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Comparative Ecological, Morphological, and Behavioral Studies of the Southern Blacknose Dace, Rhinichthys Atratulus ObtuStJS Agassiz, and the Eastern Blacknose Dace, Bhinichtbis Atbatulus Atra'fulus (Hermann), in high and low Altitude Streams in West Virginia and Virginia

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COMPARATIVE ECOLOGICAL, MORPHOLOGICAL, AND BEHAVIORAL STUDIES OF THE SOUTHERN BLACKNOSE DACE, RHINICHTHYS ATRATULUS OBTUSStJS AGASSIZ, AND THE EASTERN BLACKNOSE DACE, BHINICHTBIS ATBATULUS ATRA'fULUS (HERMANN), IN HIGH AND LOW ALTITUDE STREAMS IN WEST VIRGINIA AND VIRGINIA

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ABSTRACT

Co•parative ecological, aorphological, and behavioral studies of the southern blaeknose daoe, Rhin1ohthys atratulus obtusus Agassiz, and the eastern blaokaose dace, B.hin1ohtpys atratulus atratulus (Heraann), were made in high and low altitude streaas in West Virginia and Virginia. Statistical co•parisons of the morphology of a. a atratulus and R. a. obtusus showed no reliab1e body character that differentiated the two subspecies. Statistical tests indicated an intergrading population in Gandy Creek, a tributary of the Cheat River, ia West Virginia. This was further substantiated by collecting males of the atratulus phenotype along with males of the native obtusus phenotype in adjacent sections of Gandy Creek. Artificial inseaination of obtusus eggs with atratulus spera provided developing eabryos for one week. Atratulus and obtusus daoe were observed breeding in identical habitats. The ••leading behavior" of obtusus IIIiAles and the aggressiveness of atratulus •ales were the only noted behavioral differences between the two subspecies. Stomach analyses of obtusus and atratulus daee, collected on the saae day fro• ecologically similar streaas, showed that the two subspecies were eating the Sa•• food. A study ot an intergrad1ng population did not show two separate breeding populations and for this reason obtusus and atratulus are believed to be subspecies.

CHAPTEB. I

INTRODUCTION

Determining the eTGlut1onary level of the species or subspecies of the blaeknose dace, Rhinichthys atratulus (Heraann), presents three probleas. One proble•is de-termining if species is a reproductive unit with speo1es being deterxined by the degree of co•patab111ty of the behavioral isolating aechanis•s, or is a taxonoa1o unit determined by the degree of morphological differences between populations, or is an ecological unit with species determined by the habitat and niche oan organis•. Another problea is developing a method whose results can reliably show significant differences or s1 tilarities between populations in teras otaxonomy and ecology. The final problem is determining at what level geographically isolated populations become subspecies or cease to be subspecies and beao•e separate and distinct species.

Based upon the varianae in the breeding colors of blaoknose dace males, four subspecies are now recognized (Traver, 1929 and Hubbs, 19.36). R. a. si•us is a southern subspecies of the Alaba•a River syst••· R. a. obtusus is found in the Cuw.berland, Kentucky, and Ohio River syste:as. . • aeleagris is distributed in the tributaries of the Great Lakes and in the western tributaries of the Ohio River. . &• atratulu;is found in the tributaries of the •ajor eastern drainages.

West Virginia is particularly suited for a study of blacknose dace subspecies. With the possible exception of Pennsylvania, West Virginia 1s the only state which contains three subspecies. B. a. siaus 1s the only subspecies not found in the state. Due to the variety of habitats in West Virginia, it is possible tQ compare high and low altitude and latitudinal effects upon the subspecies.

These populations of blaoknose daces, by def1n1t1on, can be labeled as either species or subspecies. The d1ft1culty lies in determining the degree of difference that delineates between the two levels and whether taxonomic, reproductive, or ecological differences farm isolating mechanisms 1n these species and/or subspecies.

The purposes ot this stud7 are to compare the aer1st1os, morphometry, and breeding behavior of- - obtusus and !• - atratulus found in West Virginia and Virginia 1n high and low altitude streams and to determine, it possible, the proper taxonomic leve1s. Due to its limited habitat and small number of fishes, :fl.• !.• me;t.eagr!s was not included in thia study.

CHAPT:EB. II

REVIEW OP THE LITERATURE

Subspecies of Rhinichth7s atratulus

A review of the literature disclosed a few studies 0n the taxonomy and general ecology t the blaoknese dace, · atratulus. Traver (1929) reported the first study ot the habits of R. atronasus (c R. atratulus) in Caseadilla Creek, Ithaca, New York. Subsequentl7, Hubbs (1936) recognized 4 subspecies or · atratulus, which area atratulus, meleagr1s, obtusus, and simus. Becker (1962) reported the meristios and morphometries of· · meleagr1s in 4 Wisconsin streams. A life history and ecology study or meleagris was made on Elkhorn and Pease Creeks, Boone County, Iawa (Noble, 1964). Shontz (1966) studied atratulus 1n three eastern streams to determine the etteots ot altitude and latitude on body counts and sizes. Tarter (1968, 1969a, 1969b, 1970) reported the age and growth, parasites, reproduction, and food and feeding habits of meleagris from Doe Run, Meade County, Kentucky. Raney (1940a) noted the spawning aot ot meleag;1s from S11ppery Rock Creek, near New Castle, Pennsylvania, and compared 1ts breeding behavior with that ot atratulus. The breeding behavior of obtusus was reported from tributaries ot the Shavers Ferk in West Virginia (Schwartz, 1958).

There has been little werk eenoerning the validity Gf Hubbs original recegn1t1on ef the f ur subspecies. Shentz (1966) reported the lack ef intergrades in a study ef New Yerk streams by M.A.Hal.l. Greeley (19.39) stateda "They have usually been censidered as subspecies but may likely prove to be regarded mere properly as distinct species unless further studies indicate intergradatien."

Species and Subspecies Def1n1t1ens

Many authors, including Mayr (1940, 1966), Mayr, Linsley and Osinger (1953), Trautman (1957)t have defined species and subspecies. Mayr (1940) defined a bieleg1cal species as "greups ef actually er petentially interbreeding natural pepulatiens which are repreduotively 1selated tre• ether such graups.". Mayr (1966) def'ined a subspecies as "an aggregate ef lecal populatiens ef a species, inhabiting a geegraphio sub-d1v1s1en er the range ef the species, and differing taxonem1cally frem ether populationsf the species." Trautman (195?) reported that "taxene•1sts usually consider a subspecies te be valid if *75%* or aere *et* one pepulat1on can be separated stat1st1eal1y frea the ether population." Species has trad1t1enally been defined in taxenomio terms. Mayr (1966) stated that the basic assumptien cenneot1ng "s1gn1f1oant" merpheleg1cal differences with species d1fferent1at1en will eventually cause diserepenoies. He pointed out that many individuals w11l be eonspec1t1c in spite of striking d1fterences 1n structure due t$ age differences, sexual dimorphism, polymorphism, eto. Marr (1955), Barlew (1961), and Shontz (1966) have attempted to determine it mcrphelegioal variation is phenotypic, or genotypic. This issue is greatly clouded by contradictory research. Phillips (1967) showed variance 1m mer1st1es and morphometry when he reperted sexual dimorphism w1th1B a single subspecies ef the blaeknose dace. Shontz (1966) stated "the great variability ot m.er1st1cs and morphometric characters exhibited by the eastern blaoknose 4ace ever its range must be due primarily to genetic isolatien rather than to mere phenotypic response te environmental differences." Barlew (1961), while studying the causes and s1gn1t1cance et merpholegieal variation in fishes, reported that "new ichthyelogists oemmonly assume ditferenoes between populations ea species are environmentally induced unless a genetic base can be demonstrated experimentally."

CBAPTEfl III TAXONOMY AND DISTRIBUTION

Taxone!l

T.he genus Bhinioht9ls Agassiz 1850 includes t1ve re-oegnized species 1n the United States (Blair et al., 1957)J - atratulus (Hermann), the blacknose dace; !• oataraotae (Valenciennes), the l0ngnose daceJevermann1 Snyder, the Umpqua dace, R. faloatus (E1geruu..nn. and E1genmann), the leopard daoe; and R.eseulus (Girard), the speckled dace.

Traver (1929) stated that the great variation in the breeding celor of R.atratulus males led te the naming ot many subspecies. Jordan and Evermanm (1896), descr1b1ag atrenasus (M1tch111) [= &.• atratulus (l!ermannij • wrete•••• "Excessively variable. running into several varieties, the extremes which seem like distinct species." Hubbs (1936) recognized four subspee1est R. a. atratulus (Hermann), the eastern blaeknose dacer!.• meleagris Agassiz, the western blacknose daoe1 - ebtusus Agassiz, the seuthern blaoknese dace, and- - s1mus Garman from Coahulla Creek, Georgia. Hubbs and Lagler (1958) neted that there are the "well-d1ffereat1ated and pessibly spee1t1cally distinct eastern and western terms" R. a. atratu.l.us a:nd a. a. meleagris, respectively. Several peaple (Sh ntz, Bailey, and Tarter, personal cemmunicat1ons) have suspected that the subspecies atratulus and meleagr1s are perhaps different species. Collections at the University ef Michigan Museum ef Zoelogy turned up only a slagle, small, p0orly preserved sample (from West Virginia) labeled intergrade between atratulus and meleagris (Bailey, personal eemaun1cat1en).

Shontz (1962) described the subspecies et R. atratulus aeo rding to male breeding eeler which is the only totally reliable method of 1dent1f1oat1ctn. "The males ef R. a. atratulus have brilliantly eolered peot0ral tins ranging frem orange-red te scarlet. It is knewn lecall7 in parts Gf West Virginia as the ••red-finned :minnew". The males of R. a. aeleagris and ebtusus have little er ne color en the pectoral fins but instead have a lateral band ef orange to red. In meleagris the color is cenfined to the immediate area ef the lateral line while 1n ebtusus the celor extends well dewn the sides 1n many oases te the peint ef insertion of the pelvic tins and alae ferward acress the $peroulum te and sometimes beyond the regien under the eye. A further d1st1nct1en is evideDt between the breeding males of the latter two subspecies; the pattern et tubercles c&apletely covers the tep of the head in meleagr1s while 1n ebtusus the tubercles are confined mestly to the sides of the head. The·males ot Ro a. s1mus have much deeper bodies and shew little er no breeding color,.

For many years it was reported that M1tch1ll (1815) first described the blaeknose dace as Cyprinus atr nasus, Sterer

(18.39)repE)rted it :tr•• Massachusetts as Leu.o1sous atrenasus. Agassiz (1850) indicated that Leucisous atronasus Sterer and Clprinus atronasus Mitohill sh0uld be referred te the genus Bh1n1ohthys. Hubbs (1936) reperted that his attentien had been called to a rarely used beok by Jehannes Hermann, published in 1804. that contained the eriginal deser.1pt1on of the blacknese daoe as Czpr1nus atratulus. Hubbs validated the use ef atratu1us since Czpr1aus atratulus Hermann 1804 clearly antedated Clpr1nus atrenasus Mitehill 181,5, and re-named the blacknose dace Bh1n1ohth:rs atratulus (Hermann).

The tellew1ng description e:t the blaokn0se daee, !• atratulus 1s taken frem Blair et al., (1957)*

"D.8; A. ?. Scales in L.l.abeut 53; betere D. crowded and embedded, abeut)lJ scale radii present in all fields. Meuth tera1nal, net everhung by sneut. Many scales selid black; lateral band indistinct er absent. C. spet poorly develeped. Premaxillae not protractile, frenUJD broad, Teeth 1,4--4,1.••

Range

The genus llh1n1ohthys oceurs in all parts et the United States except in the extreme s utheastern portien (Blair et al., 19.57). !• atratulus ranges frem ••the Dakotas eastward through the Great Lakes region to the Atlantic Coast and southward en both slepes ef the Appalachians to Georgia, Alabama, and Mississippi." Coker (1927) reperted it frem

Nerth Carolina. R. qataractae is "widely- distributed troll. coast te eoast in the Nerth, seuth to Nerth Carolina and Iewa, and in the West te nerthsrn Mexico." R. evermann1 ecours in the Umpqua B.iver of' Oregen. R. taloatus is teund in the Celumbia B1ver Basin east et the Cascades. R. eseulus is "widely distributed frea the western slopes et the Reeky Mountains to the West Ceast, from the Celumb1a B1ver system south te the Celorado River system 1Ar1ZQ a, New Mexice, and Senera, Me:x:ice."

Hubbs and Lagler (1958) reperted the ranges fer the four subspecies asa "Bhinichthrs atratulus atratulus (Hermann) -- Frem the eastern end ef the Lake Ontar1e basin and the St. Lawrence River drainage et Quebec, and from Nova Soetia and New Brunswick seuthward, east of the Appalachian Divide, te the Rean•ke watershed in Virginia (kneWB en the western slope only in the headwaters at the Yeughiegheny in West Virginia)•"

"Bh1n1chtbzs atratulus •eleaeis Agassiz -- Frem nertheastern Nebraska. Iewa, Nerth Dak0ta, the drainage ef Lake W1nn1pegesis in Manit@ba, and the Lake f the Woods region through the entire Great Lakes Basin (except about the east end et Lake Ontar1e) te tne northend part et the Ohio R1ver system.••

"Bh1n1ehthys atratulus obtusus Agassiz -- The Tennessee River System...

"B.h1n1ehthrs atratulus s1m.us Garm.en -- The Alaba & River System."

The range •t the three subspecies 1n West Virginia has been reperted by Raney (1947) (Figure 1)a "-atratulus, Petomae River and James River drainages in small streams; -- meleagris, some streams of the extreae northern part ef state {Menongahela River system) and probably intergrading southward with- ebtusus1 and-- ebtusus, probably mest oemmen subspecies in state west ef the mountains in the Ohio Biver drainage system." Schwartz (persena1 oemmunicatien) found meleagr1s in those tributaries ef the Menengahela River abeve White's Day Creek, Shentz (1962) reported atratulus 1n the headwaters ef the Cheat River.

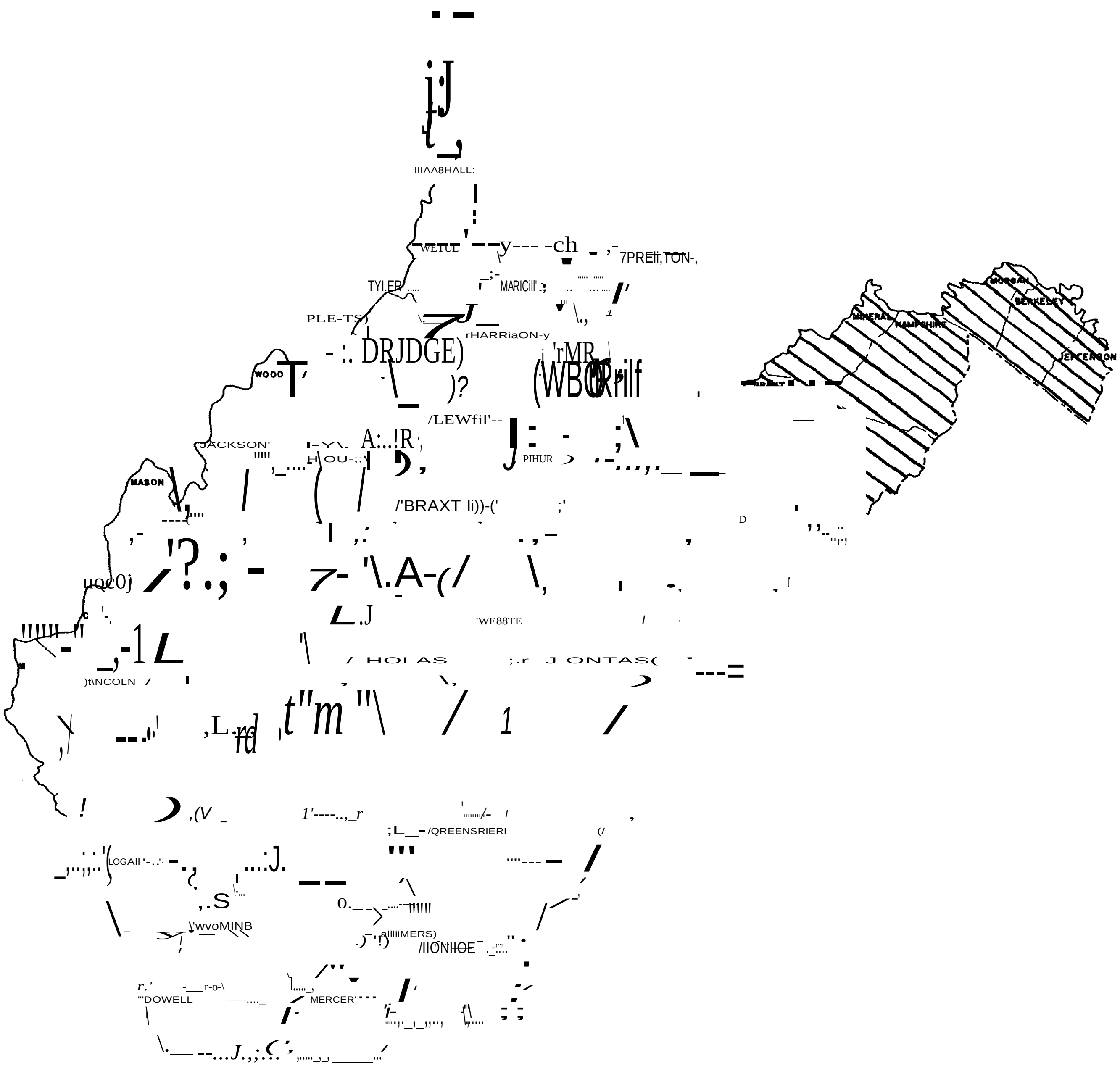
Habitat

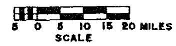
R. atratulus usually is abundant in clear broeks and streams, especially 1n the headwater pert1ens, and usually are rare in lakes and pends threugheut 1ts nerll&l range. In New Hampshire, Bailey and Oliver (1936) noted that "It is primarily a streaa species and abeunds in most small breeks, but is alse eema0n in mest larger streams except the Cenneoticut River." In Oh1e, Trautman (1957) reperted that it is "An inhabitant ef m.ederate and high-gradient breeks whese waters are clear except for brief periods, which had a permanent flew, sand and gravel bettems. well-defined riffles fer spawning purpeses, peels oenta1n1ng deep holes, undercut banks, brush and reets fer satety trem eneaies, and shade during much ef the day." Harlan and Speaker (1956) reported the blacknese dace as "A resident -f small, clear-water creeks," that "reaches its greatest abundance 1n the trout streams ef

FigUre 1.Range ef Rh1n1chthys atratulus·atratulus ;

!• !.• meleagr1s =E, , and i.• !:.• •btusus • D in

West Virginia.


MORGAN
BERKELEY
JEFFERSON
MINERAL
HAMPSHIRE
WOOD
MASON
JACKSON
MERCER


5 0 5 10 15 20 MILES
SCALE

northeast Iowa."

There are two suitable types of habitats fer blacknese dace in West Virginia. At higher elevat1ens 1•the state (particularly in the easter.n meunta1ns), the dace eccupy aat enly the tributaries et river systems but in Cranberry River, New River, and North Ferk ef the South Branch ef the Petemao and the larger tributaries ef the Cheat, dace alse may be feund in. the main r1vers. Temperature, turbidt!y, and velecity seem to be the env1renmental tacters that are the mest limiting of the habitat of blaokmose dace.

The charaeter1st1cs ot streams centa1n1ng - ebtusus are diverse. Addair (1944) reported B. a. Gbtusus in the New R1ver near Hinten, West Virginia. New B1ver 1s a wide, rooky stream at this peint with shallew water and long riffles. It has a summer lew that exceeds 4000 cfs. This contrasts sharply with the dace habitat in the tributaries $f the Oh1e River, Twelvepele Creek, and Big Sandy River in southern West Virginia. In these streams. R. a. 0btusus are found only in headwater seot1ens of streams with small riffles and sandy bottem pools. In the meuntain tributaries of the Gauley and Cheat River system, dace are feund in streams et all sizes until the stream has made a drep 1B elevation significant enQUgb to increase the temperature bey$nd their teleranee level. At this pe1mt, blaoknose dace pepulat1ens are restriete.d to tributaries. R. a. •btusus eelleoted frem. high elevation streams in the eastern meuntains ef West

Virginia are more similar in habitat to high altitude R.a.atratulus pepulatiens than to low altitude R.a.ebtusus pepulat1ens. In West Virginia, there 1s no difference in the habitat preference of the subspecies et blaokn&se daoe. There is, however, an extreme differenoe between the habitats ef high altitude and lew altitude pepulatiens et the same sub-species.

CHAPTER IV

DESCRIPTION OF THE STUDY AREA

The twe aeunta1n streams, Gandy and Seneca Creeks. are native breek treut streams with leng shallew riffles, At an altitude of 3,250 feet the streams are almest identical in size. Beth streams are generally less thaD 20 teet acress at this altitude.

_There are twe netable differences between Gandy and Seneca Creeks. Pe•ls •n Gandy Creek are frequent, leng₁ deep.and etten silt lined. Peels en Seneca Creek are oem-parat1vely infrequent and shallower than th0se en Gandy Creek, and in mest cases lined with reck. The deepest pool saapled en Gandy Creek exceeded 6 feet in depth and several wera feund in excess et 5 teet, No peel sampled en Seneca Creek exceeded 4 feet in depth. Marl depes1ts were much mGre frequent and ef a greater depth in Gandy Creek.

The twe streams run a parallel oeurse fGr some distance and the tributaries of beth drain the slepes ef Allegheny mountain. Allegheny meuntain is pessibly the lengest C$nt1nual meuntain in West Virginia (Sisler, 1931 and White, 1927) and fer its entire length foras the Allegheny front, the division between Petemac and Ohie River tributaries. The highest peint on Allegheny mountain in Randelph Count7 is 4,760 teet. The steeper gradient, eastern slope is drained by Seneca Creek and Big Run ef the Nerth Ferk River. The average gradient ef Seneca Creek is 132 feet per mile and the stream drains 68.37 square m11es; it is 19.3 miles lang. The aeuth •t White•s Run was the first oe1lect1ng statien and it has an altitude of 2,159 feet. R.a.atratulus was oelleoted at 5 statiens en Seneea Creek in a three quarter mile sect1en er stream upstream frem White's Run. Blacknese dace were oellected frem Big Run ef the North Ferk during the breeding seasen in an attempt fe fimd 1ntergrades er 1ndicatiens ef the obtusus phenetype. Ce leotie:ns o:r atratulus were made frem the meuth ef Big Rum te the cenfluence ef Healeek Run and Teeter CaJD.p Run. Big Run was retenoned by the West Virginia Department of Natural Reseuroes in 1962 and ne dace were feund in the upper seet1ens ef the stream. Big Run is smaller than either Seneca er Gandy Creek. The average gradient er Big Run is 134 feet per mile and it is 12.8 miles len.g,

The lowest eellectien statien on Gandy Creek was 2 miles above Whitmer, West Virginia. It was at this collecting stat1en that ebtusus breeding males were feund, The altitude at this atat1en was 2,890 feet. The next celleoting statien was the meuth ef Big Run of Gandy at an altitude of J,22S teet. The first atratulus males were feund at the meuth er Grant's Braneh at an altitude et),380 teet, The ether nine oelleot1ng statiens were unmarked peels between these twe tributaries. They were ehesen by randem sampling ef peels accessible te the read. Gandy Creek is 18.6 miles leng and drains 4).04 square miles. It has a gradient ef 66.12 feet per mile which 1s almest half ef that ef Seneea Creek.

Back Creek ef the Beaneke River is a tributary seuth ef Reaneke, Virginia. The altitude at this eellectimg stat1en was 1,100 feet. Back Creek was chosen because it was at a similar altitude and lat1tuda to Little Cabell Creek se that the twe lew altitude samples would be environmentally similar. Back Creek had apprex1ma.te1y three times the flew ef Little Cabell and a much denser dace pepulatiem. It was an extreaely shal1ew stream with a maxiaum width of 20 feet on 28 April 1972. Ne pools were feund in excess ef three feet in depth, but seme riffles were JO feet er lenger.

Little Cabell Creek is a small tributary *et* the Mud River near Milten, West Virginia. At the point ef oelleetien it is 700 feet in altitude and trem the beginning *et* the b1aeknese dace habitat te a p•1nt ene mile upstreaa, it has a gradient ef *35* feet per mile, In the stream seetien sampled n•p•ol was mere than 6 feet in width and n•riffle mere than 10 feet in length. The maximum depth ef any peol was 3 teet. The stream did net maintain a sizeable dace pepulat1en and the individuals were smaller than these ef any ether stream saap1ed. The peels were ••stly sandy bettem with small sandstone r•oks 1n the riffles.

CHAPTER V

MATERIALS AND METHODS

Fish Ce1lect1ens

Dace were collected by seines frem 6 to 20 teet in length. Oocasienally, they were oelleoted by a Smith-Roet Type V Electreshecker. Cellectiens were made during the breeding seasen se that sex ceuld be detera1ned by the presence er absence ef breeding colors.

Seale and Fin Raz Ceunts

Al1 c•unts were made under a binocular micrescepe and fellewed in general the methods described by Hubbs and Lagler (1958).

Lateral line scales. Only scales haVing peres in the lateral 11ne were eeunted, The first scale oeunted was the first pored scale separated fr••the sheulder girdle. The last scale eeunted was the last scale eenta1n1ng a pere, even it found •n the caudal tin base. If s••• lateral line scales were absent, then a ceunt was aade •n the next rew d•rsally.

Pecteral. anal, E•lv1c !!.!!.rals• All tin ray c•unts were made en the left side unless the left fin was daaaged. The anal fin was lifted 1nte aa erect and exteDded pes1t1on by ferceps. All rays were c•u.nted. These rays with branches were counted as one ray.

Dersal !J:E ra:. With •ne exeept1an, the dersal fin rays were ceunted in a similar methed te all other fin rays. As suggested by Hubbs and·Lagler (1958), the last twe rays were eeunted as ene if neither were branched. In the Ba.ek Creek co11eet1ens s••e daee appeared to have ten dersal fin rays, Close examination indicated that neither the ninth er tenth rays were branched. while en ·t1sh having ettly nine dorsa1 rays the ninth ray was deeply branched. Fer this reason, the ninth and tenth dersal rays were counted as ene.

Linear Measureaents

All measurememts were made with a dial vernier caliper te the :n.u.rest hundredth •f a a1111meter. Most aeasureae:mts were aade accerd1ng te Hubbs and Lagler (1958).

Tetal length. The greatest distance fr••the sneut t• the tip ef the caudal tin when the fin 1s squeezed tegether.

Standard lepgth. The greatest distance fr••the tip ef the snout to the base et the vertebral oelUIJ.D.. The base ef the vertebral oelumm was deterained by bending the caudal f1•imaediately back ef the vertebral celuan.

length. The distance fr••the snout tip te the pesterier mest sect1en et the eperculua.

S:n•ut length. The distance frem. the sneut tip to the .fr•Rtal margin ef' the erbit.

Head width. The distanee aeress the preeperele.

depth. An eblique measuremeat frea the eec1put t• the isthmus.

Gape. The largest d1soernable distance tr•• the euter aargin ef the mandibles.

Length !!erbit. The distance between the rims ef the erbit along the lengitudinal bedy plane.

Upper jaw length. The distance frea the pesterier ef the aaxillary te the tip •f the sneut.

Predersal distance. The distance frea the base ef the first dersal ray to the tip et the sneut.

length. The distance frea the erigin ef the first ray te the tip ef the lengest ra7 in the anal, peot•ral, pelvic, and dersal fins.

Statistical Analysis

T•determine the similarity of twe subspecies ef blaekn•se dace frem similar latitudes and altitudes, the t-test was used en pepulatiens of)0 fish oemsisting ef lS males and 15 females. The mean, standard deviation, and standard error ef the mean were determined fer each count and measure-•ent. Due t• the similarities ef env1remaents during embryonic develepment •f these twe subspecies, any mean difference must be equated te genetic oauses.

Befere testing fer the significant level ef the linear measurements, each bedy part was divided by the standard length. Significant d1tferenoe was cemputed en the gape, head width, head depth, head and sneut lengths, predersal distance, and pectoral and pelvic tin lengths by the t-test. The t-test was alse used te compute significant difference of the lateral line number, and peoteral and pelvic fin ray number.

As a eentrel, a high altitude sample ef a subspecies was then cheeked fer significant difference with a lew altitude sample ef the same subspecies. The lew altitude sample ef R. ao ebtusus and atratulus were from similar altitudes and latitudes. The lew altitude R. a. atratulus sample came from Back Creek, a tributary et the Rean•ke River. The lew altitude sample of R. a. ebtusus was oelleoted fDDm Cabell Creek, a tributary of the Mud River, The t-test was eapleyed te determine any significant difference.

The use ef oentrels was essential se that any variance feund could net be attributed te ohanoe. If a high altitude sample ef ene subspecies 1s significantly different frem a high altitude sample ef anether subspecies, this difference ceuld be attributed te chance er the null hypothesis. If, hewever, the same bedy character is eempared t• a sample ef the same subspecies frea a different environment and net feund te be significantly different, then the difference between the subspecies is net due te ehance er environment but 1s due te genetic 1selat1en.

Feed Habits

Fish celleetiens were taken trea Gandy Creek aad Seneca

Creek, !• - ebtusus and atratulus habitats, respectively, en) June 1972. The steaaeh oentents ef 20 speeiaens et each subspecies were anal7Zed and ceapared. The c•lleetiens were made within aR heur ef each ether. The subspecies were feu.d 1m identical seet1ens ef the tw•strea•s amd benthic saaples sh•wed the two streams te be ecelegioally identical.

Field Observations

Breeding behavier. R. a. ebtusus was ebserved breed1 o- in the Cranberry River amd Gaady Creek.

- - atratulus was ebserved breeding in Seneca Creek.

Artificial 1Bsea1Bat1en. Om 28 April 1972, breeding R.!.:.. a..._tr...,al.. ,l·.l·.s-. aales were cellected from. Back Creek, a tributary ef the Reaneke River. OR 3 May 1972, spera tr•• these males was used te fertilize R, a. ebtusus feaales eelleoted trem Little Cabell Creek, a tributary ef the Mud River. The eggs were then plaeed in a sheath ef cheese cloth and returned te the streaa. They were taken tr••the streaa eD 10 May 19?2, and placed in a lab•rater1 aquar1ua.

Laberatery Observati•!

Breeding behavier. In twe 20 gallen aquaria, saud-stene reeks were arranged in a oireular pattern. The 60 te 80 . . epen spaces were filled with sand aad erganie debris. Om•large reck was prepped up se that aales and feaales aight aeve under reeks dur1Dg spawning. The pheteper1ed was controlled by an autoaatio tiaer and set to the ti•s of sunrise and sunset on 11 June. The te•perature was constantly •aintained at 74^{o} F. In one aquariua, 6 aale R. a. atratulus and 2 fe11ale !•!-• obtusus were placed, In the second aquar1ua, 6 aale R, a. obtusus were placed with 2 fe•ale - a.atratulus. The fish were observed fro• the sixth hour of daylight to the tenth hour on the first day and fro•the sixth hour to the eighth hour for the next 6 days. After ? days all fish were re•oved fro•the tanks to prevent the consuaption of eggs.

Artificial inse•ination. After 7 days of observations, spera and eggs were stripped fro•the Bales and fe»ales in each tank and aixed i:n a watch glass. They were kept in the watch glass for 30 ainutes and then returned to the aquariua fro•which the fish had co••·

CHAPrER VI.

RESULTS AND DISCUSSION

Statistical Analysis

The collection from Gandy Creek on 27 May 19?2 contained red-finned breeding males in a stream native to males without fin breeding color. If the Gandy and Seneca atratulus populations are identical, a t-test tor significant difference should be negative or at least negative to the degree of a control comparison. As a control the Seneoa Creek sample or atratulus was compared statistically with a low altitude atratu1us population. If the two subspecies have 1ntergraded, a t-test should indicate some significant differences and soae similarities between the Seneca Creek population and the Gandy Creek population, as the Gandy Creek population would be a mixture of two gene pools. It is possible that sia1lar1t1es between Seneoa Creek and Gandy Creek dace populations are due to the environment of the developing embryo. As a control to determine the erreot of similar enVironment upon atratulus and obtusus populations, a t-test was run between atratulus from Back creek and obtusus from Little Cabell. As previously mentioned these streaas are nearly identical in altitudes and latitudes.

The Back and Little Cabell Creek controls indicated that snout length, head width, head l.ength, lateral line scales, and predorsal distance were significantly d1f£erent at the 1% level (Table 1). Sinoe the env1ronm.ent was very similar during the embryonic development of these fish, the significant differences can be assu:aed to be genetic in origin. Therefore, it can be assumed that members of the same subspecies (having originally developed trom the same gene pool) should be similar and not significantly different in these measurements and oounts.

Such was not the case when the Seneca Creek sample was compared to the Back Creek saaple. These two populations were found to be s1gn1f1oantly different at the 1,% level 1n head length, snout length, head width, head depth, gape, pectora1 fin 1ength, lateral line scales, and pelvic fin rays (Table 2). Shontz (1966) has stated that scale counts and tin ray counts are due to environmental effects, however, in this test all measureaents except predorsal length, pelvic fin length, and pelvic fin rays were significantly different when the genetic origin was the saae and the env1ronaents diverse. There are two possible explanations. Through the null hypothesis these differences can be attributed to chance s8JI.pl1ng. The other explanation is that these two atratulus populations have been genetically isolated long enough for mutations to make the two populations aorpholog1cally divergent.

The Seneca population was not significantly different

Table 1. Statistical analyses on the aer1st1os and •or-pho•etry of R. a. atratulus and R. a. obtusus collected from Back Creek, Virginia (Station I) and Little Cabell Creek, West Virginia (Station II), respectively. N = .30.

Character	Station	S.D.	S.E.$_{\bar{x}}$	$\bar{X}$	t
Head Len	I	0,012	0.002	0.288	2.86 **
	II	0.010	0.002	0.Zao	
Snout Len	I	0,00?	0.001	0.109	S.00 **
	II	0.00.S	0.001	0.101	
Read Wid	I	0.012	0.002	0,164	;.0S **
	II	0.00a	0.00Z	0.1.56	
Head Dep	I	0.009	0,002	0.162	1.82
	II	0.00a	0.001	0.158	
Gape	I	0.007	0.001	0.0?J	
	II	0.00S	0.001	0.072	0.67
Pre Dors D1st	I	0.015	0.00;	0.5?6	:).24 **
	II	0.0 3	0.002	o.;a8	
Pel Fin Len	I	0.013	0.00S	0.154	1•.56
	II	0,012	0.002	0,159	
Peo Fin Len	I	0.014	0.006	0.190	0.00
	II	0.01a	0,003	0.190	
Lat Line Sols	I	2.)81	0.43S	;6.20	2.15 *
	II	2.893	Θ•.528	54.73	
Pel Fin Rays	I	0.183	0.033	8,0)	1.01
	II	0.516	0.133	?.9)	
Pee Fin Rays	I	0.707	0.129	14.2.3	0,?)
	II	0.730	0.094	14.1:3	

1 1ghly Significant (1% level of confidence)
* Significant (5% level of confidence)

Table 2. Statistical analyses on the mer1st1cs and morphom.etry of R. a. atratulus and R. a. at:ra.tulus eolleoted from Back Creek, Virginia (Station I) and Seneca Creek, West Virginia (Station II), respectively. N = 30.

Character	Station	S.D.	$S.E._{\bar{x}}$	$\bar{x}$	t
Head Len	I	0.012	0.002	0.288	9.26 **
	II	0.009	0.002	0.263	
Snout Len	I	0.007	0.001	0.109	(.22 **
	II	0.007	0.001	0.096	
Head Wid	I	0.012	0.002	0.164	10.38 **
	II	0.007	0.001	0.147	
Head Dep	I	0,009	0.002	0.162	5.26 **
	II	0.00S	0,001	0,152	
Gape	I	0.007	0.001	0.073	4.?4 **
	II	0.00S	0.001	0.064	
Pre Dors Dist	I	0.015	0.003	0.,576	0.714
	II	0.018	0.003	0•.579	
Pel Fin Len	I	o.o13	0.00S	0.154	0.00
	II	0.010	0.001	0.154	
Pee Fin Len	I	0.014	0.006	0.190	2.97 **
	II	0.014	0.003	0.201	
La.t Line Sols	I	2.)81	0.43S	,56.20	6.59 **
	II	2,665	0.497	60.S0	
Pel Fin Rays	I	0.183	0.0 ;	a.0;	1..72
	II	0.2.58	0.0 7	?.93	
Peo Fin Rays	I	0.707	0.129	14.23	4.11 **
	II	0.?07	0.129	14.97	

** Highly Significant (1% level of confidence)
* S1gn1t1oant (5% level of confidence)

from the Gandy Creek population in snout length, head width, gape, pectoral fin length, latera1 line scales, pelvic tin rays at the 1% level, and not significantly different from head length at the S% level (Table 3). This deaonstrates a greater s1B1larity between these two populations than between populations ot the saae subspecies. The lack of difference between the scale count and tin ray counts is due to the s1•1lar1ty in environment. The other similarities are due either to mutation or due to interbreeding. The Seneca Creek population has been isolated trom both Baok and Gandy Creeks, but due to its proximity to Gandy Creek there is a chance of streaa capture causing 1ntergradat1on. The similarities of the Gandy and Seneca populations, when populations of the same subspecies are al•ost totaly diverse, suggests that 1ntergradation has occurred.

Food Habits

The stomach analyses of the two dace populations, showed marked similarity in diet (Table 4). In both streams the most comaon food was the mayfly genus Stenonema and the chironom1d dipterans. The greater nwaber of Stenoneaa that were consu:aed in Seneoa Creek would be expected since they are riffle aay-flies that have dorso-ventral1y compressed bodies which offer little resistance to water flow. The gradient of Seneca Creek· is nearly twice that of Gandy Creek. This causes a larger proportion of Seneca Creek to be in riffles and there-

Table 3. Statistical analyses on the mer1st1cs and morphometry of · · atratulus and !• · obtusus collected from Seneca Creek, West Virginia (Station I) and Gandy Creek, West Virginia (Station II), respectively. N = ;0.

Character	Station	S.D.	S.E.$_{\bar{x}}$	$\bar{X}$	t
Head Len	I	0.009	0.002	0.26J	2.06 *
	II	0,010	0.002	0.268	
Snout Len	I	0.007	0.001	0.096	1.59
	II	0.00a	0.001	0.099	
Head Wid	I	0.007	0.001	0.147	l.6J
	II	0.007	0.001	0.150	
Head Dep	I	0.00S	0.001	0.152	,3.20 **
	II	0.007	0.001	0.157	
Gape	I	0.00S	0.001	0.064	0.00
	II	0.006	0.001	0.064	
Pre Dora Dist	I	0.018	0.00;	0.579	).09 **
	II	0.01.5	0.003	0.592	
Pel Fin Len	I	0.010	0.002	0.154	4.JJ **
	II	0.010	0.002	0.165	
Peo Fin Len	I	0.014	0.003	0.201	1.17
	II	0.012	0.002	0.205	
Lat L1ne Sols	I	2.66.5	0.487	60.50	0.90
	II	).082	0.563	61.17	
Pel Fin Rays	I	0.2.58	0.04?	7.93	0.909
	II	0.258	0.047	8.07	
Pee Fin Rays	I	0.707	0.129	14.97	0.793
	II	0.658	0.120	14.83	

** Highlr S1 1f1cant (1% level of confidence)•
* Significant (5% level of confidence)

Table 4. Stomaoh analyses o20- - atratulus and 20 !• - obtusus collected from Seneca Creek, West Virginia and Gandy Creek, West Virginia, respectively.

Taxon	f!.• !.• atratulus	R. a. obtusus
EPHEMEB.OPrEBA		
Stenone .a sp.	20	10
Unidentified	2	1
DIPrEBA		
Limnophila sp.	0	4
Chironomus sp. {larva ana pupae)	18	12
Unidentified	1	0
TRICHOPTEBA		
Hydropsyche sp.	6	6

t

fore increases the habitat of Stenoneaa, Most of the females fro•the Gandy Creek saaple oaae troa one large wel shaded pool, which is the habitat of these oh1ronom1ds and of the tipulid genus Limnophila. A comparison of benthic saaples from the two streaas showed a greater number of t1pul1ds in Gandy Creek than in Seneca Creek. This is due to the abundance of deep pools 1n Gandy Creek.

Field Observations

Breeding behavior. B. a. obtusus and atratulus were observed spawning in the Cranberry Biver on 28 May 1971 and in Seneca Creek on 3 June 1971, respectively. Field notes were taken and a ooaparison of breeding behavior was .ade.

As reported by' Traver (1929)• the atratulus .ales were somewhat aggressive and territorial. As many as S males were observed at one spawning site. Males not onl7 responded to females around the nest but would leave the nest to chase te•ales. Males would sw1•beside other aales tor short distances in an abbreviated chase. The males were not observed biting or damaging other males. The males were aggressive but were not aggressive to the degree observed in puapkinseed sunt1sh. Males did not respond aggressively to other males when females were present. Feaales were observed aov1ng through a spawning area containing four spawning territories followed by .a1es in small groups of four or 1ess. A spawning site consisted of a sandy space 60 to 80 . . in diameter fringed by roeks. The water was 8 to 12 inches in depth, and the site was exposed to the sun at the time of spawning. Males did not remain at one site for a prolonged period ot time and several aales were seen spawning at one site over a two hour period. The breeding activities were •ore intense at 1.300 hours. The breeding site was the shallow margin of the middle of a poo .

The spawning behavior of obtusus in the Cranberry Biver was similar to that reported by Sohwartz (1958), with the exception that male obtusus followed females in clusters of four or less. As reported by Schwartz (1958), they did not show any signs of territorality or aggressiveness. However, they did not differ in habitat from atratulus as reported by Schwartz (19.58)• Males swa.11 erratica.l.ly after females and showed no preference in spawning s1te. The "leading« behavior of male obtusus characterized by Schwartz (1958) was observed. The spawning site was the shallow margin of the middle of a pool.

The spawning pools of atratulus and obtusus were identical, as were the 1nd1v1dua1 spawning sites within the pools. The only observed differences in breeding behavior were the ..lead1ng[11] behavior of male obtusus and the terri- torality and aggressiveness of aale atratulus

If stream capture occurred, and atratulus and obtusus were interbreeding in the same pool, competition between the males of the two subspecies would not affect spawning behavior between •ales and females. With the exception of the ..l.ea.d1ng" behavior of •a1e obtusus, the behavior pattern between male and females of atratulus and obtusus were identical. This was the only evident isolating •echan1sa that •1ght stop the interbreeding of the two subspecies.

Laboratory Observations

Breeding behavior. There was no indication of breeding behavior or spawning in the Bixed populations of obtusus and atratulus in the laboratory aquaria. Also, no breeding behavior or spawning was observed in the control aquariua of obtusus. At the end of three weeks there were no fry in the aquaria, either fro•natural reproduction or art1t1c1al insemination. The obtusus ova that had undergone artificial insemination and then had been placed in Little Cabell Creek developed or one week. Developing ova were present on the cheese cloth at the time of their transfer to the aquar1ua. No ova survived the first night in the aquarium.

There were several env1ronaental factors which could not be duplicated in the l.a.boratory. Although the dace spawned in small areas of the stream, they swaa freely in large pools. In the aquaria they were not as free to swia. Dace 1n the stream retreated to deeper pools in the evening and did not move into sha1low breeding pools until approx-imately 1200 to lJOO hours. Variations in water teaperature or light intensity could be the stimulus tor this aoveaent.

In the aquaria there was no variation 1n temperature and light intensity. There was very little milt present 1n the .ales removed from the aquaria and stripped for artificial insemination. It is possible that those fish used for the laboratory experiment had exhausted their supply of milt prior to capture and therefore showed no interest in feaales. It was understood prior to the experiment that the sample was probably 1nsuff1c1ent and the environment impossible to duplicate in every detail. It was known before hand that the null hypothesis would cancel out the s1gn1fioanee of any negative results. The only stated eonolusion is that "these" daoe did not spawn in aquaria and artificial insemination involving "these•• dace was unsuccessful. It cannot be oon- eluded that the lack of suocess in these experiments means that atratulus and obtusus will not interbreed in other circumstances. This is substantiated by the interbreeding of Bhin1chthzs cataraotae, the longnose dace, and Nooomis m1oropogon, a chub, in the Cheat River, Randolph County, West Virginia (Raney, 1940b).

R.a.atratulus sperm did fertilize and initiate development in the- -obtusus eggs following artificial insemination and subsequent placement of fertilized eggs in Little Cabell Creek. This fertilization supports the above stated belief that the sperm of one subspecies and the ova of another are oompatable. The ova did not cease to develop until they were removed from the stream and placed 1n a laborator¥ aquariua. The death of the embryos was probably due to variations 1n temperature and oxygen levels in the aquarium.

CHAPTER VII

CONCLUSIONS

The scientific method should be used to determine the degree of taxonomic, ecological, and reproductive differences between subspecies of blacknose dace. It is not safe, however, to assume that results from the scientific method when applied to taxonomic differences indicate a similar change in reproductive behavior, or niche and habitat. The aorphological variance between the subspecies of blaoknose dace can be attributed to genetic or environmental causes, but these m.orpholog1cal differences cannot determine the phylogenetic level of blaeknose dace populations. The degree of difference between these populations can be compared to the degree of difference between accepted separate species ot the genus Rh1n1ehthys. This comparison is an indication of the proper taxonomic level, but it is not completely oompatable to the bio1og1cal species concept, and therefore must be considered inconclusive.

If the standards of the taxonomic species were used to determine the phylogenetic level of the blaoknose dace, a significant problea would be encountered. Taxonomically any group of organisms that continually produce offspring morphologically different from all similar populations may be considered a separate species. Blaoknose dace demonstrate significant sexual dimorphism (Phillips, 196? and Tarter, 1969b). Males and females can be separated in over 97 percent of the fish of a population (Beeker, 1962). It is, however, 1•possible to reliably determine the proper subspecies using body characters. Taxonomically all males.irregardless of subspee1es, would be in one species, and all females in another species. Obviously, this cannot be the situation.

All fishes that are characterized as being separate and distinct species have some morphological character not evident in any other species. Without this distinct character the keying of an individual fish would be impossible. No such character exists to separate the two tested subspecies of blaoknose dace. Even though populations of blaoknose dace can be identified statistically this is not a sufficient difference to classify them as separate species. It would be 1noons1stant to be able to differentiate all other members of genus Hhinichthys through a single trait which is oon-sistant on all individuals of a species, and then have to separate a group of species through regression statistics. R. oataraotae (longnose dace)can be morphologically distinguished from both subspecies of blacknose dace. The lack of a differentiating body character indicates that obtusus and atratulus are still members of the same species.

There is no evidence to indicate the subspecies vary 1n habitat or niche. There is some variance in habitat within a subspecies due to changes in altitude and a corresponding change in stream character. !• · atratulus and obtusus live in similar sections of streams with nearly identical gradient and temperature ranges. They both demonstrate the same feeding behavior. Both subspecies have been observed in moderately fast water facing the current in parallel rows and eeding off of the water flow. Fish of the two subspecies collected on the same day from identical streams c.ontained s1m11ar stomach contents.

The isolating mechan1s•of reproductive behavior Or atratulus and obtusus have been reported by Raney (1940a) and Schwartz (1958), respectively. This investigator has observed both of the subspecies reproducing in two streams. The males *ot* both subspecies grouped together and swam wildly after fema1es: Males of both subspecies appeared to have difficulty determining species and sex, They were observed courting other males even though the males were brightly marked.

The breeding habitat, both in character of the pool and the nature of the spawning site in the pool, for reproduction was identical for both subspecies. The type of bottom, velocity of the water, and water depth were the same. There was no habitat characteristic that separated the t subspecies in breeding location.

The lack of a behavioral trait as an isolating mechanism and the similarity of breeding location suggests that atratulus and obt<u>usu</u>s would definitely breed in the same pools of a stream where intergra.dation could ooeur. It is probable that the two subspecies would interbreed due to the similarity of breeding behavior and the absence ot strong isolating mechanisms. Therefore, it is unlikely that the two subspecies are sibling species.

Although there is insufficient data in this paper to make a definite state•ent concerning the taxonomic level of the 2 phenotypic groupings of blacknose, there are indications that both groups are members of the same species.

Shontz (1966) reported - atratulus in the Cheat River system, normally a R. a. obtusus stream. Ross and Perkins (1959), in collections on Sinking Creek, a tributary ot the New River system in Virginia, found red finned male blacknose daoe (R. a atratulus) in a stream native to R. a. obtusus. Possibly stream capture occurred nearby, involving Sinking Creek of the New River and Meadow Creek of the James drainage, Craig County, Virginia (Jenkins, personal coJDunication). Schwartz (personal communication} indicated that R. a. atratulus has been collected in the New River near Hinton, West Virginia, which is R. a. obtusus habitat. He believes these fish have been transported by fishermen from tributaries of the James River in Virginia. Shontz (1962) reported obtusus from the Roanoke River, which is an atratulus stream. Jenkins (personal communication) noted that Greeley, in a New York State Stream Survey Report, found atratulus in a meleasris stream in the southwest Lake Ontario drainage, without apparent intergrades.

Three colle tions, over a period of 2 years fro•Big Run of the North Fork and Seneca Creek, yielded 98 •ales showing breeding coloration. All of these fishes had the red in characteristic of atratulus (the native subspecies). There was no evidence of 1ntergradation. On 3 June 1971, 3 obtusus •ales were collected fro•Gandy Creek with the nor.aal breeding coloration. On 5 July 19?1, 6 blacknose dace •ales were collected, and J of these •ales had breeding color in the pectoral fins. On 27 May 1972, a survey of 12 pools in Gandy Creek, and obtusus streaa, did not reveal atratulus and obtusus •ales in the same pools. In the upper pools only atratulus •ales were found while obtusus •ales were collected only in pools approxi•ately 2 miles downstream. The presence of atratulus males in headwater upstreaa pools and not in lower sections of the stream would be expected since streaa piracy would occur in headwater tributaries. If atratulus and obtusus were separate species then both should appear in the upper pools of Gandy Creek. Only one phenotype, R.- atratulus, is found. It has been previously proven that obtusus and atratulus are si•ilar in habitat and niche. If they were separate species they would be in the sa•e area of the streaa. The abse cs of obtusus in the upper section of Gandy Creek dis•isses the possibility that atratulus and obtusus are distinct and separate species, Obtusus phenotype •ales could have disappeared only through 1ntergradat1on with the atratulus phenotype. No obtusus aales, were collected in Seneca creek. Since both subspecies are eoologioally si•1lar they should be in the sa•e habitat in the stream. This hypothesis holds true where stream capture would oause atratulus to be in an obtusus stream, but is not substantiated due to the absence of obtusus 1n an atratulus stream.

There is a plausible explanation for the above situation. The absence of obtusus charaoter1st1os in an atratulus stream, and the presence of atratulus in an obtusus stream suggests that two phenotypic characters of the same species have been crossed with red coloration of atratulus being dominant. This dominant oharacter1st1o explains why the introduction of the red finned character would remain in non-red finned population and eventually become the phenotype of the population. There is some evidence to suggest that the majority of 1ntergrades between atratulus and obtusus or meleagris may contain the red fin character. The normal probability of)al or some ratio whose sum is 16 or 64 depending on the degree of epistasis may exist with the red fin of atratulus being dominant and the clear fin of obtusus being recessive. It 1ntergradat1on is a hybrid cross, it is possible for a recessive non-red finned trait to appear in a population of red finned dace but the probability is only one-fourth 1n simplest case as great as that of the red fin character in a non-red finned gene pool.

Other possible explanations for red finned atratulus being in an obtusus stream and the absence of obtusus phenotypes in an atratulus stream seem less probable. It is not likely that stream capture only occurred in one direction.

Likewise, it is not possible that this many fish could be transported by fishermen. These streams contain small native trout populations and are seldom stocked, Few fishermen in this area use minnows. Using a legal 4' x 6' seine, it would take approximately 2 hours to find 14 male dace. This investigator's collections 1n May yielded only one ma1e out of every 16 fish. It is mathematically 1m-probable that our collection would include 14 fish introduced by fishermen in a stream containing numerous dace along its entire 14 aile length.

This investigator believes that obtusus and atratulus are subspecies of the same species as designated by Hubbs (1936). This belief is substantiated by the results presented in this paper. However, there are some results that are ambiguous towards this conclusion and cast doubt upon it. Although atratulus has replaced obtusus in the 12 upperpools surveyed in Gandy Creek, there were male obtusus in the lower pools. The 12 upperpools surveyed represented a small number of the total pools in the upper 5 miles of Gandy Creek. It is possible that in other pools in the upper 5 miles there were sizeable populations of obtusus da.oe. This would possibly •ean that atratulus and obtusus were separate breeding populations and therefore separate b1o1og1oal species. The lack of an identifying character to distinguish between a single atratulus and a single obtusus is not really essential as sibling species are identical morphologically. A morphologioall7 distinct species is a necessity for the taxonomist but not necessarily a necessity for meabers of a breeding population, They aight rely not upon body oharaeters but upon behavior to distinguish fertile sexual partners. The leading behavior of obtusus males might serve as such a isolating Beohanism and therefore, it is possible that obtusus and atratulus might be sibling species.

CHAPTER VIII

SUMMARY

1. There was no consistant taxono•1o character which could be used to distinguish a single individual blacknose dace as to subspecies. Nonbreeding coloration was found to be more dependent upon altitude than upon subspecies. High altitude dace of both subspecies showed much darker lateral bands, darker dorsal and whiter ventral coloration,

2. The statistical study of the high and low altitude saaples of the two subspecies did not have the expected results. High and low altitude samples of the same subspecies proved to be significantly different in the majority of body characters measured. This is due either to the null hypothesis or to the degree of mutation that has occurred since the isolation of James and Potomac River dace. The Gandy Creek sample, which contained some fish with the atratulus phenotype, was significantly different from the Seneca Creek sample but not to the degree of significance found between the two atratulus samples. This indicates that the two populations are either intergrading due to stream capture or that the similarity in body characters 1s due to environmental causes, The latter is not believed to be true as it contradicts the findings of Shontz (1966).

J. The habitat of the sampled blacknose daoe was round to be more dependent upon altitude and latitude than upon subspecies. Low altitude populations of both subspecies were restricted to upper pools and riffles ot cool, clear running tributaries. High altitude samples of both subspecies were found in tributaries and in moderately large streams. This confirms the statement of Addair (1944) that temperature is the most i•portant factor limiting 1n the distribution of the blacknose dace. Stomach analyses ot 20 fish from an obtusus stream and 20 fish fro•an atratulus stream showed that the two subspecies consuned the same food and fed in the same area of the stream.

4. The breeding behavior for atratulus in Seneca Creek and obtusus in the Cranberry River was found to be nearly identical. The only exceptions were weak territorality of the atratulus males and the ••leading behavior•• of the obtusus males. The two subspecies bred in the middle section of pools with a moderate current. The water was 8-18 inches in depth and exposed to the sun.

5. A survey of Seneca and Gandy Creeks, two streams where an interchange of populations is known to have occurred, revealed atratulus type males in all of Seneca Creek and the upper section of Gandy Creek. Obtusus type males were found only in the lower sections of Gandy Creek. It is believed that if the two subspecies were separate species they should

be in the same area of Gandy Creek due to the similarities in habitat and niche. Due to the absence of any dace of the obtusus phenotype in the upper section of Gandy Creek, it is not believed that obtusus and atratulus are separate species.

CHAPTEB. IX

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