spatial distribution of most salamander species.

The distributional patterns of salamanders of the genus <u>Plethodon</u> have received much attention. Grobman (1944) gave a detailed account on the distribution of the salamander genus <u>Plethodon</u> in eastern United States and Canada. He discussed the

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syngeographic distribution of the genus and divided it into two groups: eastern large <u>Plethodon</u> (<u>glutinosus</u>, <u>jordani</u>, <u>yonahlossee</u>) and eastern small <u>Plethodon</u> (<u>cinereus</u>, <u>richmondi</u>, hoffmani, <u>nettingi</u>).

Highton (1972) reported on the distributional interactions among eastern North American salamanders of the genus and concluded that closely related species seem to exclude one another both ecologically and geographically more frequently than do species which are distantly related.

Much work has been done concerning the distribution of salamanders in West Virginia. Green (1948) gave an annotated list of the amphibians and reptiles known to occur in the state, including 25 species of salamanders. Green and Pauley (1987) reported on the amphibians and reptiles in West Virginia which included the known range and distribution of 29 species of salamanders. Although their species accounts included the Cumberland Plateau salamander, Plethodon kentucki, very little was known at the time concerning its range and distribution throughout the state. In fact, it was known to occur in only three counties, Cabell and Wayne in the west, and Summers in the southeast (Highton and MacGregor 1983). Because of the limited knowledge regarding the range and distribution of P. kentucki in West Virginia, a detailed investigation was warranted. In addition, a study of the range and distribution of \underline{P} . glutinosus in West Virginia was conducted to aid in understanding the distributional interactions between these two species.

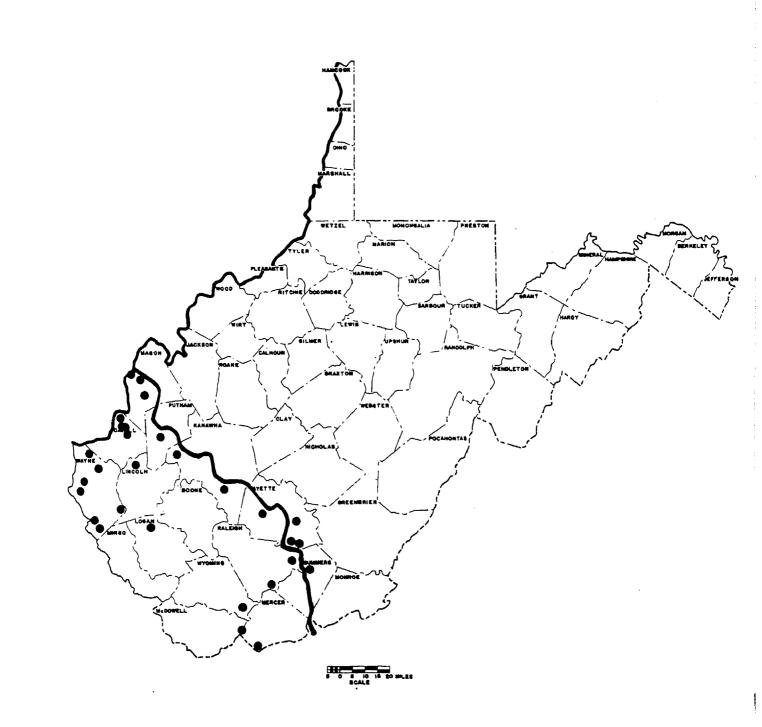
Methods and Materials

Field surveys for <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> were conducted throughout West Virginia. A likely habitat was chosen and typical searching techniques were employed. Daytime surveys involved turning all rocks and logs in the area, as well as examining other suitable microhabitats such as rock crevices. Night surveys were conducted with the aid of a flashlight and involved careful observation of the forest floor, particularly at the bases of large trees and rocks. The location of each specimen found was recorded and later plotted on a map of the state. Additional locations were obtained from personal records of Dr. Richard Highton of the University of Maryland, and by examining specimens from the West Virginia Biological Survey at Marshall University.

Results

Field surveys and museum records revealed that <u>P</u>. <u>kentucki</u> is restricted to the southwestern portion of the state (Figure 9). Its range extends from Mason County in the north to Mercer County in the south. Populations were found in 12 counties southwest of the New-Kanawha River system, and in two counties east of the New River. With the exception of Boone and Mingo, all counties southwest of the New-Kanawha River system were found to have populations of <u>P</u>. <u>kentucki</u>. Populations east of the New

Figure 9. The known range and distribution of <u>Plethodon kentucki</u> in West Virginia. Some records for Cabell, Wayne, and Summers counties are taken from Highton and MacGregor (1983).



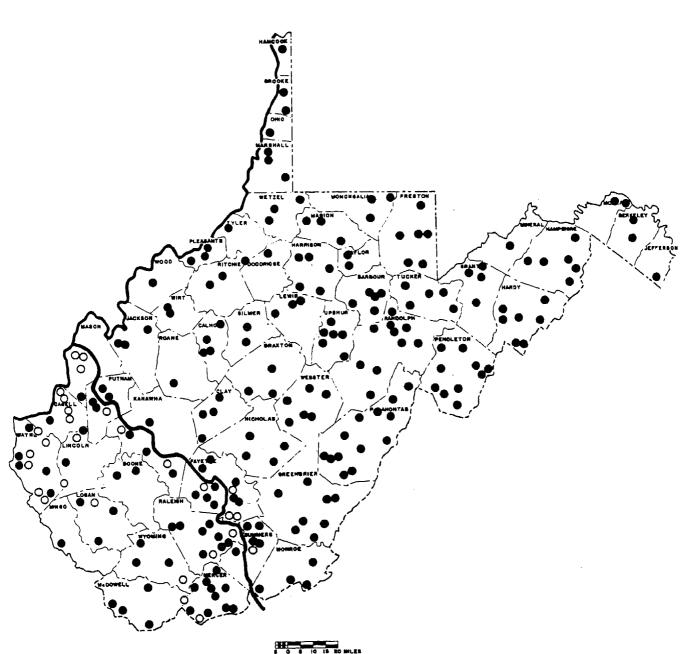
i d River were found in Fayette and Summers counties.

Research on the range and distribution of <u>P</u>. <u>glutinosus</u> revealed that it occurs in every county throughout West Virginia, and thus is sympatric with <u>P</u>. <u>kentucki</u> in 13 counties (Figure 10).

Discussion

This study revealed that <u>P</u>. <u>kentucki</u> is apparently limited to counties southwest of the New-Kanawha River system in West Virginia. With the exception of Fayette and Summers counties, <u>P</u>. <u>kentucki</u> were not found east of this river system. Several important ecological questions arise from this apparent limited distribution: (1) Does elevation play a role in limiting the range and distribution of <u>P</u>. <u>kentucki</u> in West Virginia? (2) Does the New-Kanawha River system act as a natural barrier to <u>P</u>. <u>kentucki</u>? (3) Do environmental factors (air temperature, soil temperature, air relative humidity, etc.) differ in northern counties to the degree that they effectively limit the distribution of <u>P</u>. <u>kentucki</u> to southwestern West Virginia? (4) Does competition with high density populations of <u>P</u>. <u>glutinosus</u> in northern counties play a role in limiting the distribution of **P**. kentucki?

Discrete answers to questions 1 and 2 are beyond the scope of this study; however, a discussion of each should be included to aid in understanding every possible aspect of the range and Figure 10. Geographic distribution of <u>Plethodon glutinosus</u> (closed circles) in West Virginia. Records of <u>P. kentucki</u> (open circles) are included to show counties where the two species are sympatric. Some records for <u>P. glutinosus</u> and <u>P. kentucki</u> are from personal records of Highton (unpublished data), and Highton and MacGregor (1983), respectively.



S IO IS RO HLES distribution of <u>P</u>. <u>kentucki</u> in West Virginia. For detailed answers to questions 3 and 4, refer to chapters II and III, respectively.

Elevational Differences

Elevation plays an important role in the distribution of many salamander species. In West Virginia, the Cheat Mountain salamander (Plethodon nettingi) is endemic to the Allegheny Mountain province, and limited to elevations exceeding 880 meters (USFWS 1991). The white-spotted salamander (Plethodon punctatus) is known from only two mountains along the West Virginia / Virginia border, the higher elevations of Shenandoah Mountains over 914 meters, and North Mountain over 853 meters (Highton 1972). Many salamanders species which are not typically restricted to higher elevations will often exhibit increased population densities as elevation increases. The blue ridge twolined salamander, Eurycea bislineata wilderae, occurs at elevations as low as 300 meters, but is most abundant above 600 meters and to the tops of tallest mountains (Conant 1975). Pauley (1980) reported that although Plethodon cinereus, the redback salamander, occurs at low elevations in West Virginia, it increases somewhat in percentage of occurrence with an increase in elevation. Pauley (1980) suggested that changes in occurrence along an elevational gradient are the result of either an influence from an environmental factor(s), or competitive interactions among salamander species of similar size, or both.

In his study of higher elevations in West Virginia (above 975 m), Pauley found that <u>Plethodon glutinosus</u> is significantly more abundant at lower elevation ranges than is <u>Plethodon wehrlei</u>. However, with an increase in elevation <u>P</u>. <u>wehrlei</u> replaces <u>P</u>. <u>glutinosus</u> and comprises a greater percent of the total number of salamanders.

Two important environmental factors associated with high elevations are temperature and moisture. Temperature typically decreases as elevation increases, while moisture increases with elevation (Smith 1980). <u>P. kentucki</u>, like all members of the genus <u>Plethodon</u>, is a lungless salamander and thus depends upon cutaneous respiration for gaseous exchange. Respiratory processes would then be most efficient with a cool, moist integument. Consequently, an area that has cool temperatures and a high moisture (i.e., high elevations) level would offer the conditions most suitable for the colonization of new areas by lungless salamanders such as <u>P. kentucki</u>.

The mean elevation for counties forming the boundary line (i.e., Kanawha-New River system) for the extent of <u>P</u>. <u>kentucki</u> range are given in Table 2 (WV Geological and Economic Survey 1986). Elevation increases dramatically from Mason County (mean = 191.5 m) upstream through Putnam, Kanawha, Fayette, and Raleigh counties with Summers County having the highest mean elevation (mean = 747.3 m). Although <u>P</u>. <u>kentucki</u> was found in each of the counties following the New-Kanawha River system, only Fayette and Summers counties had populations established east of this river

Table 2. Mean elevation for counties following the New-Kanawha River system.								
County	Mason	Putnam	Kanawha	Fayette	Raleigh	Summers		
Elevation (meters)	191.5	256.0	365.0	560.8	731.3	747.3		

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system. If higher elevations were conducive to more suitable habitats for P. kentucki, then populations would most likely colonize Fayette and Summers counties before the lower elevation counties, a situation which is supported by the results of this study. When discussing this situation, however, it must be kept in mind that forces other than elevational differences may be operating to limit the range of P. kentucki, such as the natural barrier effect imposed by the rivers, and the prospect of competition with high density populations of P. glutinosus on the opposite side of the rivers. Although environmental data for each county are not available for comparison, it appears feasible to postulate that the favorable conditions offered by higher elevations as in Fayette and Summers counties significantly increase the chances of survival for those presumably few individuals of P. kentucki which successfully cross the New River. An intense study of environmental factors such as air and soil temperature, air relative humidity, and soil moisture along the boundary of P. kentucki range is needed for further evaluation of this problem.

Rivers as Natural Barriers

It is generally accepted that bodies of fresh water such as rivers do not normally act as barriers to the dispersal of <u>Plethodon</u>. Highton (1972) remarked that most species which occur on either bank of most major rivers such as the Mississippi, Ohio, Columbia, and Hudson, also occur on the opposite side. In

West Virginia, several species of the genus <u>Plethodon</u> occur on both sides of the New-Kanawha River system (Green and Pauley 1987). <u>Plethodon cinereus</u> and <u>P. glutinosus</u> have extensive ranges throughout eastern United States and can be found on both sides of the New-Kanawha River system in West Virginia. Although its total range is less extensive, <u>P. wehrlei</u> also occurs on both sides of this river system in West Virginia. This evidence suggests that large rivers seldom act as barriers to most species of the genus <u>Plethodon</u>. Although <u>P. kentucki</u> occurs east of the New-Kanawha River system in Fayette and Summers counties, its absence east of the rivers in other counties which follow the system is of particular interest.

The New River enters southern West Virginia via Monroe and Summers counties and flows north where it meets with the Gauley River in Fayette County. The joining of these two rivers forms the Kanawha River which flows in a northwesterly direction through the state to Mason County where it joins the Ohio River. If this river system were a barrier to <u>P</u>. <u>kentucki</u>, it would most likely exert its effects along the Kanawha River in Mason, Putnam, and Kanawha counties. This is hypothesized because the Kanawha River is a higher order stream and would be wider throughout its length than the New River. Results of this study support this hypothesis since no <u>P</u>. <u>kentucki</u> were found east of the Kanawha River in these counties. As with the elevational differences discussed above, it is important to consider the possibility that several factors may be involved (i.e.

competition, environmental factors) in limiting the range and distribution of P. kentucki. Highton (1972) discussed a similar situation concerning distributional interactions between P. hoffmani and P. richmondi. The known ranges of these two species come closest in the New River Valley in Giles County, Virginia, and in Summers, Mercer, and Monroe counties, West Virginia. In this area, Highton found only P. hoffmani east of the river and only P. richmondi west of the river. He suggested that the New River, in combination with the presence of a closely related form on the opposite side of the river, effectively prevents the species from crossing. This may be representative of the situation occurring with P. kentucki. The absence of P. kentucki east of the Kanawha River in Mason, Putnam, and Kanawha counties may be the result of the combined effects of the river acting as a barrier, and competition with P. glutinosus.

Summary

Field surveys and museum records revealed that <u>P. kentucki</u> is restricted to southwestern counties in West Virginia. It was also determined that <u>P. glutinosus</u> occurs throughout West Virginia, and is sympatric with <u>P. kentucki</u> in 13 counties. With the exception of Fayette and Summers counties, <u>P. kentucki</u> were not found east of the New-Kanawha River system. Thus, this river system appears to partially mark the boundary line for the range of this salamander in West Virginia. Although it was not determined what mechanism(s) limit the range and distribution of <u>P</u>. <u>kentucki</u> in West Virginia, two hypotheses were discussed. Since elevation is important regarding the distribution of many salamander species, it was hypothesized that the higher mean elevation for Fayette and Summers counties provided the environmental conditions most suitable for the colonization of new areas by <u>P</u>. <u>kentucki</u>. Contrastingly, those counties with lower mean elevations (Mason, Putnam, Kanawha) would theoretically offer conditions less suitable (higher temperatures-less moisture) for the establishment of new populations of P. kentucki.

Because the New-Kanawha River system marks the boundary line for the extent of <u>P</u>. <u>kentucki</u> range in all counties except Fayette and Summers, it was hypothesized that the Kanawha River, which is typically wider, is acting as a natural barrier and preventing the establishment of new populations on the opposite side. It was further suggested that the barrier effects of the rivers may not be working alone, and in fact may be operating in combination with competition involving <u>P</u>. <u>glutinosus</u> to limit the range and distribution of <u>P</u>. <u>kentucki</u> (see chapter 3).

It must be noted that several factors are often involved in determining the spatial distribution of salamanders. Abiotic factors such as soil and air temperature, air relative humidity, soil moisture, and soil pH, are important components of the microclimate of salamanders, and thus influence their distribution. Biotic factors such as competition, predation, and

plant species component are important and should be considered when the distribution of a salamander species is in question.

CHAPTER II

Aspects of the Natural History of <u>Plethodon kentucki</u> in West Virginia

Introduction

Although <u>Plethodon kentucki</u> is relatively common throughout its range, information concerning its natural history and ecology is limited. Existing studies have focused on validating its distinctness as a true species by means of morphometric measurements, immunological studies, and or electrophoretic analyses (Clay et al. 1955; Maha and Maxson 1983; Highton and MacGregor 1983; Highton et al. 1989). These studies are of great importance to the species, however they contribute little information to its natural history and ecology.

Behavioral interactions among and between <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> including aggressive behavior, chemical communication, intra- and interspecific interference competition have been studied in Kentucky (Delpont 1987; Marvin 1990). These studies are limited due to the artificiality of the laboratory setting; however, they are generally accepted and contribute to the understanding of competitive interactions between species even in natural settings.

Although information regarding aspects of the ecology of \underline{P} . <u>kentucki</u> is limited, \underline{P} . <u>glutinosus</u> and several other members of the genus <u>Plethodon</u> have been studied intensely. Semilitsch (1980) studied populations of <u>P</u>. <u>glutinosus</u> in Maryland and Pennsylvania to obtain information on population structure and density. A study in North Carolina involving <u>P</u>. jordani was conducted by Hairston (1983) in which reproduction, survival, and growth were investigated. Pauley (1977) studied habitat partitioning between <u>P</u>. <u>cinereus</u> and <u>P</u>. <u>wehrlei</u> in West Virginia. Jewell (1991) evaluated the factors (environmental and or autecological) which regulate the distribution of <u>P</u>. <u>richmondi</u> and P. cinereus in West Virginia.

The purpose of this study was to provide insights into the natural history of <u>P</u>. <u>kentucki</u>. Topics of study included: (1) surface density (2) microclimatic conditions (3) seasonal and diel activities (4) movement patterns (5) cover object preference (6) aspect studies (7) vegetation analysis at study site (8) observations of sympatric amphibians and reptiles at study site.

Methods and Materials

Surface Density / Seasonal and Diel Activities

In order to determine what time of the year <u>P</u>. <u>kentucki</u> was most active, field surveys were conducted and surface density was recorded for each month of the study. Surveys involved lifting all rocks and logs and examining other suitable habitats such as rock crevices. After locating a salamander, soil and air temperature readings were taken at the position each specimen was

occupying. These on-site microclimatic data were plotted against monthly salamander abundance in order to elucidate any existing relationships. Scheduled surveys of the quadrats were made between 12:00 PM and 4:00 PM EST with two-week intervals, at which time microclimatic data were collected. All salamanders found in the quadrats were marked using a method of toe-clipping as described in Ferner (1979). Schnabel method T mark and recapture techniques were employed to provide an estimate of population size. Unscheduled surveys were conducted as a result of rainy and wet weather conditions. During these surveys (usually at night), quadrats and areas outside were searched.

The snout-vent length and sex of salamanders found during surveys were recorded to determine if adults, juveniles, and sexes were active at different times of the year. A vernier caliper was used to measure SVL (snout to anterior angle of cloaca) to the nearest 0.1 mm, and gender was determined by examining cloacal glands.

In order to determine diel activity of <u>P</u>. <u>kentucki</u>, the time of day was recorded for every salamander found on the surface, i.e., any salamander which was visible without removing cover objects. Most surveys were conducted on rainy and or wet nights during the hours of 6:00 PM through 12:00 AM EST.

Microclimatic Conditions

In order to determine the conditions which define the \underline{P} . <u>kentucki</u> habitat, microclimatic data were recorded at the study

site every two weeks during the period of June 1990 to August Relative humidity and air temperature (°C) readings were 1991. obtained using a Bacharach Sling Psychrometer (model # 12-70130). Five readings were taken (approximately 5 cm above soil surface) in each of the seven 25 m² quadrats. The procedure involved recording a separate reading in each of the four corners, and one in the center of the quadrat. A Weston (model # 2282) soil thermometer was used to obtain five soil temperature (°C) readings (approximately 5 cm below soil surface) in each quadrat. Two soil samples were collected in each quadrat and returned to the laboratory for analyses of moisture content and pH. Soil moisture was obtained by removing two 20 g subsamples of soil from each quadrat sample and placing them in a petri dish for drying. Care was taken to remove small stones and any other debris. After drying, soil subsamples were weighed using an Ohaus Digital Scale (model # E120) and percent (%) moisture was calculated from the differing weights. Soil pH was determined by combining 10 g of soil from each quadrat sample with 100 ml of distilled water of neutral pH. The pH of the mixture was recorded using a Redox pH Meter (model # 707mV).

All data collected in the quadrats were obtained between the hours of 12:00 PM and 4:00 PM EST. Monthly averages for each microclimatic condition were obtained by combining readings of the two days the study site was visited. In order to determine the relationship between each microclimatic condition and salamander density, averages were plotted against the number of

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<u>P</u>. <u>kentucki</u> observed each month. Air and soil temperature readings obtained in the quadrats were compared to on-site air and soil temperature readings. On-site readings are those recorded at the exact position of each salamander during surveys.

Laboratory studies were conducted to determine the relationship between P. kentucki and environmental temperature and moisture. Critical Thermal Maxima (CTM) studies were performed to determine the maximum temperature (°C) P. kentucki can withstand before disorientation. Twenty-six adult P. kentucki (16 males, 10 females) were acclimated to room temperature (21.0 °C) for 48 hours and then individually placed into a perforated plastic container. The container and specimen were submerged into a water bath at room temperature (21.0 °C). A mercury thermometer graduated to 0.1 °C was inserted through a hole in the container to obtain water temperature readings. In order to prevent thermal shock to the salamander, the CTM chamber was adjusted to increase the water temperature 1 °C every 3 minutes (Brooks and Sassman 1965). Disorientation and the inability of a salamander to right itself were indicative of the CTM temperature value for each specimen (Hutchinson 1961). Twenty adult P. glutinosus (8 males, 12 females) were also tested for comparison.

Dehydration studies were conducted to determine the percentage of body water <u>P</u>. <u>kentucki</u> can lose before death. Eleven adult <u>P</u>. <u>kentucki</u> were individually placed into a wire cage of known weight. The cage with specimen inside was placed

into a cylinder containing 200 mg of anhydrous calcium chloride, which acted as a dehydrant. An air pump was connected to the cylinder via a rubber hose and thus pumped a steady flow of air across the salamander. An outlet hose was connected to the opposite end of the cylinder and lead to a flask of water. Air bubbles in the flask indicated that air was passing through the cylinder. Salamanders were weighed every 30 minutes until death. All tests were performed at a constant temperature of 21.0 °C. Dehydration values obtained for <u>P</u>. <u>kentucki</u> were compared to values obtained for <u>P</u>. <u>glutinosus</u> in West Virginia (Pauley unpublished data).

Movement Patterns

Progressive linear movements of <u>P</u>. <u>kentucki</u> were monitored by utilizing recaptured marked salamanders from population studies. The position of capture for each toe-clipped salamander was marked by inserting a stake endowed with florescent flagging into the ground. If salamanders were captured in unique microhabitats such as rock crevices, their position was marked with a small application of florescent paint. The location of recaptured salamanders was noted and the distance moved from the original point of capture was measured to the nearest 0.01 m.

Cover Preference

Cover object preference for <u>P</u>. <u>kentucki</u> was determined by recording the type of cover each salamander was associated with

during field surveys. Cover was divided into three categories: rocks, logs, and leaves.

Aspect Studies

To determine the relationship between surface density of \underline{P} . <u>kentucki</u> and aspect, a 1000 m² quadrat (#8) was constructed at the study site. It was positioned to obtain three aspects, southwest, west, and northwest. Survey techniques followed the procedures as described above and were conducted biweekly during scheduled visits. Unscheduled visits were conducted as result of wet weather.

Vegetation Analysis

A vegetation analysis was performed in order to determine the plant species component of the study area. The analysis involved dividing the plant community into three strata, canopy, understory, and shrubs. All identifications and counts were made using a modified version of the line intercept method for sampling plant communities as described in Smith (1980). Twentymeter intervals along the horizontal line transect (80 m) and vertical line transect (75 m) at the study site were utilized for the identifications and counts.

Sympatric Amphibians and Reptiles

Observations of amphibians and reptiles living sympatrically with <u>P. kentucki</u> at the study site were recorded.

Most observations were inadvertent and made during surveys for \underline{P} . kentucki.

Results

Surface Density / Seasonal and Diel Activities

Seventy-four P. kentucki (34 females, 26 males, 14 juveniles) were observed at the study site during the 15 month study period. A summary of the number of females, males, and juveniles observed each month is presented in Table 3. The study began in June 1990, and during this month 3 females and 4 males were observed. The number of P. kentucki observed thereafter decreased dramatically, with only 4 individuals (4 juveniles) representing the months of July 1990 through December 1990. A juvenile with a snout-vent-length of 32.0 mm was the first to appear (January 31) during 1991. Surface density increased during February as 3 adult females, 1 adult male, and 1 juvenile were found. A dramatic increase occurred during March and April of 1991 in which approximately 70 percent of the observations were made. This increase was met with a sudden drop during May with only 3 adult females and 2 adult males constituting the month's total. Surface density was low during the final three months (June, July, and August) of the study with just two additional P. kentucki (2 juveniles) found in August.

Mean air and soil temperatures (55 readings each) recorded at the position of each salamander observed were plotted against

Table 3. Monthly surface abundance of <u>Plethodon kentucki</u> at Beech Fork State Park in Wayne County, West Virginia.							
MONTH	# FEMALES	# MALES	# JUVENILES	Total			
June '90	3	4	0	7			
July '90	0	0	1	1			
Aug. '90	0	0	1	1			
Sept. '90	0	0	2	2			
Oct. '90	0	0	0	0			
Nov. '90	0	0	0	0			
Dec. '90	0	0	0	0			
Jan. '91	0	0	1	1			
Feb. '91	3	1	1	5			
Mar. '91	11	9	4	24			
Apr. '91	1.4	10	2	26			
May '91	3	2	0	5			
June '91	0	0	0	0			
July '91	0	0	0	0			
Aug. '91	0	0	2	2			
Total	34	26	13	74			

surface density for February, March, and April 1991 (Figure 11). A progressive increase in both air and soil temperature occurred from February to April. Surface density followed a similar pattern as the number of <u>P. kentucki</u> increased from five in February, to 24 and 26 for March and April, respectively.

Twenty-one <u>P</u>. <u>kentucki</u> were toe-clipped within the eight quadrats at the study site (Table 4). As a result of its larger size, approximately 71 percent of the salamanders were marked in quadrat 8. Three salamanders were marked in quadrat 7, two in quadrat 5, and one in quadrat 3. Four recaptures were recorded during the study. One juvenile (17.5 mm SVL) and one adult female (60.1 mm SVL) represented recaptures for quadrat 8, and one adult female (63.2 mm SVL) was recaptured twice in quadrat 5. Due to the low number of recaptures, an estimate of the population was not feasible.

There was no obvious difference in seasonal activity between sexes. Although the total number of juvenile observations for the study was low, they did appear to be more active during the summer and fall seasons when adults were virtually absent from the surface.

Results of diel activity studies revealed that <u>P</u>. <u>kentucki</u> is most active between the hours of 9:00 PM and 10:00 PM EST (Figure 12). Salamander activity began after 7:00 PM and gradually increased throughout the evening until it reached a climax between 9:00 PM and 10:00 PM (46.2% of total observations). Although activity was relatively high after

Figure 11. Mean on-site air and soil temperatures, and number of <u>Plethodon kentucki</u> observed for February, March, and April 1991. (Based on 55 temperature readings).

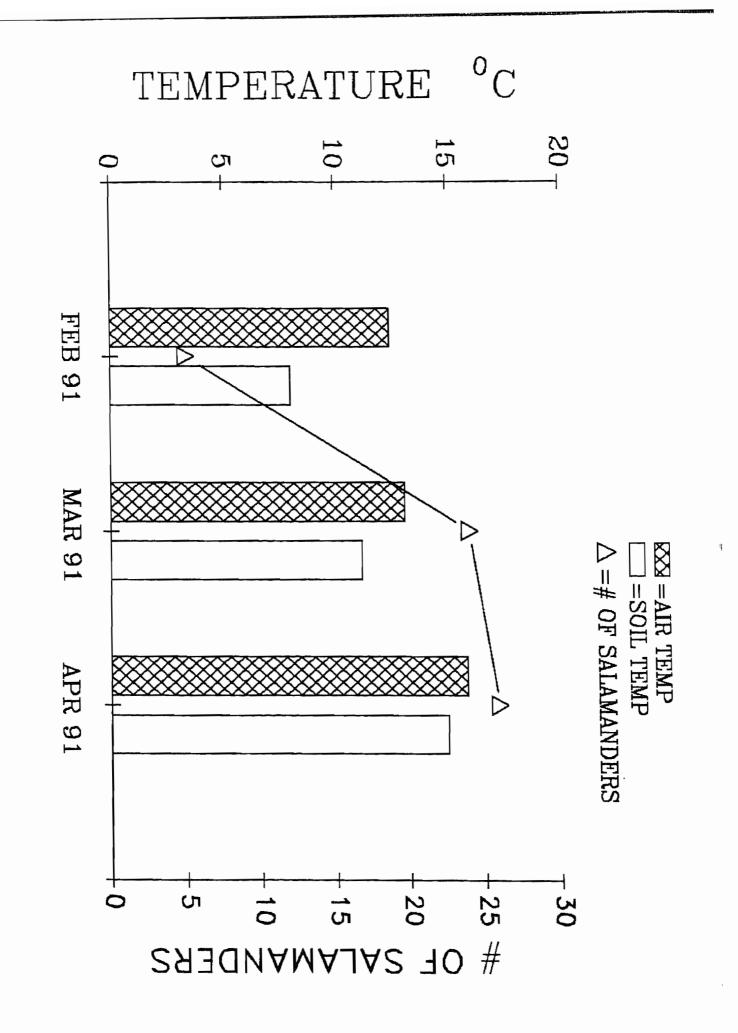
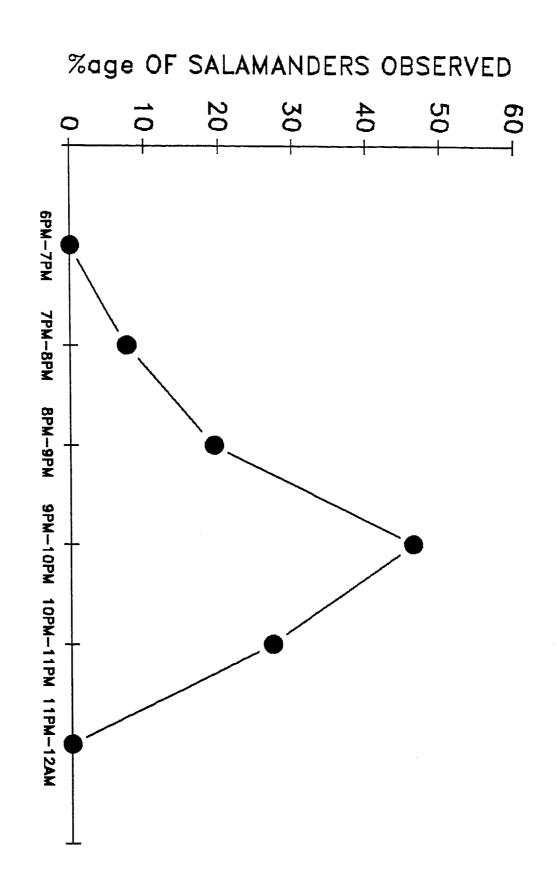


Table 4. Number of <u>Plethodon</u> <u>kentucki</u> marked in each quadrat at Beech Fork State Park.							
QUADRAT #	# FEMALES	# MALES	# JUVENILES	TOTAL			
1	0	0	0	0			
2	0	0	0	0			
3	11	0	0	1			
4	0	0	0	0			
5	1	1.	0	2			
6	0	0	0	0			
7	1	2	0	3			
8	8	6	1	15			
TOTAL	11	9	1	21			

Figure 12. Diel activity profile for <u>Plethodon</u> <u>kentucki</u> at Beech Fork State Park. Percentages based on 67 specimens.



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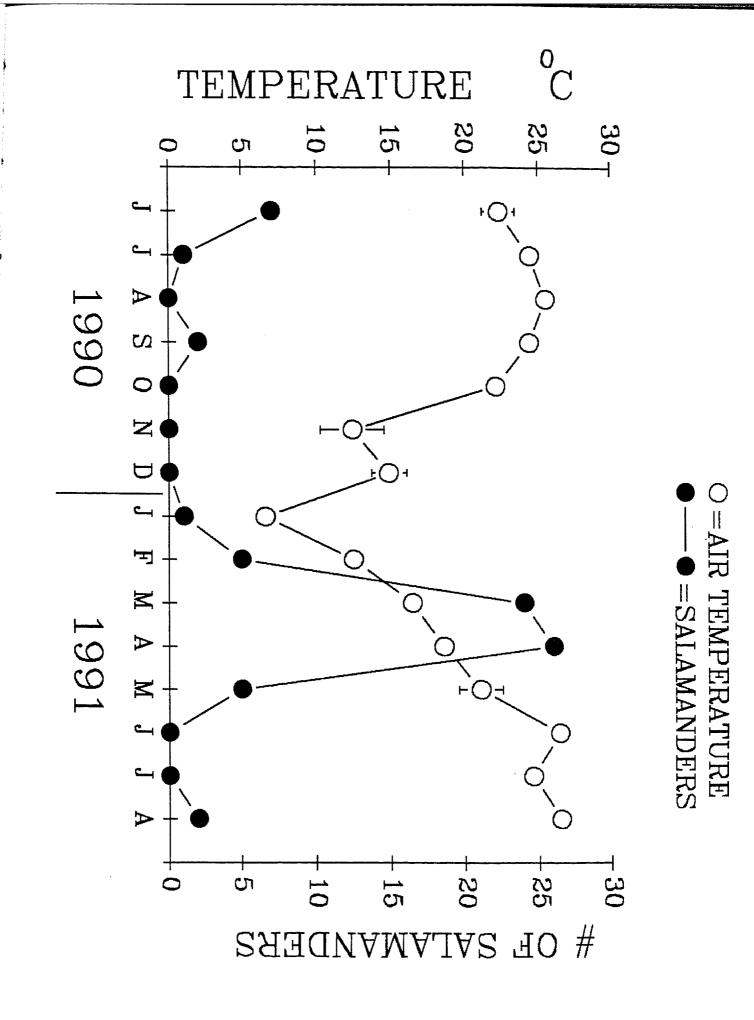
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10:00 PM, it dropped sharply after 11:00 PM and subsequently came to a halt.

Microclimatic Conditions

Mean air temperatures (°C) (± 1 S.E.) and numbers of salamanders observed for each month of the study are presented in Figure 13. Surface density was not significantly correlated with air temperature during the 15 month study: Spearman's rank correlation coefficient, two-sided; P > 0.025 (Ott and Mendenhall 1985). The study began in June of 1990 with a mean air temperature of 22.3 °C. Seven P. kentucki were observed during this month. Surface density decreased as air temperatures steadily increased during July (mean = 24.4 °C) and August (mean = 25.5 °C). Although there was a steady decline in air temperature during September, October, and November, only three P. kentucki (3 juveniles) were observed during this time period. A slight increase in air temperature occurred thereafter, as the mean air temperature for December was slightly higher than that of November. Although January 1991 was the coolest month of the study (mean = 6.6 °C), a juvenile with a snout-vent-length of 32.0 mm was observed. As air temperatures began to increase during February, March, and April, a dramatic increase in salamander density occurred. Fifty of the total 74 P. kentucki observed during the study were found during March and April of 1991, with mean air temperatures of 16.5 °C and 18.6 °C, respectively. As temperatures continued to increase during May

Figure 13. Relationship between mean air temperature °C (bars indicate ± 1 S.E.) and surface density of <u>Plethodon</u> <u>kentucki</u> for each month of the study. Surface density was not significantly correlated with air temperature: Spearman's rank correlation coefficient, two-sided; P > 0.025.



(mean = 21.0 °C) and June (mean = 26.4 °C), salamander abundance concomitantly declined as only five salamanders were found during the two months. The warmest month of the study was August 1991, which had a mean air temperature of 26.5 °C. Two juvenile <u>P</u>. <u>kentucki</u> were added to the total during this month.

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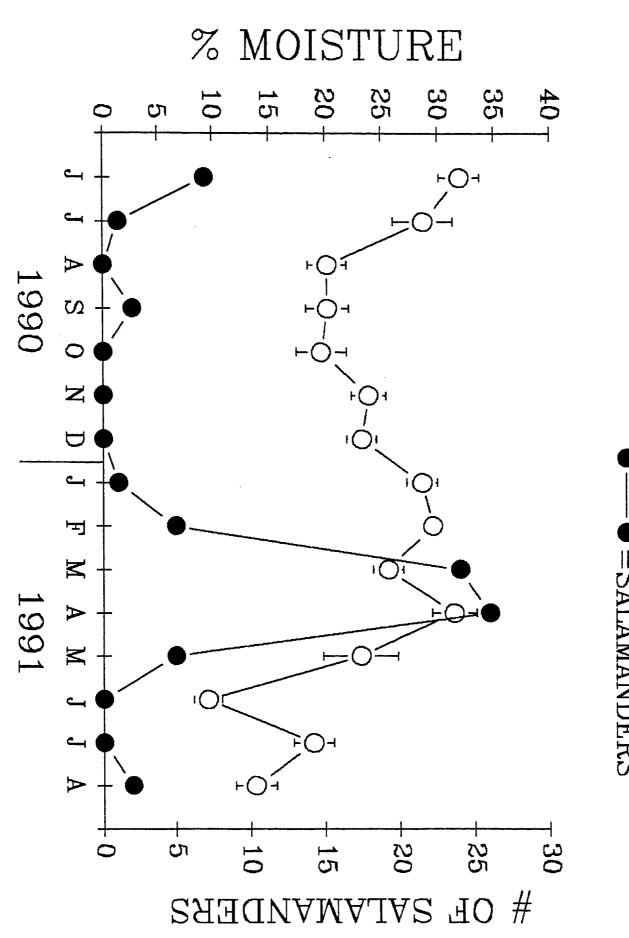
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Mean soil temperatures °C (± 1 S.E.) and numbers of salamanders observed for each month of the study are presented in Figure 14. Surface density of salamanders was not significantly correlated with soil temperatures: Spearman's rank correlation coefficient, two-sided; P > 0.025. Monthly soil temperatures followed a pattern similar to that of air temperatures, but with fewer fluctuations. Salamander density is the same for each microclimatic condition studied, thus monthly surface density for soil temperature is identical to that of air temperature. An increase in soil temperature occurred from June until July. This was followed by a decrease in mean monthly temperatures beginning with August and ending in February, which had the lowest mean soil temperature (3.4 °C) during the study. Soil temperatures increased thereafter with only one fluctuation occurring during July. Soil temperature reached a high during August 1991, with a mean of 23.7 °C. As was the case with air temperatures, 50 of the 74 P. kentucki observed during the study were found during March and April of 1991. Mean soil temperatures for March and April were 10.5 °C and 15.6 °C, respectively.

Mean air relative humidities (± 1 S.E.) and numbers of salamanders observed for each month of the study are presented in

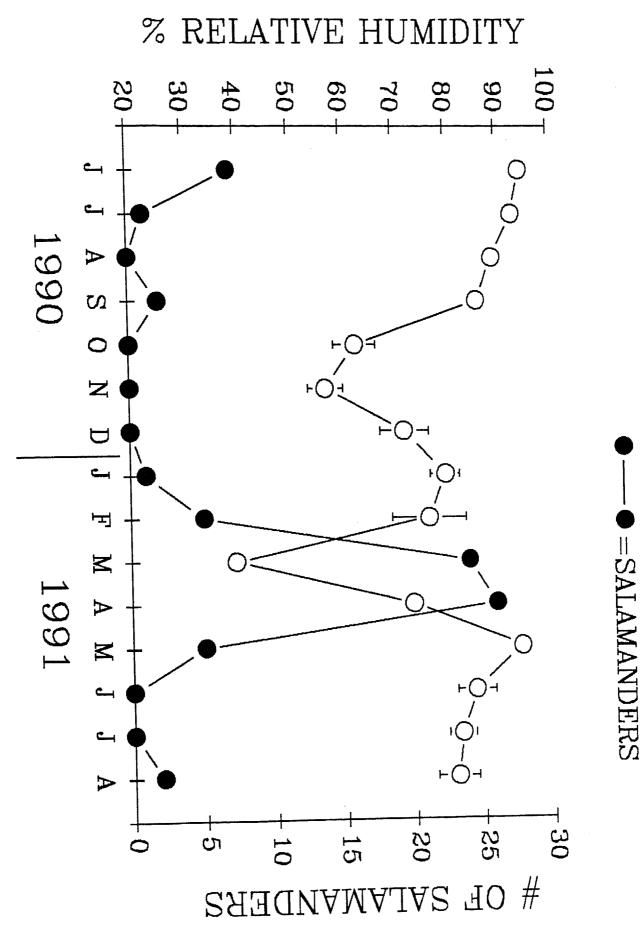
Figure 14. Relationship between mean soil temperature °C (bars indicate ± 1 S.E.) and surface density of <u>P. kentucki</u> for each month of the study. Surface density was not significantly correlated with soil temperature: Spearman's rank correlation coefficient, two-sided; P > 0.025.



O =% SOIL MOISTURE ● ---- ● =SALAMANDERS Figure 15. Surface density was not significantly correlated with air relative humidity: Spearman's rank correlation coefficient, two-sided; P > 0.025. Seven P. kentucki were observed during June 1990, which exhibited the highest mean air relative humidity for the study at 94.6 percent. A gradual decline in air relative humidity occurred after June and continued through November. This was followed by a similar decrease in surface density, with only four P. kentucki (4 juveniles) representing the months of July 1990 through November 1990. Rapid fluctuations in air relative humidity occurred thereafter, with December and January showing slight increases, and February and March (mean = 39.4 °C March) showing drastic decreases. This drastic decrease was met with a subsequent increase in surface density of P. kentucki during March and April (approximately 70% of total observations). A final increase in air relative humidity occurred during April and May (mean = 73.2% April; 94.2% May), which was followed by a gradual decline during June, July and August. A similar decline in surface density occurred as only 2 P. kentucki were added to the total during June, July, and August.

Mean soil moisture values (\pm 1 S.E.) and numbers of salamanders observed for each month of the study are presented in Figure 16. Surface density was positively and significantly correlated with soil moisture: Spearman's rank correlation coefficient, two-sided; P < 0.025. The highest soil moisture readings were obtained during June 1990 (mean = 31.99%). Seven <u>P. kentucki</u> were found during this month. A gradual decrease in

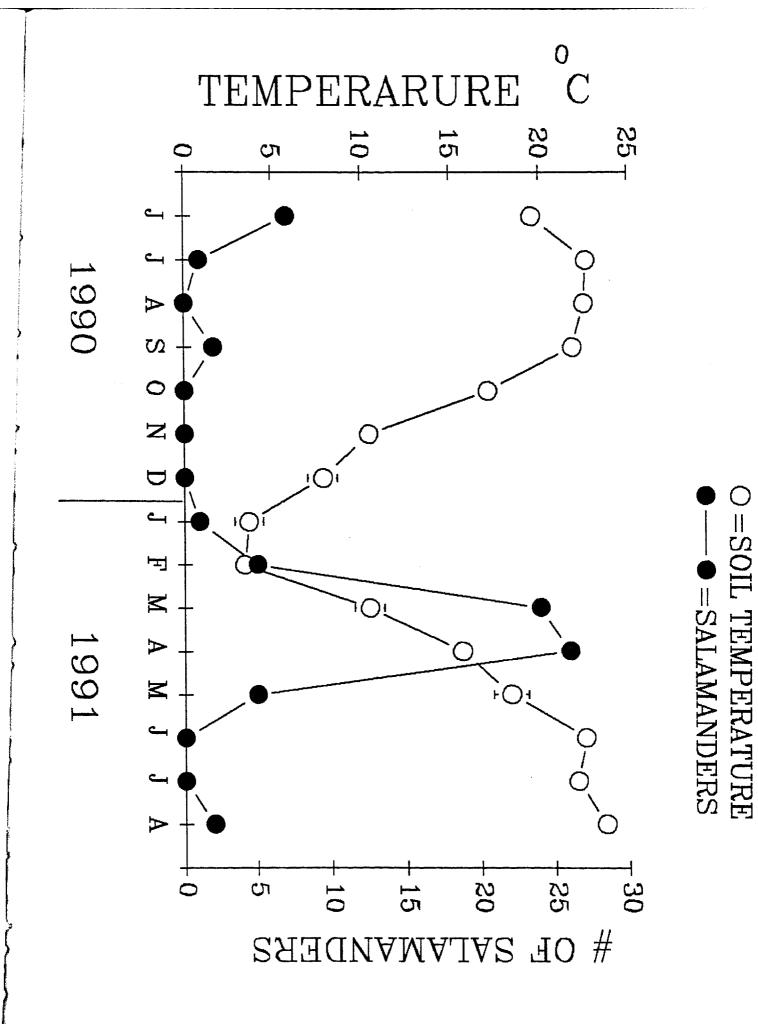
Figure 15. Relationship between mean % air relative humidity (bars indicate ± 1 S.E.) and surface density of <u>P. kentucki</u> for each month of the study. Surface density was not significantly correlated with air relative humidity: Spearman's rank correlation coefficient, two-sided; P > 0.025.



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Figure 16. Relationship between mean % soil moisture (bars indicate ± 1 S.E.) and surface density of <u>P. kentucki</u> for each month of the study. Surface density was significantly correlated with soil moisture: Spearman's rank correlation coefficient, two-sided; P < 0.025.

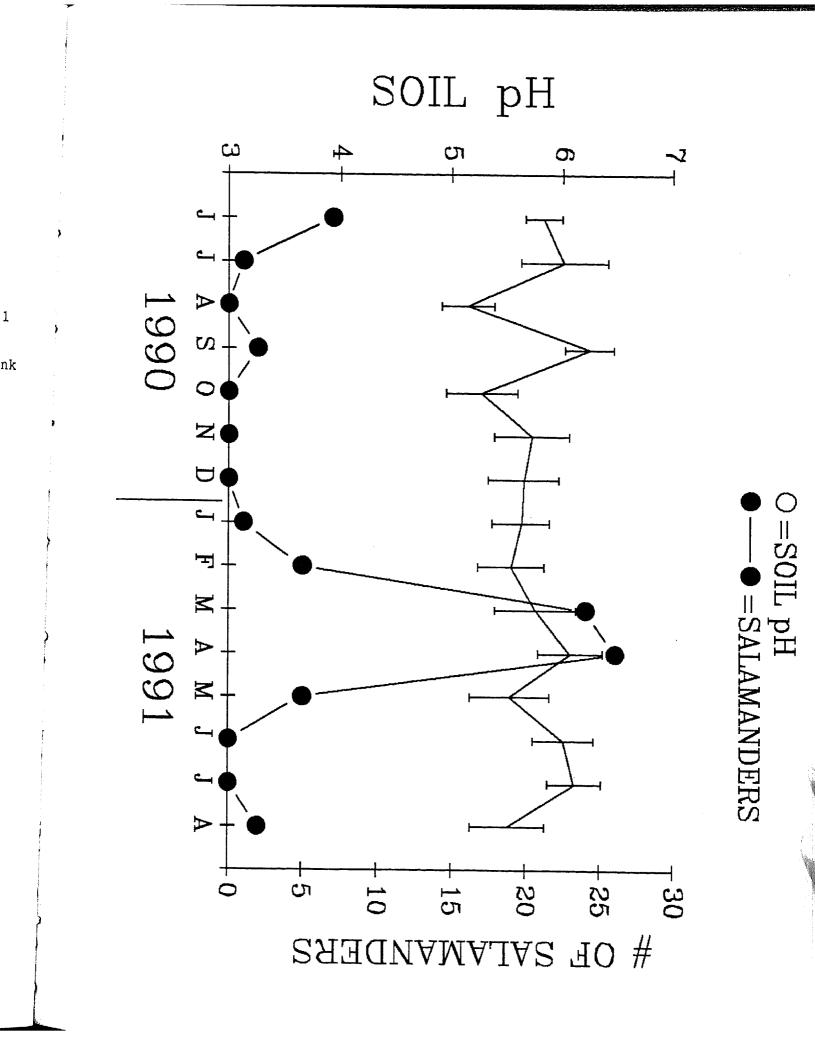


soil moisture was the trend from July to October 1990. Surface density exhibited a similar decrease as only 4 <u>P</u>. <u>kentucki</u> were observed during July, August, September, and October. A gradual increase in soil moisture was the trend from November 1990 through April 1991, with two slight decreases during December and March. Surface density was highest during March and April 1991 (approximately 70% of total observations) when soil moisture was relatively high (mean = 25.62% March; 31.48% April). Soil moisture declined thereafter and remained low during the final months (May through August 1991) of the study. Similarly, surface density of <u>P</u>. <u>kentucki</u> declined and remained relatively low, with seven individuals representing the months of May through August.

Mean soil pH values (\pm 1 S.E.) and numbers of salamanders observed for each month of the study are presented in Figure 17. Surface density was not significantly correlated with soil pH: Spearman's rank correlation coefficient, two-sided; P > 0.025. The month exhibiting the highest mean soil pH was September 1990 (mean = 6.24). Two juvenile <u>P. kentucki</u> were observed during this month. The month exhibiting the lowest mean soil pH was August 1990 (mean = 5.15), a month in which salamanders were not found. There were several fluctuations in soil pH throughout the 15 month study, however most values were within the interval of 5.5 - 6.0. Surface density was highest during March and April 1991, with mean soil pH values of 5.75 and 6.06, respectively.

Quadrat air and soil temperatures obtained during March and

Figure 17. Relationship between mean soil pH (bars indicate ± 1 S.E.) and surface density of <u>P. kentucki</u> for each month of the study. Surface density was not significantly correlated with soil pH: Spearman's rank correlation coefficient, two-sided; P > 0.025.



April when surface density was highest were compared with on-site (salamander's position) air and soil temperatures (Figure 18). Air temperatures were significantly different for both March and April (t-test, P < 0.05). Soil temperatures were not significantly different for March or April (t-test, P > 0.05).

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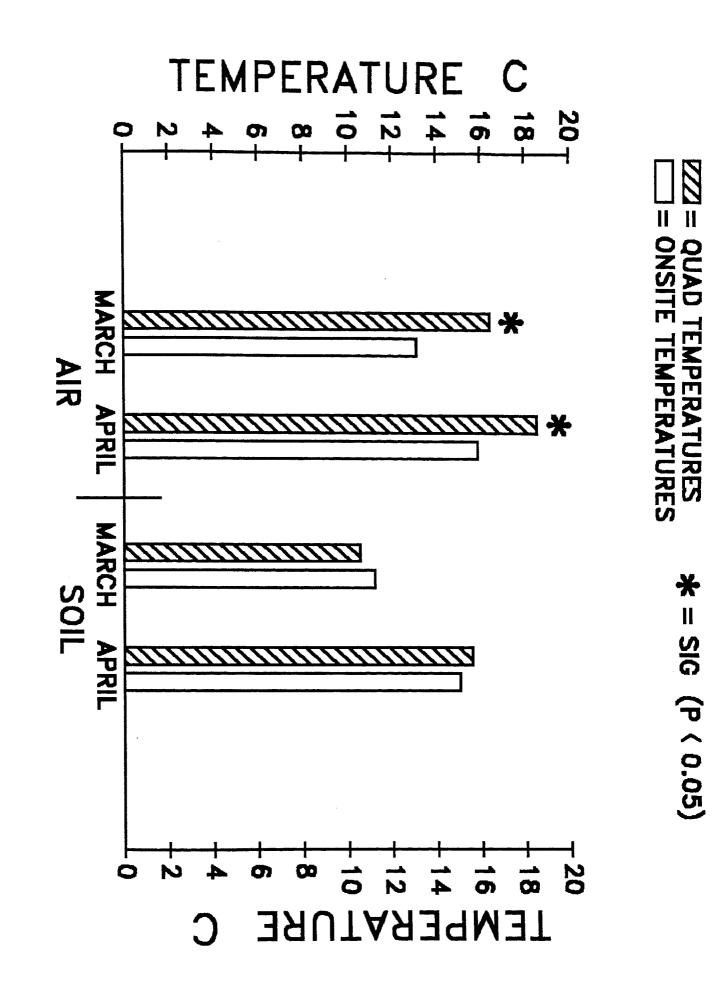
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Soil and air temperatures, and air relative humidity readings obtained at the study site in southwestern West Virginia were compared to readings obtained from a northwestern site (Jewell 1991). There were no significant differences in mean air temperatures, soil temperatures, and air relative humidities between Wayne County and Ritchie County for the months of June 1990 through March 1991 (Table 5). There was no significant difference in average annual rainfall (1947-1976) from areas near the two sites (Lee et al. 1977).

Mean CTM values (± 1 S.E.) for <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> are presented in Table 6. The mean CTM value for <u>P</u>. <u>kentucki</u> was 33.4 °C, while the CTM value for <u>P</u>. <u>glutinosus</u> was slightly higher at 33.6 °C. These values were not significantly different (t-test, P > 0.05).

Mean dehydration values (± 1 S.E.) for <u>P. kentucki</u> and <u>P. glutinosus</u> are presented in Table 7. The mean dehydration value for <u>P. kentucki</u> was 38.21 percent, while the mean dehydration value for <u>P. glutinosus</u> was slightly higher at 40.27 percent. These values were not significantly different (t-test, P > 0.05).

Figure 18. Comparison of on-site air and soil temperatures with quadrat temperatures for March and April 1991, when surface density of <u>P</u>. <u>kentucki</u> was highest. Air temperatures were significantly different (P > 0.05); soil temperatures were not significantly different (P < 0.05).



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Table 5. Mean air and soil temperatures °C, air RH %, and rainfall for northeastern and southeastern counties in West Virginia (values not sig. dif. P > 0.05).								
SITE	N	AI MEAN			IL SE	AIR MEAN		RAINFALL " MEAN
NE	372	14.6	2.9	11.7	2.4	72.3	4.2	43.7
SE	635	18.2	2.0	13.9	2.5	74.9	5.6	44.3
* N E data taken from Jewell (1991), at North Bend State Park, Ritchie County, West Virginia (from June 1990 - March 1991). * Rainfall is annual average for years 1947-1976 at areas near N E and S E sites (Lee et al. 1977).								

Table 6. Mean CTM values (± 1 S.E.) for <u>Plethodon</u> <u>kentucki</u> and <u>P. glutinosus</u> .				
SPECIES	N	MEAN	S. E.	
<u>P. kentucki</u>	26	33.4	0.092	
P. glutinosus	20	33.6	0.250	
Values not significantly different: t-test, P > 0.05.				

Table 7. Mean dehydration values (± 1 S.E.) for <u>P</u> . <u>kentucki</u> and <u>P. glutinosus</u> .				
SPECIES	N	MEAN	S. E.	
<u>P. kentucki</u>	11	38.2%	1.5	
P. glutinosus	12	40.3%	1.4	
Values not significantly different: t-test, P > 0.05.				

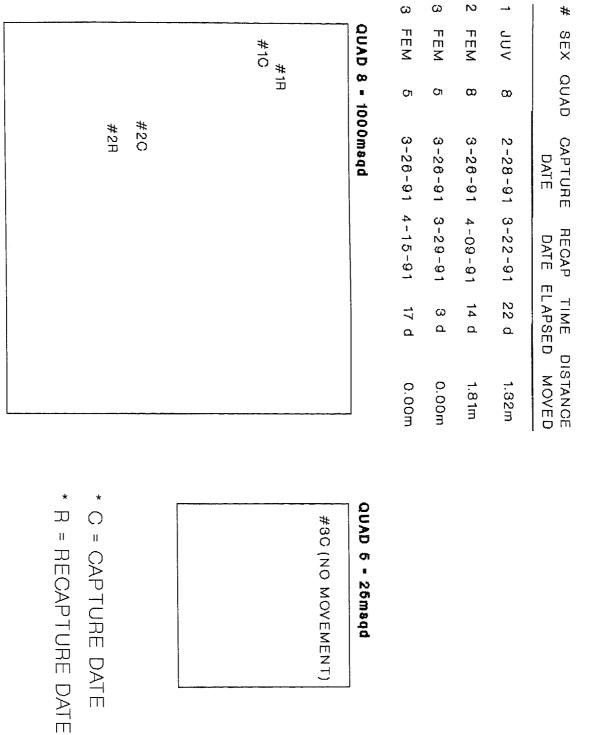
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Movement Patterns

The mark and recapture study for a population size estimate produced only four recaptured individuals, thus information on movement patterns is limited. Results obtained during the study are submitted in Figure 19. The first recapture was a juvenile (17.5 mm SVL) found March 22, 1991. The time elapsed between mark and recapture dates was 22 days, and the distance moved was measured at 1.32 m. An adult female was recaptured on March 29, and once again on April 15, 1991. Elapsed time between the first recapture and mark date was 3 days, while the second recapture occurred 17 days after marking. This individual was observed using the same refugia at both recaptures, thus no data regarding movement were obtained. The final recapture was a female and occurred on April 9, 1991. The time elapsed between mark and recapture dates was 14 days, and distance moved was 1.81 m.

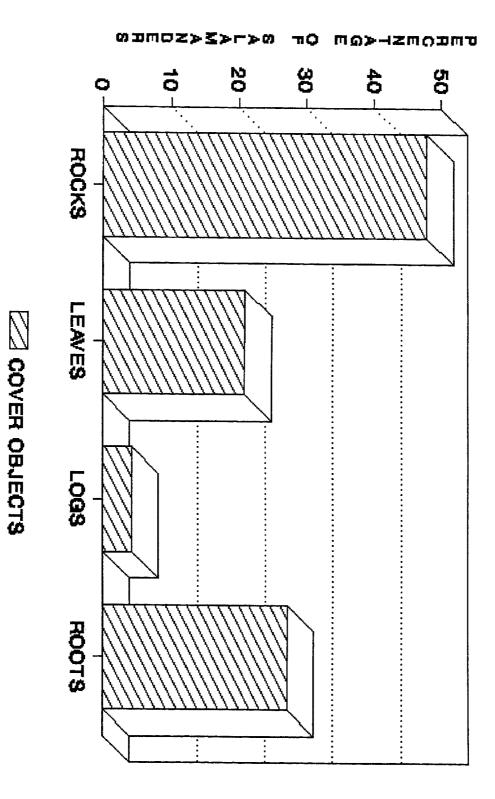
Cover Preference

The type of cover object utilized by 48 individual \underline{P} . <u>kentucki</u> was recorded during the study. Results indicated that rocks are utilized more frequently than leaves and logs (Figure 20). The percentage of salamanders utilizing rocks was 47.9 percent, while leaves constituted 20.8 percent of cover utilization, and logs 4.2 percent. A value of 27.1 percent was obtained for those <u>P</u>. <u>kentucki</u> found in crevices created by exposed roots of living trees. Figure 19. Movement of recaptured P. <u>kentucki</u> in quadrats 8 and 5. No movement was recorded for number 3 since it was recaptured twice under the same refugia.



#3C (NO MOVEMENT)

Figure 20. Percentage of <u>P</u>. <u>kentucki</u> found under rocks, logs, leaves, and in crevices beneath exposed roots of living trees. Percentages based on 48 specimens.



Aspect Studies

Aspect studies revealed that <u>P</u>. <u>kentucki</u> is present on west-facing slopes more frequently than on southwest-facing and northwest-facing slopes (Figure 21). The percentage of salamanders present on west-facing slopes was 44.8 percent, while southwest-facing and northwest-facing slopes exhibited lower percentages, with values of 31 percent and 14.6 percent respectively.

Vegetation Analysis

Identifications and counts of plant life along the horizontal line transect (80 m), and vertical line transect (75 m) were pooled to give a description of the plant species component (canopy, understory, shrubs) at the study site. Nineteen species were observed in the study. The percentage distribution of species in canopy, understory, and shrub layers are presented in Table 8. The most important canopy species in terms of frequency of occurrence was Fagus grandifolia (23.5%). Quercus prinus and Q. alba were common at the study site and constituted 20.6 percent, and 17.6 percent of the canopy species, respectively. The most common understory species was Fagus grandifolia (34.0%), followed by Oxydendrum arboreum (22.0%) and Cornus florida (16.0%). The shrub layer exhibited the greatest diversity with Vaccinium vacillans and Acer rubrum sharing the highest percentage of occurrence at 20.0 percent each. Asimina triloba and Cornus florida were important species and represented Figure 21. Percentage distribution of <u>P. kentucki</u> on southwest-, west-, and northwest-facing slopes. Percentages based on 47 specimens.

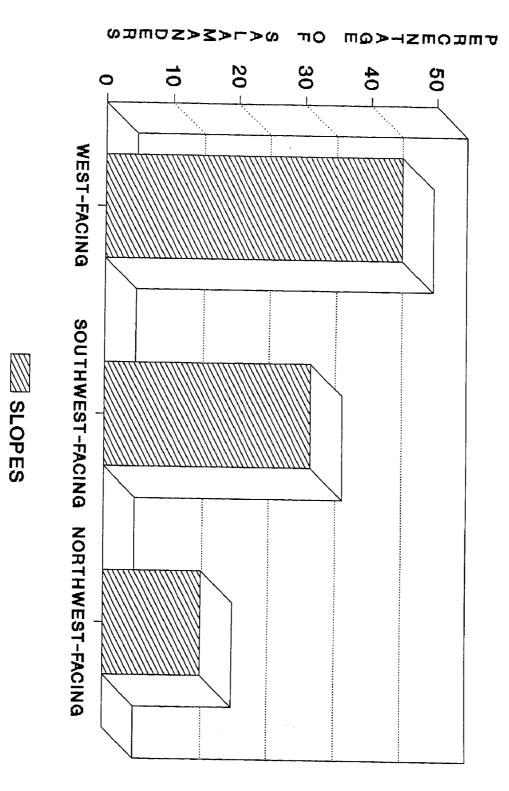


Table 8. Percent frequency occurrence of species in canopy, understory, and shrub layer at Beech Fork State Park.				
Canopy	Understory	Shrub		
Quercus alba (17.6%)	<u>Fagus</u> grandifolia (34.0%)	Acer rubrum (20.0%)		
<u>Quercus</u> velutina (2.9%)	Acer rubrum (8.0%)	Fagus grandifolia (1.7%)		
Quercus (2.9%)	Oxydendrum arboreum (22.0%)	<u>Fraxinus americana</u> (10.0%)		
Quercus prinus (20.6%)	Ulmus <u>americana</u> (2.0%)	Ulmus <u>americana</u> (1.7%)		
<u>Fagus</u> <u>grandifolia</u> (23.5%)	<u>Cornus</u> <u>florida</u> (16.0%)	<u>Sassafras</u> <u>albidum</u> (3.3%)		
Ulmus americana (14.7%)	Asimina triloba (10.0%)	<u>Cornus</u> <u>florida</u> (11.7%)		
<u>Carya</u> <u>ovata</u> (2.9%)	<u>Cercis</u> <u>canadensis</u> (6.0%)	<u>Cercis</u> <u>canadensis</u> (3.3%)		
<u>Nyssa</u> <u>sylvatica</u> (2.9%)	<u>Sassafras</u> <u>albidum</u> (2.0%)	Asimina triloba (15.0%)		
<u>Pinus</u> <u>virginiana</u> (11.8%)		<pre>Vaccinium vacillans (20.0%)</pre>		
		<u>Smilax</u> <u>rotundifolia</u> (6.7%)		
		Rhododendron calendulaceum (6.7%)		

15.0 percent and 11.7 percent of the shrub layer flora, respectively.

Sympatric Amphibians and Reptiles

Amphibian life was relatively diverse at the study site as there were nine species of salamanders, four species of frogs, and one toad species observed during the 15 month study (Table 9). Diversity was less for reptiles, however all three major groups were represented including four species of snakes, three species of lizards, and one species of turtle.

Discussion

Several studies have shown that environmental conditions, especially temperature and moisture, are important factors in the natural history and ecology of plethodontid salamanders (Heatwole 1962; Spotila 1972; Pauley 1977). Results of the present study indicate that such factors are important in the ecology of <u>P</u>. <u>kentucki</u> in West Virginia, particularly with reference to its activity. The population of <u>P</u>. <u>kentucki</u> studied appeared to be extremely seasonal with regard to activity. In fact, fifty of the seventy-four <u>P</u>. <u>kentucki</u> observed during the study were found within a two-month period. Within this short period of activity, similar numbers of males and females appeared on the surface, indicating no obvious difference in seasonality between sexes. However, a small number of juveniles were active on the surface

Table 9. Amphibians and reptiles living sympatrically with \underline{P} . <u>kentucki</u> at the study site.				
AMPHIBIANS	REPTILES			
Ravine Salamander (<u>Plethodon</u> richmondi)	Northern Ringneck Snake (<u>Diadophis punctatus</u> edwardsi)			
Slimy Salamander (<u>Plethodon</u> glutinosus	Eastern Garter Snake (<u>Thamnophis s. sirtalis</u>)			
Northern Dusky Salamander (Desmognathus <u>f</u> . <u>fuscus</u>)	Eastern Worm Snake (<u>Carphophis a. amoenus</u>)			
Southern Two-Lined Salamander (<u>Eurycea</u> <u>cirrigera</u>)	Northern Copperhead (<u>Agkistrodon contortrix</u> <u>mokasen</u>)			
Longtail Salamander (<u>Eurycea longicauda</u>)	Ground Skink (<u>Scincella lateralis</u>)			
Midland Mud Salamander (<u>Pseudotriton</u> <u>montanus</u> <u>diasticus</u>)	Broadhead Skink (<u>Eumeces laticeps</u>)			
Red-spotted Newt, Eft (<u>Notopthalmus v</u> . <u>viridescens</u>)	Northern Fence Lizard (<u>Sceloporus</u> <u>undulatus</u> <u>hyacinthinus</u>)			
Marbled Salamander (<u>Ambystoma opacum</u>)	Eastern Box Turtle (<u>Terrapene c. carolina</u>)			
Spotted Salamander (<u>Ambystoma</u> <u>maculatum</u>)				
Eastern American Toad (<u>Bufo a. americanus</u>)				
Wood Frog (<u>Rana sylvatica</u>)				
Northern Spring Peeper (<u>Pseudacris c. crucifer</u>)				
Mountain Chorus Frog (<u>Pseudacris b. brachyphona</u>)				
Cope's Gray Treefrog (<u>Hyla chrysoscelis</u>)				

during summer months when adults were virtually absent. It might be presumed that seasonal activity would be in direct relation to However, of the five the five microclimatic conditions recorded. conditions recorded in the seven quadrats during the study (air and soil temperature, % air RH, % soil moisture, and soil pH), percent soil moisture was the only condition identified as having a significant correlation with surface density, and thus seasonal activity of P. kentucki. Salamander activity, as indicated by the number of individuals on the surface, was greater when soil moisture was high than when it was low. This suggests a vertical migration from underground refugia to the surface when soil moisture is high enough to prevent desiccation. Similar vertical migrations related to soil moisture conditions have been demonstrated for other species of plethodontid salamanders. Heatwole (1962) reported that P. cinereus exhibits a vertical seasonal migration, and that surface abundance was greater when substrate moisture was high than when it was low.

In contrast to soil moisture, quadrat air and soil temperatures were not significantly correlated as individual factors with seasonal activity of <u>P</u>. <u>kentucki</u>. An example begins with the first month of the study, June 1990. Air and soil temperatures were comparatively high during this month. Similarly, the number of salamanders on the surface was relatively high. This situation might not be expected because higher temperatures increase the risk of desiccation to plethodontid salamanders. However, the increased risk of

desiccation are negated somewhat when moisture is high. Consequently, surface density was relatively high during June 1990 because soil moisture was high, which probably acted to negate the risks of desiccation accompanied by high temperatures. During the months that followed, soil moisture decreased and did not increase substantially until January 1991, where it stabilized and remained high until April. Surface density followed the same pattern and did not exhibit a substantial increase until February 1991. It was during March and April 1991 that surface abundance was highest. However, soil and air temperatures for March and April were comparably much lower than those obtained in June 1990 when surface density was also high. In fact, there was a difference of over nine degrees °C in soil temperature between June 1990 and March 1991. Contrastingly, soil moisture for April 1991 differed from June 1990 by as little as one half of a percent. The extreme differences in temperature, but subtle differences in soil moisture between the months of highest surface densities, illustrate the fact that air and soil temperatures are not significantly correlated with seasonal activity as individual factors. It appears that there is a broad range of temperatures in which P. kentucki might be active, granted soil moisture is near optimum.

Quadrat air and soil temperatures were compared to on-site air and soil temperatures for March and April 1991, when surface density was highest. Results indicated that quadrat air temperatures were significantly higher than on-site air

temperatures for both months, while quadrat and on-site soil temperatures did not differ significantly. It is likely that air temperatures differed because most of the on-site temperatures were recorded at night, while all quadrat temperatures were taken between 12:00 PM and 4:00 PM EST. The insulating effects of leaf litter most likely keep soil temperatures uniform throughout a 24 hour period, thus explaining why soil temperatures were not significantly different. Although air temperature was not significantly correlated with seasonal activity, it is probable that it would be so correlated with diel activity. This is suggested because 90 percent of P. kentucki observed during the study were found at night, when air temperatures were significantly cooler than those obtained during the hours of 12:00 PM and 4:00 PM. In fact, results of diel activities revealed that P. kentucki is nocturnal, and most active between the hours of 9:00 PM and 10:00 PM EST. This is consistent with Gordon et al. (1962) in which surface abundance of adult \underline{P} . yonahlossee was found to peak between 9:00 PM and 10:00 PM. Soil temperature might also be correlated with diel activity. However, because of its uniformity during a 24 hour period, soil temperature probably doesn't restrict salamander activity to the degree that air temperature does.

Air relative humidity was not significantly correlated with seasonal activity of <u>P</u>. <u>kentucki</u> at the study site. However, studies have shown that air relative humidity influences the daily activity of some <u>Plethodon</u> salamanders. Spotila (1972)

studied several <u>Plethodon</u> species including <u>P</u>. <u>glutinosus</u>, and found that salamander activity was apparently dependent on the moisture content of the air. When subjected to RH gradient tests, he found that salamanders actively select the highest available level of atmospheric moisture. Air relative humidity is generally lower during the day than at night (Smith 1980). Because most of the <u>P</u>. <u>kentucki</u> observed during the study were found at night, it is probable that air relative humidity is related to its daily activity.

Soil pH was relatively uniform throughout the study, and thus it was not significantly correlated with seasonal activity. The average soil pH at the study site was 5.7. Although this value is acidic, studies have reported that many amphibians appear to be relatively acid tolerant and can survive in acid solutions of pH 4-4.5 (Pierce 1985).

As stated on several occasions in this study, the interactions of <u>P</u>. <u>kentucki</u> with <u>P</u>. <u>glutinosus</u> most likely play a key role in the distribution and ecology of this species. In terms of total range and distribution, <u>P</u>. <u>glutinosus</u> is one of the most successful plethodontid species in eastern United States (Conant and Collins 1991). Consequently, this species is successful in a wide range of environmental conditions. Comparably, <u>P</u>. <u>kentucki</u> is not as successful and occupies but a small portion of eastern United States. Thus, one assumption might be that <u>P</u>. <u>kentucki</u> is not sympatric with its congener throughout eastern United States because it lacks the ability to

adjust to the varying environmental conditions at different geographic locations. In the present study, each species was subjected to Critical Thermal Maximum (CTM) and dehydration studies for comparisons of tolerances to high temperatures and water loss. Results indicated that P. glutinosus has a higher CTM value, however, it was not significantly higher than the value obtained for P. kentucki. In addition, dehydration values were not significantly different for the two species. This suggests that each is equally well adapted for survival in the thermal and moisture conditions they are confronted with in West Virginia. However, it does not explain why P. kentucki is absent in northern counties of West Virginia where P. glutinosus is abundant. This matter was further complicated when environmental conditions from the study site in southwestern West Virginia were compared to conditions from a northwestern site (Jewell 1991). There were no significant differences in mean air temperatures, soil temperatures, and air relative humidities between Wayne County (southwestern WV), and Ritchie County (northwestern WV) for the months of June 1990 through March 1991. In addition, there was no significant difference in average annual rainfall (1947 - 1976) from areas near the two sites (Lee et al. 1977). Similar environmental conditions for the two sites might explain why P. glutinosus is successful in both southern and northern West Virginia. However, based on this information, there is no apparent reason why P. kentucki does not occupy counties in northern West Virginia. It may be that competition with \underline{P} .

<u>glutinosus</u> is effectively limiting the range and distribution of <u>P. kentucki</u> in West Virginia, and possibly throughout eastern United States. This situation is not unique among salamanders of the genus <u>Plethodon</u>. Hairston (1951) reported that the restriction of <u>P</u>. <u>glutinosus</u> to lower elevations in the Blue Ridge Mountains is apparently due to intense competition with <u>P</u>. <u>jordani</u>. Jaeger (1972) demonstrated that both <u>P</u>. <u>shenandoah</u> and <u>P. cinereus</u> choose moist, deep soil, but that <u>P</u>. <u>cinereus</u> via competitive interactions, restricts its congener to the dry talus. Thus, in the present study, it is postulated that <u>P</u>. <u>kentucki</u> is limited in its range and distribution as a consequence of competition with <u>P</u>. <u>glutinosus</u>. Cool moist spots may be the limited resource for which these two species compete. This is supported by the results of CTM and dehydration studies.

Information regarding movement patterns of <u>P</u>. <u>kentucki</u> is limited to four individuals. An adult female was recaptured twice under the same cover object. Although zero movement was recorded for this individual, it most likely did not remain stationary during the time period between captures. It is assumed that this individual actively moved about within the general vicinity of its territory. A movement of 1.32 m was recorded for one juvenile, while the distance moved for an adult female was 1.81 m. Although these distances appear insignificant, they are consistent with results of other studies showing that movements and home ranges for <u>Plethodon</u> are relatively small. Wells and Wells (1976) studied movement

patterns of <u>P</u>. <u>glutinosus</u> in North Carolina and reported home ranges of less than 9 m in diameter for adult females, and less than 6 m in diameter for juveniles. Merchant (1972) studied <u>P</u>. <u>jordani</u> and found mean movement distances to be 2.3 m for females, and 2.0 m for juveniles.

Plethodon kentucki utilized rocks as cover more frequently than leaves and logs. It appeared that most of the rocks with salamanders under them extended deeper into the soil than rocks without salamanders. In addition, soils under the deeper rocks appeared to have a higher moisture content. Although data were not collected for analysis, it is postulated that these rocks were preferred as cover because they provide a microhabitat with higher substrate moisture, which would effectively reduce the risk of desiccation and provide areas to obtain food when leaf litter is dry. Although logs were relatively abundant at the study site, they were utilized as cover less frequently than rocks and leaves. Soils beneath logs appeared drier than soils under rocks. This may be due to the fact that most logs did not extend into the soil as deep as rocks. Although several \underline{P} . kentucki were observed in crevices created by exposed roots near the base of living trees, they were not considered to be cover objects in this study. This is due to the fact that they could not be turned in the same manner as rocks and logs. However, since they appeared to be important microhabitats for \underline{P} . kentucki, these observations were included. Soil moisture in this microhabitat may have been supplemented by stemflow during

periods of rain (Smith 1980). It appeared that <u>P</u>. <u>kentucki</u> was restricted to cover objects and microhabitats that provided ample substrate moisture.

Aspect is an important factor to consider when discussing microdistribution patterns of terrestrial salamanders such as \underline{P} . kentucki. Since moisture appears to be the critical factor influencing activity of P. kentucki, it seems justified to assume this species would selectively "choose" the direction of slope that fulfills its moisture requirements. North- and east-facing slopes receive lesser amounts of insolation than south- and westfacing slopes. The amount of solar energy on a slope controls its moisture content. Thus, high temperatures tend to increase evaporation of moisture from the soil (Smith 1980). Aspect studies revealed that P. kentucki occurs on west-facing slopes more frequently than on southwest- and northwest-facing slopes. This contradicts what was expected since west- and southwestfacing slopes are, in general, warmer and drier than northwestfacing slopes. Quantitative data for soil moisture from each aspect was not collected for comparison, thus microdistribution patterns cannot be explained using this factor. Although the numbers were low (5), P. glutinosus was observed at the study site. Plethodon kentucki may have been confined to west-facing slopes as a result of competition with \underline{P} . glutionsus.

Plant life was relatively diverse at the study site with 19 species representing the canopy, understory, and shrub layers. Beech Fork State Park was aptly named as <u>Fagus</u> <u>grandifolia</u> was

the most important canopy and understory species in terms of frequency of occurrence. <u>Vaccinium vacillans</u> and <u>Acer rubrum</u> were equally important in the shrub layer. Although not attempted in the present study, it has been shown that specific plant species often act as indicators of occurrence for <u>Plethodon</u> species. Pauley (1977) in Harrison County West Virginia, reported that high densities of <u>P</u>. <u>cinereus</u> tended to correlate with the occurrence of sugar maple-beech as the understory, along with an abundance of herbaceous plants and absence of shrubs. Contrastingly, he found that the occurrence of <u>P</u>. <u>wehrlei</u> was correlated with sourwood-red maple as understory, and the prevalence of shrubs rather than herbaceous plants. If such a correlation existed for <u>P</u>. <u>kentucki</u> at the study site, it most likely would involve <u>Fagus grandifolia</u>.

Observations of amphibians and reptiles living sympatrically with <u>P</u>. <u>kentucki</u> at the study site were recorded. This information was valuable for extrapolating the contribution of certain species to factors affecting <u>P</u>. <u>kentucki</u>, such as competition and predation. As Table 8 indicates, amphibian life was diverse, particularly with regard to salamanders (9 species). Although exact numbers of individuals of each species are not available, <u>P</u>. <u>richmondi</u> (ravine salamander) appeared to be the most abundant salamander, followed by <u>Eurycea cirrigera</u> (southern two-lined salamander). Although smaller than <u>P</u>. <u>kentucki</u>, <u>P</u>. <u>richmondi</u> is a terrestrial salamander that could compete with its larger congener for limited resources such as moist spots and

food. Eurycea cirrigera is also smaller than <u>P</u>. <u>kentucki</u> and is generally considered to be a semi-terrestrial salamander. The top competitor for <u>P</u>. <u>kentucki</u> at the study site was probably <u>P</u>. <u>glutinosus</u>. However, it was found in very low numbers (5 specimens), thus its interactions with <u>P</u>. <u>kentucki</u> may be limited.

Several species of snakes were encountered during the study, including two which are known to prey on terrestrial salamanders (personal observation). <u>Diadophis punctatus</u> <u>edwardsii</u> (northern ringneck snake) and <u>Thamnophis s. sirtalis</u> (eastern garter snake) appeared to be the most abundant snakes at the study site. Conant and Collins (1991) noted that salamanders are part of the diet of each of these snakes.

Summary

The population of <u>P</u>. <u>kentucki</u> studied appeared to be extremely seasonal with regard to activity. Approximately 70 percent of all salamanders were found within a two month period (March and April 1991). Seasonal activity was not significantly correlated with air temperature, soil temperature, air relative humidity, or soil pH. Seasonal activity was significantly correlated with soil moisture. Surface density was greatest when soil moisture was high. This was true when temperatures were high and low, suggesting that there is a wide range of temperatures at which <u>P</u>. <u>kentucki</u> might be active, given soil

moisture is near optimum. This further suggests a vertical migration from underground refugia to the surface when soil moisture is high enough to effectively prevent desiccation of the salamander.

There was no obvious difference in seasonal activity between males and females. However, a small number of juveniles appeared on the surface during summer months when adults were virtually absent.

There was no significant difference in CTM and dehydration values between <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u>. This suggests both are equally well adapted to moisture and temperature conditions in West Virginia. However, due to its absence in northern counties where <u>P</u>. <u>glutinosus</u> is abundant, it is postulated that <u>P</u>. <u>kentucki</u> is limited to southwestern West Virginia as a consequence of competition with its congener. A possible resource for competition might be cool moist spots, a postulation which is supported by similar CTM and dehydration values.

Daily activity of <u>P</u>. <u>kentucki</u> began after 7:00 PM EST and gradually increased throughout the evening until it reached a climax between the hours of 9:00 and 10:00 PM.

Movement patterns of <u>P</u>. <u>kentucki</u> were limited to two individuals. A juvenile moved a linear distance of 1.32 m, and an adult female was recaptured 1.81 m from its original position of capture.

Rocks were utilized as cover more frequently than leaves and logs. It appeared that <u>P</u>. <u>kentucki</u> was restricted to refugia

that provided adequate substrate moisture, which might act to reduce the risk of desiccation. Rocks appeared to extend deeper into the soil than logs, and soil moisture beneath them was apparently greater. Although they were not considered to be cover objects, crevices beneath exposed roots of living trees appeared to be important microhabitats for <u>P. kentucki</u>.

Contrary to what was expected, <u>P</u>. <u>kentucki</u> was more abundant on west-facing slopes than on southwest- and northwestfacing slopes. Because comparative data from each aspect was not obtained, this microdistribution pattern was not explained using environmental conditions. However, it was suggested that <u>P</u>. <u>glutinosus</u> might be confining <u>P</u>. <u>kentucki</u> to west-facing slopes at the study site.

Fagus grandifolia was the most important canopy and understory plant species at the study site in terms of frequency of occurrence. Important shrubs were <u>Vaccinium vacillans</u> and Acer rubrum.

Amphibian and reptile life at the study site was relatively diverse. Important salamanders with regard to possible competitive interactions with <u>P. kentucki</u> were <u>P. richmondi</u>, and <u>P. glutinosus</u>. Several species of snakes were observed, including two which are known to prey upon salamanders. <u>Thamnophis s. sirtalis</u> and <u>Diadophis punctatus edwardsii</u> appeared to be the most common snakes.

CHAPTER III

Competition as a Possible Factor Limiting the Range of Plethodon <u>kentucki</u> in West Virginia

Introduction

Several studies concerning competition among salamanders have been conducted. Hairston (1980) demonstrated that competition is important in determining the altitudinal distributions of <u>P</u>. <u>glutinosus</u> and <u>P</u>. <u>jordani</u> in North Carolina. In Virginia, Jaeger (1972) concluded that the allopatric distribution of <u>P</u>. <u>cinereus</u> and <u>P</u>. <u>shenandoah</u> is maintained via competitive interactions, with <u>P</u>. <u>cinereus</u> excluding its congener from habitats preferred by both species. Thurow (1975) studied several species of large and small eastern <u>Plethodon</u> and observed behaviors such as aggression, feeding competition, social dominance, and territoriality.

A laboratory approach was followed in the present study in which an attempt was made to measure behavioral aspects of competition between <u>P. kentucki</u> and <u>P. glutinosus</u>. <u>Plethodon</u> <u>kentucki</u> and <u>P. glutinosus</u> maintain sympatric distributions throughout the extent of <u>P. kentucki</u> total range. However, in West Virginia, <u>P. kentucki</u> is restricted to the southwestern portion of the state, whereas <u>P. glutinosus</u> occurs statewide. Possible explanations for this limited range include the natural barrier imposed by the New-Kanawha river system (see Chapter 1). Although rivers are not typically considered to be barriers for Plethodon species, Highton (1972) suggested that the New River in West Virginia, in combination with the presence of a closely related species (P. richmondi), may be effectively confining P. hoffmani to the Valley and Ridge province. This suggestion may explain the limited range of P. kentucki in West Virginia. Behavioral interactions among and between P. kentucki and P. glutinosus including aggressive behavior, chemical communication, intraspecific, and interspecific interference competition have been studied in Kentucky (Delpont 1987; Marvin 1990). Delpont (1987) found that male and female P. kentucki appeared to use chemical signaling as well as threat displays to deter conspecific intruders. Marvin (1990) concluded that male and female P. kentucki and P. glutinosus will aggressively defend areas against conspecifics and congenerics in laboratory situations, indicating that they may compete for some limited resource(s). Furthermore, P. glutinosus tended to be the dominant species in interspecific encounters. If similar results were obtained from other geographical locations including West Virginia, much evidence would be available to support the contention that competition exists between these two species and most likely plays a key role in limiting the range of P. kentucki in West Virginia. There were two objectives in the present study. The first was to determine if competition exists between P. kentucki and P. glutinosus in West Virginia. The second was

to determine what effects competition might have on the limited range of <u>P</u>. <u>kentucki</u> in West Virginia.

Materials and Methods

Salamanders were collected in the field, returned to the laboratory and randomly placed in individual one gallon jars. Each jar was placed on its side and contained approximately one inch of potting soil. This produced a substrate that was level with the mouth of the jar. Jars were placed mouth to mouth, but for seven days lids isolated the salamanders so that they could not exchange physical, visual, or olfactory communications. The seven-day period was provided to allow time for salamanders to mark areas with pheromones and establish territories. The salamanders were kept at 20 °C with a photoperiod of 12L:12D. They were fed <u>Drosophila melanogaster</u> once a week and watered twice a week. Techniques were similar to those described by Jaeger et al. (1982).

During a three week period, 21 pairs of salamanders ($\underline{P} \cdot \underline{k}$. vs. $\underline{P} \cdot \underline{q} \cdot$) were observed for competitive interactions for one hour under a red light by removing the lids between paired jars. Only males were used in the experiment with a total of 21 interactions observed.

Seven behavioral states were observed for a salamander. Initiator: the first salamander to move from its original position, or raise its head in response to airborne pheromones. Aggressor: that salamander which threatens or attacks the other, including biting (Thurow 1975). In the present study, aggressive behavior included exhibiting certain characteristic poses or movements such as vertically extending the fore- and or hindlegs, raising the head, and thus forming a "U" shaped appearance (Jaeger et al. 1982). Appeaser: that salamander which positions itself to avoid or hinder an attack from another (Thurow 1975). Intruder: a salamander that entered the chamber (territory) of the other during the experiment (Jaeger et al. 1982). Defender: that salamander whose chamber (territory) was approached and entered by another (Jaeger et al. 1982). Escaper: that salamander which attempted to flee the area due to a physical interaction or in response to a perceived threat via airborne pheromones or threat displays. In the present study, movement towards the periphery of the test chamber was indicative of an escape attempt (Tristram 1977). Biter: that salamander which grasped or snapped another during an interaction (Wrobel et al. 1980).

A statistical analysis of the results included chi-square, where P < 0.05, and df=1. This test was used to determine significant differences in the frequencies of behavioral states exhibited by both salamander species.

Results

The data were partitioned and a separate Chi-square was

calculated for each behavioral state (Figure 22). There were no significant difference in frequency (P < 0.05) for the initiator, aggressor, escaper, and biter behavioral states. <u>Plethodon</u> <u>kentucki</u> was the appeaser significantly more often than <u>P</u>. <u>glutinosus</u> (P < 0.05). <u>Plethodon kentucki</u> was the intruder significantly more often than <u>P</u>. <u>glutinosus</u> (P < 0.05). <u>Plethodon kentucki</u> was the intruder significantly more often than <u>P</u>. <u>glutinosus</u> (P < 0.05). <u>Plethodon kentucki</u> was the intruder significantly more often than <u>P</u>. <u>glutinosus</u> (P < 0.05). <u>Plethodon kentucki</u> (P < 0.05).

Discussion

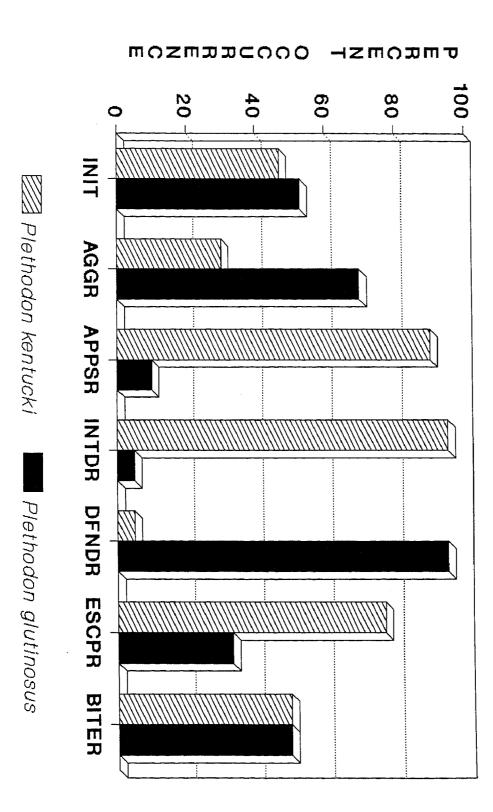
Previous laboratory studies indicated that individuals of two species could influence each other's spatial position through aggressive interactions (Thurow 1975). The present study provides evidence that a competitively superior salamander species (based on a higher incidence of territoriality) may be effectively excluding another from otherwise suitable habitats. Results of the present study for behavioral states observed are discussed below.

Initiator

An initiator is that salamander which moves from its initial position, possibly in response to airborne pheromones. In this experiment, frequency for the initiator category was not associated significantly with either species. Marvin (1990) found that residents of either species initiated aggression more

Figure 22. Results of the seven behavioral states observed during competition studies between <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u>.

APPSR, INTDR, DFNDR, SIG. (P < 0.05)



often than intruders with no significant differences between species. This may be due to similarities in the ability to detect the presence of another salamander either by visual or chemical cues. It may be that <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> responded approximately equally in this category because neither species appeared to be better at detecting airborne or visual cues. However, Marvin (1990) found that individuals of <u>P</u>. <u>kentucki</u> exhibit head-swaying more than individuals of <u>P</u>. <u>glutinosus</u>, which may be a behavior used in detecting airborne pheromones.

Aggressor

Although P. glutinosus exhibited the highest percentage of aggressive behaviors, there was no significant difference in frequency for the aggressor category. Aggressive behaviors such as characteristic poses are important since they often work to avoid physical confrontation (biting), which can be detrimental to the individuals involved.

Appeaser

A salamander appeases to avoid or hinder an attack from another. Frequency for the appeaser category was associated significantly with <u>P. kentucki</u>. The appeasing salamander is often associated with characteristic poses such as extending the legs horizontally, with the venter and chin pressed against the substrate (Jaeger 1981). Jaeger (1981) suggested that this

posture is exhibited most often when escape from an attacking salamander is impossible, and it prevents foraging (Jaeger et al. 1981). This provides direct evidence that \underline{P} . <u>kentucki</u> is an inferior competitor when compared to \underline{P} . <u>glutinosus</u>. Applied to natural settings, \underline{P} . <u>kentucki</u> would, in effect, be the "loser" if a particular competitive interaction involved food.

Intruders / Defenders

An intruder is defined as a salamander that invades the area (territory) of another. Plethodon kentucki was the intruder significantly more often than P. glutinosus in this experiment, suggesting that it is less territorial in nature. Territoriality implies that an area is defended for the purposes of having a store of necessary resources. Based on this implication, it is possible to measure or detect the competitive ability of a species by evaluating its inherent territorial behaviors. Jaeger et al. (1981) suggested that individuals of \underline{P} . <u>cinereus</u> establish territories in nature in defense of a limited but reliable supply of food. This behavior may be an indication of intense competition with another species, and may have evolved as a result of competition at some time in its evolutionary history. Since P. kentucki was the predominant intruder, it thus appears to be less territorial, and consequently, less apt to allocate and or secure resources which may be limited in the presence of competition. Contrastingly, P. glutinosus was the predominant

defender (more territorial), and thus more apt to successfully allocate limited resources in the presence of intense competition. This supports the contention that <u>P</u>. <u>glutinosus</u> is competiting with <u>P</u>. <u>kentucki</u>, and that competition appears to be playing a role in limiting <u>P</u>. <u>kentucki</u> to counties southwest of the Kanawha and New Rivers in West Virginia.

Escapers / Biters

Although P. kentucki exhibited a higher percentage of escapes, there was no significant difference in frequency for the escaper category. Escape behavior is important because an intense aggressive interaction can be costly to the loser (i.e. the one receiving the bite). Jaeger (1981) noted that a bite to the trunk can do little harm, while a bite to the tail of another may result in the loss of energy reserves due to autotomy of the tail. Bites to the snout may also be detrimental, as damage to the nasolabial grooves can reduce chemoreception in the bitten animal (Jaeger 1982). Although the animal that escapes is often regarded as the loser, its difficult to ascribe the escaper a loser. This is because an escaper is avoiding a confrontation which may cause physical harm. On the other hand, an escaper may be effectively excluded from a potential necessary resource. At this point, it is virtually impossible to determine which of the above scenarios out-weighs the other with regard to competition.

Summary

Based on results of a laboratory study, competitive interactions occur between P. kentucki and P. glutinosus in West This can be seen in the results of the appeaser, Virginia. intruder, and defender categories, each of which were Plethodon kentucki was the predominate appeaser significant. during the study. During appeasements, the chin and venter of appeasing salamanders are pressed against the substrate, a position which may effectively prevent foraging (Jaeger et al. 1981). Plethodon kentucki was the predominate intruder, and \underline{P} . glutinosus the predominate defender, suggesting a higher degree of territoriality in the latter. This was taken as evidence that P. glutinosus is better able to successfully allocate and secure necessary resources in the face of intense competition. Marvin (1990) found several lines of evidence concerning competition between these two species including territorial defense of areas via aggressive behaviors, and demonstration of aggressive behaviors such as biting, lunging, and snapping by both species. He also found that P. glutinosus was the dominant species in interspecific encounters. Thus, if these two species were competing for cover objects, <u>P</u>. <u>glutinosus</u> would exclude <u>P</u>. kentucki from microhabitats favorable to both. Field observations of competitive interactions between these two species will add valuable information to existing laboratory findings. In addition, field studies will aid in understanding

the importance of competition with regard to the limited range of \underline{P} . <u>kentucki</u> in West Virginia. With this idea in mind, this preliminary study leads to the following conclusions: (1) competition does occur between \underline{P} . <u>kentucki</u> and \underline{P} . <u>glutinosus</u> in West Virginia; (2) competition is suggested through appeasements and territoriality; (3) competition is most likely playing a key role in limiting the range \underline{P} . <u>kentucki</u> in West Virginia.

CHAPTER IV

A Comparative Analysis of Prey Selection between <u>Plethodon kentucki</u> and <u>Plethodon glutinosus</u> in West Virginia

Introduction

A wealth of information exists concerning the feeding habits of salamanders. A popular consensus is that most species are euryphagic and feed on a wide variety of small invertebrates, especially insects. Their indiscriminate feeding habits appear to be restricted only by the size of prey, taking whatever they are physically able to ingest. Mitchell and Taylor (1986) reported that collembolans, centipedes, spiders, and insect larvae were important food items for <u>P. jordani</u> in North Carolina. Pauley and Little (unpublished data) studied the feeding habits of several salamander species including <u>P</u>. <u>cinereus</u>, in the Fernow Experimental Forest in West Virginia. Their data suggested that ants, winged hymenopterans, beetles, and arachnids are important in the diet of P. cinereus.

An important factor that is often considered when investigating the feeding habits of sympatric species is competition. Burton (1976) investigated the feeding ecology of five salamander species in the Hubbard Brook Experimental Forest in New Hampshire. His interest focused on determining whether food was a item for which salamanders were competing interspecifically. He concluded that interspecific competition for food was reduced via gradations in the sizes of salamanders, varying life histories, and differential utilization of habitats.

Jaeger (1972) conducted a study to determine if food was a limited resource in interspecific competition between <u>P</u>. <u>cinereus</u> and <u>P</u>. <u>shenandoah</u> in Virginia. He suggested that food could be a limited resource for these two species when conditions were dry enough to limit the availability of prey.

Results of competition studies (Chapter 3) revealed that \underline{P} . <u>glutinosus</u> is apparently a superior competitor to \underline{P} . <u>kentucki</u> in the laboratory. Since food is often suggested to be a limited resource for competition between species, a comparative analysis of prey selection was conducted in the present study to determine the importance of food as a limited resource for competition between P. kentucki and P. glutinosus.

Methods and Materials

<u>Plethodon kentucki</u> and <u>P</u>. <u>glutinosus</u> were collected at various locations throughout southwestern West Virginia. Daytime collecting techniques involved turning all suitable rocks and logs, and raking leaf litter. Night collections involved using flashlights to scan the forest floor. Most night collections were made during rainy and or wet weather. After capture, specimens were immediately placed into small jars containing 5

percent formalin, and returned to the laboratory for analysis of stomach contents. Several specimens of <u>P</u>. <u>glutinosus</u> were obtained from the West Virginia Biological Survey at Marshall University, and were used for stomach content studies. Prey items were removed from the stomachs and identified to order. Spearman's rank correlation coefficient was employed to determine the similarity of diet between the two species. Snout-ventlength (SVL) and jaw width of <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> were compared to determine if feeding habits might be partitioned by prey size.

Results

The stomachs of 70 <u>P</u>. <u>kentucki</u> and 79 <u>P</u>. <u>glutinosus</u> were analyzed for food contents (Table 10). Prey selection was significantly correlated, thus indicating a high degree of similarity in feeding habits for the two species: Spearman's rank correlation coefficient, two-sided; P < 0.025. This correlation was based on pooled numbers of 18 individual prey categories.

A total of 634 individual food items were obtained from stomachs of <u>P</u>. <u>kentucki</u> specimens, and 526 food items from <u>P</u>. <u>glutinosus</u>. Stomachs of 5 <u>P</u>. <u>kentucki</u> and 11 <u>P</u>. <u>glutinosus</u> were empty. Hymenopterans (mostly ants) were found in stomachs (percent frequency) of both species more frequently than any other prey type (<u>P.k</u>. 71.4%; <u>P.g</u>. 54.4% of stomachs), followed by coleopterans (<u>P.k</u>. 40.0%; <u>P.g</u>. 32.9%) and gastropods (<u>P.k</u>.

Table 10. Distribution of food items in stomachs of 70 \underline{P} . <u>kentucki</u> and 79 \underline{P} . <u>glutinosus</u> in West Virginia.						
Group	Plethodor % of stomachs	n <u>kentucki</u> % of food items	Plethodon % of stomachs	glutinosus % of food items		
Coleoptera	40.0	7.1	32.9	8.7		
Hymenoptera	71.4	44.6	54.4	48.9		
Lepidoptera	5.7	0.6	2.5	0.4		
Hemiptera	1.4	0.3	1.3	0.2		
Diptera	20.0	4.1	10.1	2.3		
Orthoptera	2.9	0.3	2.5	0.4		
Collembola	24.1	9.8	8.9	3.6		
Thysanura	1.4	0.2	0.0	0.0		
Dermaptera	0.0	0.0	1.3	0.2		
Araneidae	28.6	5.7	13.9	3.2		
Acari	21.4	3.6	16.5	4.0		
Chilopoda	12.9	1.6	10.1	2.1		
Diploda	15.7	2.2	15.2	3.2		
Gastrapoda	30.0	5.0	30.4	7.6		
Isopoda	4.3	0.6	15.2	4.9		
Pseudoscorp	25.7	5.0	6.3	1.1		
Nematoda	0.0	0.0	1.3	0.2		
Oligochaeta	5.7	0.9	3.8	0.8		
Other	48.6	8.2	36.7	8.2		

30.0%; <u>P.g.</u> 30.4%). Although there was a significant correlation in overall feeding habits, four prey groups appeared to be more important in the diet of <u>P. kentucki</u>: spiders (Araneida) were found in 28.6 percent of <u>P. kentucki</u> stomachs, and in 13.9 percent of <u>P. glutinosus</u> stomachs; flies (Diptera) 20.0 percent of <u>P. kentucki</u> stomachs, 10.1 percent of <u>P. glutinosus</u>; springtails (Collembola) 24.1 percent of <u>P. kentucki</u> stomachs, 8.9 percent of <u>P. glutinosus</u>; pseudoscorpions (Pseudoscorpionida) 25.7 percent of <u>P. kentucki</u> stomachs, 6.3 percent of <u>P. glutinosus</u>. Sowbugs (Isopoda) appeared to be more important in the diet of <u>P. glutinosus</u> (15.2% of stomachs) than in <u>P. kentucki</u> (4.3%).

The mean (± 1 S.E.) SVL, and head width at the widest point for <u>P. kentucki</u> and <u>P. glutinosus</u> are presented in Table 11. <u>Plethodon glutinosus</u> (mean svl = 66.27 mm) was significantly larger than <u>P. kentucki</u> (59.6 mm): P < 0.05. However, head width was not significantly different (<u>P. g. mean = 9.94 mm; P. k</u>. mean = 9.69 mm): P > 0.05.

Discussion

Based on the diversity of food types found in stomachs of <u>P. kentucki</u> and <u>P. glutinosus</u>, it appears each is euryphagic and will take any available prey they are able to capture and physically ingest. A comparison of diets for the two species revealed a high similarity in food types eaten. In fact, the

Table 11. Comparison of snout-vent-length and head width for <u>P</u> . <u>kentucki</u> and <u>P</u> . <u>glutinosus</u> . * Indicates significant difference, where P < 0.05.					
SPECIES		N	MEAN	S. E.	
	SVL	53	* 59.6	0.64	
\underline{P} . \underline{k} .	HW	32	9.69	0.14	
_	SVL	105	* 66.3	0.69	
<u>P</u> .g.	Н₩	40	9.94	0.15	

three most important food groups were identical for both species. This might indicate that food is an object of competition.

Based on snout-vent lengths, <u>P</u>. <u>glutinosus</u> is significantly larger than <u>P</u>. <u>kentucki</u>. More importantly, head width at the widest point is not significantly different for the two species. Because head width is similar, it is predicted that prey size does not partition the feeding habits of <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u>, and probably increases the intensity of competition. This would further indicate that food is most likely a limited resource for which these species compete.

As was mentioned earlier (Chapter 3), competitive interactions occur between <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> in West Virginia. In addition, <u>P</u>. <u>glutinosus</u> was more territorial in nature, and thus appeared to be the superior competitor. Due to the similarity in diet, it is suggested that the limited range of <u>P</u>. <u>kentucki</u> in West Virginia is due, in part, to competitive interactions for food with <u>P</u>. <u>glutinosus</u>.

Summary

<u>Plethodon kentucki</u> and <u>P. glutinosus</u> feed on the same types of prey. This similarity in diet leads to the conclusion that competition for food probably occurs between these two species. Head width was not significantly different, thus suggesting that prey size probably does not partition feeding habits, and most likely increases the intensity of competition for food.

Conclusions

<u>Plethodon kentucki</u> is restricted to southwestern counties in West Virginia. With the exception of Fayette and Summers counties, no <u>P</u>. <u>kentucki</u> were found east of the New-Kanawha River system. Since elevation is important regarding the distribution of many salamander species, it was hypothesized that the greater mean elevation for Fayette and Summers counties, as compared to lower lying counties along this river system, provide the environmental conditions most suitable for the colonization of new areas by <u>P</u>. <u>kentucki</u>. In addition, it was hypothesized that the Kanawha River, which is typically wider throughout its length than the New River, is acting as a natural barrier preventing the establishment of new populations on the opposing side.

The population of <u>P</u>. <u>kentucki</u> at Beech Fork State Park appeared to be extremely seasonal with regard to activity. However, there was no obvious difference in seasonal activity between males and females. This seasonality was significantly correlated with soil moisture at both high and low temperatures. This suggests that there is a wide range of temperatures at which <u>P</u>. <u>kentucki</u> might be active, given soil moisture is near optimum. In addition, it suggests that <u>P</u>. <u>kentucki</u> migrates from underground burrows to the surface when soil moisture is high enough to prevent desiccation.

<u>Plethodon kentucki</u> and <u>P. glutinosus</u> appear to be equally well adapted to the moisture and temperature conditions in West

Virginia. With this evidence, it was then postulated that competition, rather than environmental factors (temperature and moisture), is limiting <u>P</u>. <u>kentucki</u> to the southwestern counties in West Virginia. The limited resource for competition may be cool moist spots, a suggestion supported by results of CTM and dehydration studies.

Nocturnal habits of <u>P</u>. <u>kentucki</u> were evident in that peak activity occurred between 9:00 and 10:00 PM EST. As expected, linear movements of <u>P</u>. <u>kentucki</u> were limited to less than two meters. A juvenile moved a linear distance of 1.32 meters, and an adult female 1.81 meters.

Rocks were utilized as cover more frequently than leaves and logs. It is probable that <u>P</u>. <u>kentucki</u> preferred this type of cover as a result of the apparent wetter substrate beneath them.

<u>Plethodon kentucki</u> was found more frequently on west-facing slopes than on southwest- and northwest-facing slopes. This was not expected since environmental conditions are typically more extreme on west-facing slopes. Because comparative data were not collected from each aspect, this microdistribution pattern could not be explained using environmental conditions. Although it was found in low numbers at the study site, <u>P. glutinosus</u> might be confining <u>P. kentucki</u> to west-facing slopes.

Based on the results of a laboratory study, competition occurs between <u>P</u>. <u>kentucki</u> and <u>P</u>. <u>glutinosus</u> in West Virginia. <u>P</u>. <u>kentucki</u> was the predominate appeaser during the study. Because the appeasing salamander will take positions that are

thought to prevent foraging, it was suggested that <u>P</u>. <u>kentucki</u> would be the loser in a competitive interaction that involved food. <u>Plethodon kentucki</u> was the predominate intruder, and <u>P</u>. <u>glutinosus</u> the predominate defender, suggesting a higher degree of territoriality in the latter. This was taken as evidence that <u>P</u>. <u>kentucki</u> is less able to successfully allocate and secure necessary resources in the face of competition with its congener. Thus, it was concluded that competition is playing a key role in limiting the range and distribution of <u>P</u>. <u>kentucki</u> in West Virginia.

<u>Plethodon kentucki</u> and <u>P</u>. <u>glutinosus</u> were found to feed on the same types of prey. Based on this similarity, it was concluded that food is most likely an object for which these species compete. Because head width was not significantly different for the two species, it was suggested that prey size does not partition feeding habits, and most likely increases the intensity of competition for food.

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