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HEALTH PHYSICS DIVISION

Radiation Ecology Section

GROWTH AND MOVEMENT OF SMALLMOUTH BUFFALO, ICTIOBUS BUBALUS

(RAFINESQUE), IN WATTS BAR RESERVOIR, TENNESSEE

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Submitted as a thesis to the Faculty of the Graduate School of The University of Tennessee in partial fulfillment of the requirements for the degree of Doctor of Philosophy in Zoology.

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ABSTRACT

The smallmouth buffalo, <u>Ictiobus bubalus</u> (Rafinesque), population of Watts Bar Reservoir, Tennessee, was investigated in order to describe its age distribution, growth rates, dispersion, and importance as an accumulator of radionuclides. Measurements and scale samples were taken from commercially-caught fish and fish caught in the ORNL tagging operations. Scale impressions were analyzed for age and growth phenomena. Dispersion of smallmouth buffalo was investigated by conventional tagging methods and by autoradiographic analyses of scales. Stable and radiochemical composition of scales was examined by spectrographic analysis, flame spectrophotometry, and radiometric surveys.

Watts Bar smallmouth buffalo in the commercial catch ranged from four to fifteen years of age. The largest number of fish in the catch was from year class six, the youngest year class which was completely vulnerable to commercial fishing gear. Annulus formation occurred prior to June. The total survival rate was found to be 49 per cent for year class six, 35 per cent for year class seven, 26 per cent for year class eight, and 19 per cent for year class nine.

The rate of change in weight as length increased was lo0 g/cm for fish exceeding 31 cm in total length. Absolute growth was 422 mm at three years, 441 mm at six, 487 mm at seven, 522 mm at eight, and 609 mm at nine. The species characteristically exhibited the largest relative growth during the second year of life. Conditions for growth evidently had improved for the past six years as was indicated by an increase in total length attained at the end of succeeding years. Growth compensation was evident during the fourth and fifth years of life.

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Calcium was the most abundant element in fish scales with at least twenty-three other elements present in varying quantities. Fish scales and bone were found to contain radionuclides of ruthenium, cesium, zirconium, zinc, and cobalt. Radiometric surveys of scales revealed the Watts Bar Reservoir smallmouth buffalo population was a relatively minor accumulator of radionuclides with only 0.08 per cent showing the presence of artificially produced radionuclides. Approximately 6 per cent of the Clinch River fish and 77 per cent of the White Oak Creek fish had accumulations.

Limited data on dispersion were determined from conventional tags. Much more dispersion and life history data were determined from autoradiographic analyses of scales. These dispersion data were applied only to individuals because the number was too small for generalizations for the population as a whole.

All normal scales containing radionuclide accumulations were found to produce identical autoradiographic patterns of concentric circles which were associated with growth of the fish in contaminated areas. This phenomenon was combined with conventional capture-recapture methods of population estimates in a proposed technique of population studies. A laboratory experiment showed that scales could be tagged with cesium-134, but this radionuclide was found to accumulate in much larger concentrations in the soft tissues than in the bony tissues.

Data on population characteristics of the smallmouth buffalo are biologically significant in that they increase our basic knowledge of this commercially important species. The dispersion study is especially important in that an entirely new technique of study was developed and found to be superior to conventional tagging methods.

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CHAPTER I

INTRODUCTION

In ecological investigations it is necessary to determine the interspecific and intraspecific relationships between organisms, their effects on the physical environment, and effects of the physical environment upon the organisms. The study of organisms in such an ecological investigation often follows the form of a population study designed to determine the characteristics of that population. A population is considered to be a group of organisms of the same species occupying a particular space and possessing characteristics of the group which are not characteristics of the individuals of the group. Some of these characteristics are: density, birth rate, death rate, age distribution, biotic potential, dispersion, and growth form (Odum, 1959).

The primary objective of this study was to determine the age distribution, growth rates, and dispersion characteristics of a selected fish population in the Clinch River. Because this investigation was a part of the continuing Clinch River Study (Morton, 1961), it included an investigation into the species' importance as an accumulator of radionuclides.

The smallmouth buffalo, <u>Ictiobus</u> <u>bubalus</u> (Rafinesque), was selected for this investigation of population characteristics for several reasons. An examination of the fish tagging records of the Radiation Ecology Section revealed that the species is abundant in the

area throughout the year. The species is commercially important as indicated by the fact that over one million pounds are harvested annually from Tennessee Valley Authority impoundments. A preliminary radiometric survey of fish species from the Clinch River indicated that the smallmouth buffalo was one of the biotic accumulators of radionuclides released into the river as waste from the Oak Ridge National Laboratory.

The study of population characteristics was based on the examination of scales from the fish. It is a general principle that the scales register all the stages of growth of fish and that every factor influencing this growth is expressed on the sculptured, outer surface (Bertin, 1958). Periods of rapid growth, retarded growth, and even periods of spawning activity may be interpreted from the relative position of marks on the scales of most fish. Lea (1910) established that there is a constant relationship between the size of the fish and the size of its scales. Examination of the outer surface of the scale reveals the animal's current age. The knowledge of age is essential in the study of growth. In conventional growth studies the scales regularly are used to compute the length of the fish at the end of previous growing seasons, as indicated by the spacing of year marks (Ricker, 1958).

Emigration, immigration, and migration are movements of individuals which may affect several of the population characteristics. The investigation of these movements now is limited to capture-recapture study methods and conventional marking techniques. A major disadvantage exists in the conventional methods of marking fish. The attachment of metal or plastic tags to the animal's body has been shown to influence its behavior and inhibit growth (Ricker, 1942).

A preliminary autoradiographic examination of scales from several species of Clinch River fish revealed that the radionuclides had accumulated in patterns of concentric circles. This accumulation was assumed to be the result of growth of the animal in a contaminated area. If any of the nutrient materials used by the animal in forming new body tissues contain radioisotopes of essential elements, these isotopes will follow the same pathway as stable isotopes of the element and be incorporated into these tissues. When accumulation of radionuclides occurs in the scales it can be detected by autoradiography. A comparison of scale autoradiograms to the growth of the specimen should reveal when that individual was in a contaminated area.

In recent years radioisotopes have been applied effectively to the investigation of several phases of aquatic biology. Biological productivity and rates of biogeochemical cycling have been measured by radionuclide methods. They have been used in the investigation of fish diseases and nutrition. They also have been applied in tracing the movement of water and pollutants in water in hydrologic studies. Some application of radionuclides has been made to the marking of aquatic animals, but little success has been achieved. Most of these studies have been based on the use of radioactivity as a means of locating the radioactive individuals. One such study was conducted by Kondrat'ev (1962) in which he tagged commercial fich species by holding them in water containing a weak concentration of radionuclides for several Then these fish were released and recaptured by commercial hours. fishing methods. The catch of fish was passed through tanks equipped with radiometric counting devices and the number of radioactive fish

was recorded. This method had some success in testing the efficiency of commercial fishing gear.

Pendleton (1956) pointed out the advantages of radionuclides for marking animals in ecological studies. The radionuclides are not detectable by the senses of the animals and they are easily applied with minimal handling to large groups of animals. Some techniques do not require that the organisms be captured, handled, or even seen by the investigator. The radionuclides are incorporated into the individual's body, thereby tending to prevent loss of the tag. Seymour (1958) discussed the tagging of fish with radionuclides, but concluded that present-day marking methods are much more practical. His objections to tagging fish with radionuclides were that tagged fish are difficult to detect because of the shielding effect of water, the high energy radiation necessary in radioactive tags might have a detrimental effect on the fish, the fish tagged with radionuclides might constitute a health hazard if consumed by humans, and the identification of individuals by such tags would be extremely complicated. However, Seymour appears to have considered using radionuclides primarily as a means of locating fish, as most previous investigators have done. Hooper, Podoliak, and Snieszko (1961) stated that future use of radionuclides in the marking of aquatic animals will be only in situations where there is complete control over the fish harvest or where the tagged fish can be handled without danger to the public.

CHAPTER II

REVIEW OF LITERATURE

A. Population Characteristics

1. Age distribution

Lotka (1925) concluded that a population tends to develop a stable age distribution. Movement of individuals from other populations or changes in environmental conditions may disrupt this balance. However, the population eventually regains its old stability or a new one after the disturbance. Allee, et al. (1949) discussed the relationships between age distribution and other characteristics of the population. There is a preponderance of young individuals in the population soon after spawning because of the high fecundity of fish. However, the survival rates for young fish usually are low because of the intense pressure of predation. Predation continues until the young fish reach a size where they no longer are suitable prey for larger fish. Survival rate is the factor determining the number of individuals entering a new age group.

2. Dispersion

Populations of stream fish in natural habitats cannot be assumed to be isolated units. In the absence of physical barriers movement of individuals occurs between adjacent populations. The movement of fishes may be random (Thompson, 1933), but more likely the movements have cause in population pressures, environmental changes, or migration behavior.

Funk (1955) presented evidence supporting a concept of stream fish populations being divided into sedentary and mobile groups. His data on fourteen species revealed that each species included a sedentary group which remained near the point of capture and release and a mobile group which ranged more or less widely. Carp seemed to adapt their movements to the physical conditions of their habitat. They usually were sedentary in stable habitats, but mobile in habitats subject to flooding. Some carp, the only rough fish species included in this work, ranged as far as 200 miles. Gerking (1953), Larimore (1952), Funk (1955), and others have concentrated on the investigation of game fish movements. home ranges, and homing behavior. Game fish are much less mobile than rough fish. Miller and Bryan (1948) after making a limited investigation of movements of fish in Tennessee Valley Authority impoundments. concluded that the fish populations of creek embayments studied were more or less independent of the main reservoir and few fish moved back and forth between them. Present knowledge of the movements of rough fish is limited because of the past emphasis placed on the study of game fish movements, but it is generally believed that rough fish do not maintain home ranges or exhibit homing behavior and that they range considerably farther than game and pan fish species.

3. Growth

Growth is defined as an increase in size. Rounsefell and Everhart (1953) described growth by two different approaches. Absolute growth is the average size of fish at each age. This size may be either length or weight measurements. The absolute growth rate curve is sigmoidal in shape and the inflection indicates the point at which the

rate changes from a continually increasing rate to a decreasing rate of growth. Relative growth is defined as percentage growth in which the increase in size in each time interval is expressed as a percentage of the size attained at the beginning of the time interval. Relative growth is most rapid in younger fish and constantly declines. Total lengths were used in describing growth of smallmouth buffalo in this study because commercial fishermen removed the viscera before bringing the fish to the collection point.

B. Fish Scales in Population Studies

1. Methods

Carlander (1956) evaluated the methods currently used in studying age and growth. Recapture of tagged fish has been the method used in population studies by most investigators. Black (1957), Ricker (1958), Woodbury (1956), and many others have found that the presence of a tag on the fish's body inhibits growth and influences behavior. Hile (1941) analyzed the uses of length-frequency groupings for age determination and found that considerable inaccuracies existed because varying growth rates of individuals eliminate peaks of abundance at older ages. Most investigators agree that the interpretation of growth rings on scales, vertebrae, otoliths, opercular bones, spines, and fin rays is the best source of information on the age and growth of fish in natural habitats.

2. Scale formation and structure

Van Oosten (1957) summarized information on the formation and development of teleost scales. The scale has its origin in a mass of

fibroblast cells in the dermal layer of the skin. This cell mass flattens out to form two distinct layers between which there appears a fibrous network. Surrounding osteoblast cells initiate the formation of the bony layer by secreting calcium salts into the osteoid tissue. The fibrillary plate next appears as a thin sheet between the bony scale and the lower layer of osteoblasts.

Growth of the formed scale is continued by addition to the margin of the bony surface layer and the deposition of thin fibrous layers below it. Since the surface layer grows by deposition of materials at the edge, it does not increase in thickness with age and the early surface sculpturing does not change except for wear. This fact makes it possible to determine the age of the fish from its scales. The thickest part of the scale is always in the center. Scales may be thought of as greatly flattened cones (Figure 1). The fibrillary plate is largely or entirely uncalcified and without vascular canals. The bony layer is composed of an organic framework impregnated with inorganic salts, mainly calcium phosphate and calcium carbonate. The surface sculpturing of scales has been described in detail by Creaser (1926).



Fig. 1. Cross-Section Diagram of a Fish Scale.

C. Accumulation of Radionuclides by Fish

The advent of atomic energy installations has led to the contamination of some aquatic environments with low-level radioactive wastes. The distribution of these radionuclides in any aquatic environment will vary with the physical, chemical, and biological characteristics of that environment. Concentrations of radionuclides will vary between species and tissues and will fluctuate according to food habits, life cycles, and seasonal changes. A major quantity of the radionuclides within the biota will be held by organisms which make up the primary trophic levels in the early stages of contamination of aquatic habitats where the standing crop of producers exceeds that of the consumers. However, the radionuclides will move to other trophic levels later where they may be concentrated in large quantities (Davis and Foster, 1958).

Krumholz, Goldberg, and Boroughs (1957) summarized the factors which contribute to the accumulation of radionuclides in living organisms. The accumulation and loss of radionuclides depends on their physical half-lives and biological factors contributing to their incorporation in, retention by, and disappearance from the organisms. Water characteristics, such as salinity, per cent composition of the dissolved solids, pH, oxygen-carbon dioxide ratio, and the presence of complexing agents, also affect the accumulation of radionuclides.

The radionuclides of strontium, cesium, cobalt, and ruthenium are considered to be the most important waste products released into the Clinch River from a bioaccumulation point of view. Lack of investigation prohibits generalizations on the accumulation of cobalt

by fish, but the other radionuclides have been investigated to some extent.

Boroughs, Chipman, and Rice (1957) traced an ingested dose of cesium-137 in small tuna, <u>Thunnus</u> spp., and found the radionuclide was taken up rapidly by the liver, heart, spleen, and kidneys, but was lost rapidly by these organs. Muscle, gonads, brain, and integument continued to accumulate cesium-137 faster than they lost it. Davis and Foster (1958) suggested that absorption was the primary method of cesium uptake, but experiments into this specific problem are inconclusive. Data on cesium-134 uptake by sunfish in this study support the idea that radiocesium enters the fish's body in considerable amounts through ingestion and accumulates in the soft tissues.

Jones (1960) discovered that bottom-feeding plaice, <u>Pleuronectes</u> <u>platessa</u>, accumulated nitrosyl ruthenium-106 in the liver and spleen by eating organisms embedded in contaminated silt. The skin activity of these fish was low. When menhaden, <u>Brevoortia</u> spp., were fed ruthenium-106 there was only 0.05 per cent of the ingested dose remaining in the digestive tract after 128 hours. There was 0.25 per cent in the fish's body or on the skin surface and 0.01 per cent in or on the gills. It can be concluded that ruthenium-106 enters the fish's body by ingestion and accumulates in the active tissues, but only in small quantities.

The radionuclides of strontium have been studied extensively because of their long half-lives and tendency to concentrate in bony tissues. In one of the earliest studies on the absorption of radionuclides by fish, Prosser, <u>et al.</u> (1945) immersed goldfish, <u>Carassius</u> auratus (Linnaeus), in a solution containing strontium-89. They

determined that the gills, skeleton, and integument of large goldfish were ten to twenty times more radioactive with strontium-89 than muscle tissue. The scales contained about 80 per cent of the total activity of the integument and the bony element of the gills contained more than the soft portions. Fat tissues were higher than integument in strontium-89 accumulation. Muscle and eggs were the lowest in strontium activity. Brain, heart, liver, testes, and swim bladder were relatively low in strontium activity. Of the total radioactivity of goldfish immersed in radiostrontium, two-thirds of the activity was in the integument, one-sixth in the skeleton including the fins, and one-tenth was in the gills including the bony element. Saurov ÷. * (1957) found teleosts absorbed strontium-90 from an environmental solution with a higher accumulation in scales and bone than in muscle and internal organs. Bidwell and Foreman (1957) placed rudd, Scardinius erythrophthalmus (Linnaeus), in fresh water tagged with strontium-90 and after 272 days found a high accumulation in scales, a low accumulation in skin, and an intermediate accumulation in bone. Danil' chenko (1958) concluded that strontium-90 enters the vertebrate body and settles in skeletal structures, replacing calcium. Ophel (1962) observed that shiners, Notropis spp., living in Perch Lake, Ontario, which had contained strontium-90 for approximately five years, had a whole body concentration factor of 950 times that of the water. The flesh of perch in the same lake had an average concentration factor of five, while the bone of perch had an average concentration factor of 3.000 at the equilibrium which was reached in the fifth year. Martin and Goldberg (1962) found 95 per cent of the radiostrontium fed to Pacific mackerel, Scomber japonicus Houttuyn, was excreted in

twenty-four hours. The remaining five per cent was fixed for at least 235 days with 80 per cent of this activity being in the calcareous tissues. Boroughs, Chipman, and Rice (1957) working with <u>Tilapia</u> observed that about 70 per cent of the radiostrontium accumulated in bone was readily exchangeable and that the remainder was firmly bound in a lattice or to an organic matrix with a slow turnover rate. It can be concluded that radiostrontium enters the fish's body primarily through absorption and accumulates in varying concentrations in all tissues. The highest accumulations occur in the bony tissues where the element has a slow turnover rate.

CHAPTER III

STUDY AREAS, CRECIEC, AND METHODS OF STUDY

A. Study Areas

Data on fish in this study were collected from White Oak Creek, the Clinch River, and Watts Bar Reservoir. White Oak Creek, the major source of radioactive waste contamination (Morton, 1961), is within the backwaters of Watts Bar Reservoir and at full pool has an area in excess of five acres. White Oak Creek water is diluted an average of 450 times at the point where it enters the Clinch River, 20.8 river miles upstream from the confluence of the Clinch and Tennessee rivers. Clinch River water is diluted an average of 5.6 times as it enters the Tennessee River at TRM 567.7. Watts Bar Reservoir on the main stream of the Tennessee River is formed by Watts Bar Dam at TRM 529.9. This reservoir contains a surface area of 38,600 acres at full pool with a shoreline of 783 miles (Fig. 2).



Fig. 2. Clinch River and Watts Bar Reservoir, Tennessee.

B. Species Description

The smallmouth buffalo, <u>Ictiobus</u> <u>bubalus</u> (Rafinesque) is a member of the Sucker Family, Catostomidae. The species is widely distributed from Lake Erie south to Mexico. It is common in the Mississippi, Missouri, Ohio, and Tennessee rivers and their larger tributaries. The smallmouth buffalo reaches a size in excess of 30 inches and 25 pounds. Schoffman (1944) reported a specimen 30 inches long weighing 25 pounds 8 ounces from Reelfoot Lake, Tennessee. A 33.8 inch specimen weighing 23 pounds was taken from White Oak Creek in May 1962. Specimens of 15 pounds are relatively common in commercial catches on TVA reservoirs, but the average weight is near three pounds.

The smallmouth buffalo are bottom-feeders preferring muddy or silty bottoms. They eat both plant and animal foods. Aquatic insects, mollusks, other small aquatic animals, and algae are common in their diet. Local commercial fishermen occasionally find their stomachs packed with plant seeds. Weiss (1950) reported that in season this species may pack their stomachs with cotton from cottonwood trees or the seeds of other plants and trees. Since the introduction of carp which utilize the same foods and habitats, the two species have been in direct competition. However, both species are abundant in Watts Bar and this competition has not been observed to affect either species adversely.

Smallmouth buffalo evidently inhabit the deeper, swifter waters of the large rivers. No mass migrations, such as spawning runs, have been noted locally. They spawn in the early spring in sloughs and shallow weedy areas. An eight to ten pound female lays 300,000 to 400,000 eggs which are fertilized and scattered on the bottom and left without parental care (Weiss, 1950). There is undoubtedly a high mortality rate for the eggs and young fish. In spite of this, the smallmouth buffalo have flourished in the impoundments of the Tennessee Valley Authority system. They build up large populations. Yields of

up to 700 pounds per acre have been reported from small lakes in Missouri.

CHAPTER IV

AGE DISTRIBUTION OF SMALLMOUTH BUFFALO

A. Specialized Methods

1. Annulus determinations

The scale method of age determination was proved to be valid for smallmouth buffalo by Schoffman (1944) and by Eschmeyer, Stroud, and Jones (1944). However, annulus formation in this species is not distinct and this fact leads to some difficulties in age determinations. Cutting over is a term applied to the presence of incomplete circuli between complete circuli. These incomplete circuli are the result of cessation of growth during spawning or adverse environmental conditions. Incomplete circuli may even be formed if the fish is injured. Known age buffalo from Wisconsin were examined and their scales were found to be similar to the Watts Bar fish. The annuli on the scales of the Wisconsin fish were not complete. This phenomenon can be considered to be a characteristic of the species.

Gross inspection of the buffalo scales gave a good idea of the different seasonal growth rates and was found to be useful in aging the fish. There was definite crowding of the circuli immediately inside the annuli toward the focus or center of the scale which corresponds very well to the reduced growth rate that would be expected during the winter. Annulus formation seems to be followed by wider spaces between the circuli during the summer growth period which is due to the increased growth rate in the summer.

Several criteria were established for defining the true annuli on scales of the smallmouth buffalo. Usually there was some cutting over by the annulus in the lateral fields of the scale near the borders of the anterior and posterior fields. There definitely was some irregularity or pattern change in the posterior field along the annulus which was most obvious in gross inspection of the scale impression. In many scales there appeared to be crowding of the circuli prior to annulus formation and wider spacing of circuli after annulus formation. The change in spacing was most obvious in the anterior field. In many instances the scale was observed to have cracked along the annulus during pressing.

2. Tagboard strip manipulations

Two scales were examined from each fish on two different occasions giving four scales from each specimen in the calculations. In order to achieve the most consistent results in annuli determinations, tagboard strips were employed in this study. A tagboard strip is a strip of paper which is laid directly over the projected image of the scale impression. The strip is marked at the focus, margin, and each annulus of the scale on a radius through the center of the anterior field. The distance from the focus to each annulus and to the margin was measured in millimeters. A ratio was calculated between the distance from the focus to each annulus and the distance from the focus

to the margin. A figure representing the percentage of the total distance from the margin to the focus was given to each annulus.

Usually four tagboard strips were made on scales from each fish. In some instances regenerated scales in the sample limited the readable scales to less than four. The tagboard strips from one individual were compared to each other. When the distance ratio to the same annulus corresponded closely on all four tagboard strips the average distance ratio was taken as the correct one. When obvious deviations existed between corresponding distance ratios on any of the four tagboard strips, the scales were reread to determine the correct location of the annulus.

When the tagboard strip examinations were completed the age of the individual was compared to the total length. If the length was out of proportion to the fish's apparent age, the scales were reexamined to determine if the correct age had been calculated. In most instances such fish were determined to have had exceptional growth, either fast or slow, and the calculated age was allowed to stand.

A preliminary age-frequency grouping was made after the completion of scale readings and annuli determinations. Fish in each age group were arranged according to total length. The median total length for each age group was determined. Individuals which deviated widely from the median were reexamined to verify their calculated age. Most of the deviants were found to be in the correct age group and to have had an extremely fast or slow growth rate which had placed them on the margin of the size range for their age group.

B. Analysis of Smallmouth Buffalo Age

After the final determination of the age of each individual all the specimens were grouped into year classes. A year class designation indicates that the fish has lived through a certain number of winters. Year class 1 had passed through one winter, but had no annulus. Year class 2 had passed through two winters and had one annulus. This method of year class designations continues through year class 15 which had passed through fifteen winters and had fourteen annuli.



Fig. 3. Age-Frequency Distribution of Watts Bar Smallmouth Buffalo by Month of Collection.

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Figure 3 represents the age-frequency distribution for the monthly collections of smallmouth buffalo from Watts Bar Reservoir for June, July, August, and September 1962. Figure 4, page 20, represents the age-frequency distribution for the four-month total. Figure 5, page 21, represents the length-frequency distribution of Clinch River smallmouth buffalo for 1960 and 1961, and the Watts Bar smallmouth buffalo for the summer of 1962.

The smallmouth buffalo of year class 6 were the most common in the commercial catch from Watts Bar during June, July, and August 1962 (Figure 3). However, year class 5 fish became most numerous in the September catch. This was due to recruitment into the vulnerable size during the late summer. The nets used by the commercial fishermen were of three inch mesh, and therefore, were selective for fish that had reached a size of approximately 400 mm in length. During June, July, and August, only the fish in the year class 6 and upwards had reached this minimum catchable size in large numbers. As the temperatures rose during the summer and growth rates increased, fish of year class 5 became large enough to be caught in large numbers. These data do not mean that either the fifth or sixth year class was dominant. The smallmouth buffalo in Watts Bar probably correspond to the theoretical age distribution curve for fish if the population is stable. The younger year classes contain larger numbers of individuals and become succeedingly smaller in each following year as the result of mortality. There is a smaller number of individuals in each succeeding year class unless a dominant year class is formed by exceptional survival for one year class allowing a large number of individuals to enter the next





year class. No indications of such a dominant year class were found in the smallmouth buffalo population in Watts Bar Reservoir.

The length-frequency distribution graph of Clinch River and Watts Bar smallmouth buffalo (Figure 5, page 21) illustrates the effect of net selectivity. Commerical fishing gear allows only the larger individuals of year class 4 to be captured, although it is probable that more individuals of this year class are present than individuals in year class 5 or 6. The greater frequency of the smaller size fish in the



Fig. 5. Length-Frequency Distribution of Clinch River Smallmouth Buffalo for 1960 and 1961, and Watts Bar for 1962.

catches from the Clinch River in 1960 and 1961 resulted from the use of nets made of smaller mesh. A minimum mesh size of about one inch was used on the Clinch River, whereas the minimum mesh size on Watts Bar was three inches.

Assuming that the Watts Bar smallmouth buffalo numbers are representative from year class 6 through 10, the survival rates for the species in these year classes were calculated by the formula:

$$s = \frac{N_{t+1}}{N_t}$$

where $N_t =$ the number of fish in any year class and $N_{t+1} =$ the number of fish in the succeeding year class. Four hundred and ninety fish per thousand in year class 6 enter year class 7, 350 of year class 7 enter year class 8, 260 of year class 8 enter year class 9, and 190 of year class 9 enter year class 10. The number of individuals in the year classes older than ten was too low for calculation of survival rates.

Survival rates for Watts Bar smallmouth buffalo appear to be higher than those calculated from data on Wisconsin fish (Frey and Pedracine, 1938). The Wisconsin fish had 580 individuals per 1,000 surviving from year class 3 to 4, 130 from year class 4 to 5, 400 from year class 5 to 6, and 130 from year class 6 to 7. The Wisconsin data were characterized by the presence of dominant year classes.

Time of annulus formation affects the results of population studies where scale reading forms the basis for age determinations. Table I shows the calculated average total length at the last annulus for fish in each year class. These data are grouped according to month of collection. Calculated length increment since the last annulus was observed to increase steadily from June through September in all year classes except eight, where the June group had 3 mm more growth since the last annulus than the July collection of the same year class. This. increase in length since the formation of the last annulus would indicate the annulus is formed sometime prior to June in Watts Bar smallmouth buffalo. Average total length at capture revealed an expected increase in total length between June - July and between August -September in most year classes. However, in all the year classes there was a noticeable decrease in the average total length between the July and August collections. These data indicate that the larger fish from all year classes are caught in June and July and that the smaller fish of the same year classes are caught in August and September. Presumably, fish caught in August should have an added month's growth over

TABLE I

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	Month Col-		Average Total Length (mm)	Average Calculated Total Length (mm)	Calculated Length Increase Since
Age	Tected		at capture	at Last Annulus	Last Annuius
4	Jun	53	451	424	27
	Jul	11	453	426	27
	Aug	17	441	406	35
	Sep	7	473	437	36
5	Jun	177	459	440	19
	Jul	67	470	449	21
	Aug	58	461	436	25
	Sep	90	469	439	30
6	Jun	180	470	454	16
	Jul	117	473	457	16
	Aug	86	469	450	19
	Sep	75	475	449	26
7	Jun	72	482	470	12
	Jul	53	488	474	14
	Aug	70	474	456	18
	Sep	30	477	456	21
8	Jun	12	536	521	15
	Jul	38	498	486	12
	Aug	21	490	473	17
	Sep	8	500	475	25
9	Jun Jul Aug Sep	96 M Q	551 549 500 478	540 537 481 459	11 12 19 19
10	Jun	3	650	643	7
	Aug	1	526	505	21
11	Jun	1	500	495	5
	Jul	1	736	714	22
	Aug	1	5 ¹ 15	523	22
13	Jun	l	625	619	6
15	Jun	l	850	842	8

CALCULATED AVERAGE TOTAL LENGTHS (BY MONTH)

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those caught in July. Since all the collections were taken from a region on the main stream of the Tennessee River, it is possible that smallmouth buffalo populations from different tributaries combine to form the Watts Bar mainstream population. If different growth rates existed in the various tributary populations, it would be possible that a segment of the Watts Bar population with a faster growth rate could move into the fishing area early in the summer and that a segment with a slower growth rate could arrive later. This possibility of a segmented population could not be tested from data in this study because all collections were made in the same area.

Recruitment is defined as the addition of new fish to the vulnerable population by growth of smaller size categories. Ricker (1958) described the modal age in the frequency distribution of the catch as lying quite close to the first year in which recruitment can be considered complete. Year class 6 was the modal age in the catch of Watts Bar smallmouth buffalo. The smallest sixth year class fish caught was 400 mm in length. Net selectivity allowed most fish less than 400 mm in length to pass through the 3 inch mesh. Data in this study indicate that recruitment is complete at age six and that fishing pressure is equal on all fish from year class 6 upward.

Recruitment was determined for year classes four through, nine by back-calculation of the total length at previous annuli. Total length of 400 mm was considered the minimum size for fish caught in 3 inch mesh nets and the percentage of fish exceeding this minimum length at each age was determined (Table II). Increasing growth rates for Watts Bar smallmouth buffalo in their early years have resulted in
younger fish being recruited into the fishery each year for the past five years.

TABLE II

		Year Class										
Age	4	5	6	7	8	9						
1	0	Ņ	Ò	0	0	0						
2	1	0	0	0	0	0						
3	84	26	6	2	1	5						
4	100	98	72	29	18	15						
5	_	100	99	91	76	55						
6	-	-	100	100	100	100						

PER CENT VULNERABLE AT EACH AGE

CHAPTER V

GROWTH OF SMALLMOUTH BUFFALO

A. Length-weight Relationship

In the tagging operations at Oak Ridge National Laboratory during 1960 and 1961, 655 smallmouth buffalo were taken from the Clinch River. These fish were measured to the nearest one-half centimeter and weighed to the nearest ten grams. In the normal course of operations all the fish-tagging data are recorded on IBM record cards. The length and weight data on the smallmouth buffalo were used in calculating a regression equation of weight as a function of length. This equation expressed the rate of change in fish weight as total length increased. Calculations were made by IBM 7090 computer. The lengthweight relationship of the 655 smallmouth buffalo from the Clinch River



Fig. 6. Length-Weight Relationship of Clinch River Smallmouth Buffalo.

is illustrated by the scatter graph (Figure 6) with each point representing one individual. The calculated regression line is plotted on the graph.

Weight of a fish is considered to be a function of length (Hile, 1936). If the form and specific gravity of a fish were constant throughout its entire life the relationship between length and weight could be expressed as a constant. The length-weight relationship is expressed usually by the formula:

$$W = aL^n$$

where W = weight in grams, L = total length in millimeters, <u>a</u> is a constant, and <u>n</u> is an exponent. The calculated regression coefficient (a) is 0.9976 and the exponent (<u>n</u>) is 3. These data result in the formula for the rate of change in fish weight:

 $W = 0.9976 L^3$

The 95 per cent confidence interval on <u>a</u> is (0.9749 - 1.0204). Standard error of the regression coefficient is 0.0116. The length-weight regression line may be used as a nomogram for the conversion of measured total length to estimated weight for smallmouth buffalo within the length range covered by the nomogram.

B. Growth Analyses

Growth rate calculations were made on 1,271 smallmouth buffalo collected over a one week period each in June, July, August, and September 1962 from Watts Bar Reservoir. Total length of each fish to the nearest millimeter was used in conjunction with the distance ratio between focus-annulus and focus-margin of scales from the age determination study. The total length of each individual at each previous annulus was determined by use of the formula (Bertin, 1958):

$$L_1 = \frac{e_1}{e_m} \times L_t$$

where $L_1 = \text{total length of fish at the time the first annulus was}$ formed, $e_1 = \text{distance from scale focus to the first annulus, } e_m = \text{distance from scale focus to margin, and } L_t = \text{total length of fish at}$ capture. This formula is based on the fact that the size of the scales increases proportionally as the size of the fish increases. Annulus distance ratios and the individual's total length at time of capture were recorded on IBM record cards. Total length of each fish at each successive annulus was back-calculated, summed, and averaged for each year class by month of collection. All calculations were made by IBM 1420 Computer.

Absolute growth is the average size attained by the fish at each age. Length was the parameter selected for describing the growth of Watts Bar smallmouth buffalo because the fish were sampled at a local fish wholesale house after having been gutted on the lake by commercial fishermen. Absolute growth of the smallmouth buffalo has varied widely over the past fourteen years (Table III). The number of individuals in the older age groups (ten, eleven, thirteen, and fifteen) was too small for accurate generalizations on these year classes. The calculated total length at the end of the first year's growth has increased steadily from 98 mm for year class nine through 134 mm for year class four.

TABLE III

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ABSOLUTE GROWTH OF SMALLMOUTH BUFFALO

		Average Total Length (mm)		Ca	lculated	Total	Length	Average Range	at Succ	essive A	nnuli	
Age	N	at Capture	1	2	3	4	5	6	7	8	9	10
4	88	451 391-520	134 82 - 246	303 206-400	422 368-484							
5	392	463 420-543	119 75 -1 81	277 152-372	382 257 - 445	441 356-50	2			·		
6	458	471 400-595	110 71 - 175	253 1 47 - 344	348 232 - 465	412 312 - 53	453 4380 - 559	I				
7	225	480 430-594	104 71-215	233 143-372	318 238 - 485	385 299 - 50	432 8363-550	465 398 - 582				
8	79	502 451-600	103 61-179	219 146 - 318	306 245 - 450	372 311 - 52	422 2 356-540	459 397 - 576	487 424 - 594			
9	20	535 465-650	98 71 - 149	207 152 - 268	290 230 - 406	355 296 - 45	412 7 363 - 489	458 402 - 552	428 - 598	522 446 - 631		
10	4	619 526 - 805	120 89 - 177	231 205 - 290	314 279 - 378	381 337 - 45	439 9 384-531	491 421 - 604	535 452 - 676	57 1 479 - 733	609 505 - 797	
11	3	594 500-736	91 65 - 125	165 140 - 191	238 210 - 280	328 270 - 42	397 0 330 - 508	444 370 - 559	476 400-596	505 430 - 633	538 460 - 670	577 495 - 714

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The increase in calculated total length at the end of the first year's growth amounted to 5, 1, 6, 9, and 15 mm respectively for year class eight through four.

The relationships between absolute growth of the various year classes are apparent in Figure 7. The dashed lines connect the length for year classes nine through four at corresponding ages. The increasing slope of the dashed lines indicates that Watts Bar smallmouth buffalo have been increasing in total length in each successive year class. The increase in absolute growth for successive year classes probably was the result of improved food availability through removal of competing fish by commercial fishing.

Fishing pressure on the smallmouth buffalo has increased in Watts Bar Reservoir since 1958 when 15,687 pounds were caught through 1961 when 59,328 pounds were caught. The increase in fishing pressure would





result in decreased population density, in turn leading to improved food availability. However, smallmouth buffalo density data are not available at this time.

Absolute growth in weight was calculated from data on year classes five through nine. The average calculated total lengths for these year classes at annulus 4, 5, 6, 7, and 8 (Table III, page 29) were averaged. These average total lengths were converted to estimated weights by use of the length-weight regression nomogram (Figure 6, page 26). Watts Bar smallmouth buffalo had an average weight of 895 g at the time their fourth annulus was formed. The fish gained 275 g during their fifth year of life, 260 g during the sixth, 290 g during the seventh, and 355 g during the eighth. Insufficient numbers of individuals in the sample from other year classes prohibited calculation of weight increases in other years.

The average annual growth increment (Table IV) is largest for the second year of life in all year classes where adequate numbers exist in the sample. In year classes four through nine the second year's growth exceeded that of the first year by 35, 39, 33, 25, 13, and 11 mm respectively. In year classes ten, eleven, and thirteen the average annual growth increment for the second year of life was less than the first. The smaller increment for the second year of life in these three year classes is questionable because of small numbers of individuals in these year classes and the fact that annulus determinations of these older fish are subject to considerable inaccuracies. In year classes four through nine the third year's growth was less than that of the second year by 50, 53, 48, 44, 29, and 26 mm respectively.

TABLE IV

Year							Y	ear						
Class	1	2	3	4	5	6	7	8	9	10	11	12	13	14
4	134	169	119											
5	119	158	105	59										
6	110	143	95	64										
7	104	129	85	67	47	[.] 33								
8	103	116	87	66	50	37	28							
9	9 8	109	83	65	57	46	35	29						
10	120	111.	83	67	58	52	44	36	38					
11	91	74	73	90	69	47	32	29	33	39				
13	113	93	75	50	57	43	32	37	31	32	31	25		
15	119	170	36	77	68	59	43	42	43	25	17	17	17	9

AVERAGE ANNUAL GROWTH INCREMENT (MM)

The decrease in average annual growth increment continued through succeeding years after the second year's growth for all year classes examined. However, there were some fluctuations up and down, probably as a result of some favorable growth seasons. A graphic illustration of the annual growth increments (Figure 8) clearly shows the relationships between the amount of total length added each year by year classes nine through four. Apparently the habitat conditions for fish during their first three years of life have been improving in Watts Bar Reservoir since 1954, the year of spawning for year class nine. Each year class has been successively larger at the time it formed its first,



Fig. 8. Average Annual Growth Increments of Watts Bar Smallmouth Buffalo.

second, and third annuli. There was one unexplained exception where the difference was only 2 mm. Year class seven had a smaller growth f increment during its third year of life than year class eight.

Absolute growth of Watts Bar smallmouth buffalo was compared to growth of the species in other areas (Table V, page 35). Calculated lengths at each age for year classes four through nine were averaged. These data were compared to back-calculated growth data on smallmouth buffalo from Grand Lake, Oklahoma (Thompson, 1950), Wister Reservoir, Oklahoma (Hall, 1951), Chickamauga Reservoir, Tennessee (Eschmeyer, Stroud, and Jones, 1944), and Reelfoot Lake, Tennessee (Schoffman, 1944). Smallmouth buffalo in Grand Lake, Oklahoma, were larger than those in Watts Bar at the end of the first year. The species was similar in size at the end of two years in both areas. However, Watts Bar smallmouth buffalo at three, four, and five years of age were larger than those in Grand Lake by 40, 53, and 60 mm respectively. Smallmouth buffalo in Wister Reservoir, Oklahoma, exceeded those in Watts Bar Reservoir at every age from one through six. This species is larger in Reelfoot Lake, Tennessee, than in Watts Bar at every age from one through seven. Smallmouth buffalo growth data from Grand Lake, Wister Reservoir, and Reelfoot Lake were only parts of pre-impoundment studies which included many fish species. The above-mentioned reports only described the growth and did not attempt to analyze it.

Smallmouth buffalo growth in Chickamauga Reservoir, a mainstream reservoir located immediately downstream from Watts Bar, should have been more similar to the growth of the species in Watts Bar than the growth of smallmouth buffalo in any of the other three areas. However, Chickamauga smallmouth buffalo were considerably smaller at ages one and two than Watts Bar fish. The Chickamauga fish were collected in 1944 and the Watts Bar fish were collected in 1962. It is possible that growth conditions have improved considerably in both reservoirs

TABLE V

TOTAL LENGTHS (MM) OF SMALLMOUTH BUFFALO

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Age	Watts Bar, Tennessee 1952	Grand Lake, Oklahoma Thomp≋on, 1950	Wister Res., Oklahoma Hall, 1951	Chickamauga Res., Tennessee Eschmeyer, Stroud, and Jones 1944	Reelfoot Lake, Tennessee Schoffman, 1944
1	111.4	154.9-218.4	127.0	96.5	284.5
2	248.7	215.9-256.5	342.9	162.6-182.9	388.6
3	344.7	264.2-304.8	408.9		439.4
4	393.0	302.3-340.4	487.7	· ·	467.4
5	429.8	337 .8- 370.8	520.7		543.6
6	460.7		571.5		594.4
7	490.0				647.7
8	522.0				
9					782.3
12					835.7

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during the eighteen year time lapse between the two collections. No other possible explanations were found for the difference.

Relative growth is percentage growth in which the increase in size in each time interval is expressed as a percentage of the size attained at the beginning of that time interval. Relative growth was calculated by dividing the annual growth increment by the total length of the fish at the beginning of that year (Table VI).

Relative growth in most species is most rapid in the younger fish and constantly declines. If fish size at hatching were considered to be zero the percentage growth at the end of the first year would be infinite. Undoubtedly smallmouth buffalo at hatching have a measurable size, but lack of data on these small fish prohibited determination of the exact relative growth for year one. Walker and Frank (1952) reported fish in year class one had reached a total length of approximately 50 mm within one or two months after hatching, indicating a measurable size at the time of hatching. Watts Bar smallmouth buffalo were concluded to correspond to the theoretical relative growth curve with a high rate of relative growth during the first year and a constantly declining rate in succeeding years.

Per cent growth per year was averaged for the first eight years of life for Watts Bar smallmouth buffalo in year classes four through nine. When these data are compared to per cent growth of smallmouth buffalo from other areas (Thompson, 1950; Hall, 1951; Eschmeyer, Stroud, and Jones, 1944; and Schoffman, 1944) some striking dissimilarities are seen. Relative growth of Watts Bar buffalo averaged 125 per cent for the second year of life and was exceeded only by Wister Reservoir buffalo which had 170 per cent growth during the same year.

TABLE VI

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PER CENT GROWTH PER YEAR

Class 2 3 4 5 6 7 8 9 10 4 126.1 39.2 5 132.7 37.9 15.4 6 130.0 37.5 18.3 9.9 7 10 26 10 10 7.6	11 12 13 14
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	
5 132.7 37.9 15.4 6 130.0 37.5 18.3 9.9 7 124.0 26.4 21.0 10.0 7.6	
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	
[124.0 JU.4 21.0 12.2 [.0	
8 126.0 39.7 21.5 13.4 8.7 6.1	
9 111.2 40.0 22.4 16.0 11.1 7.6 5.8	
1 0 9 2.5 35.9 21.3 15.2 11.8 8.9 6.7 6.6	
11 81.3 44.2 37.8 21.0 11 .8 7.2 6.0 6.5 7.2	
13 82.3 36.4 17.7 17.2 11.0 7.4 7.9 6.2 6.0	5.5 4.2
15 1 42.8 47.0 18.1 13.5 10.3 6.8 6.2 6.0 3.3	2.1 2.1 2.0 1.0

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Grand Lake, Reelfoot, and Chickamauga buffalo had 27, 36, and 82 per cent growth respectively during their second year. Watts Bar fish had 39 per cent growth during their third year which exceeded that of all the other areas. Grand Lake buffalo had 20 per cent, Wister 19 per cent, and Reelfoot 13 per cent during the third year. Respective relative growth rates for Watts Bar, Wister, Grand Lake, and Reelfoot during the fourth year were 20, 19, 13, and 6 per cent. During the fifth year they were 13, 7, 10, and 16 per cent. Wister and Reelfoot both had 10 per cent relative growth during the sixth year which exceeded the Watts Bar rate of 9 per cent. During the seventh year Watts Bar smallmouth buffalo had 7 per cent relative growth which was exceeded by Reelfoot's 9 per cent. During the eighth year Watts Bar fish had a rate of 6 per cent. Variations in relative growth rates for smallmouth buffalo from different areas in the third through eighth years of life indicate extrinsic factors, such as habitat changes or variations in food availability through changing population densities, are influencing the increase in size.

Instantaneous growth rate is the natural logarithm of the ratio of final size to initial size for a unit of time, usually one year. Instantaneous growth rates for Watts Bar smallmouth buffalo were computed from all the calculated average total lengths of all fish in each age group for each previous year. Annual instantaneous growth rates (Table VII) indicate the highest value for any year occurred in the 1-2 year interval in year class fifteen. The rates then steadily declined to a low in the eleventh year class. The data for year classes ten through fifteen cannot be considered valid because of the low

TABLE VII

ANNUAL INSTANTANEOUS GROWTH RATES

Year						Time In	verval	(year)					
Class	1-2	2-3	3-4	4 - 5	5 - 6	6-7	7 - 8	8-9_	9-10	10-11	11-12	12 -1 3	13-14
4	0.815	0.329											
5	0.846	0.322	0.140										
6	0.833	0.322	0.166	0.095									
7	0.807	0.308	0.191	0.113	0.077								
8	0.751	0.337	0.199	0.122	0.086	0.058							
9	0.747	0.337	0.199	0.148	0.104	0.077	0.058						
10	0.658	0.308	0.191	0.140	0.113	0.086	0.068	0.068					
11	0.593	0.365	0.322	0.191	0.113	0.068	0.058	0.068	0.068				
13	0.599	0.308	0.166	0.157	0.104	0.068	0.077	0.058	0.058	0.058	0.039		
15	0.888	0.385	0.166	0.131	0.095	0.068	0.058	0,058	0.030	0.020	0.020	0.020	0.010

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ω Ø number of individuals in the samples. However, there was a constant rise in the first year's annual instantaneous growth rate from year class nine through year class five, followed by a slight decline in year class four. The annual instantaneous rates of growth for these year classes in the second and succeeding years do not appear to have followed a particular pattern, but rather to have fluctuated from year to year with the variations in environmental conditions. Instantaneous growth rates also were calculated on a monthly basis, but the data were inconclusive because of the slight differences in rates.

Growth data on Watts Bar smallmouth buffalo were compared to the growth of buffalo in Wisconsin (Frey and Pedracine, 1938). Comparisons were complicated by the fact that the Wisconsin data included the largemouth buffalo, <u>Ictiobus cyprinella</u> (Valenciennes), in small numbers with about equal numbers of smallmouth buffalo and black buffalo, <u>I. niger</u> (Rafinesque). Wisconsin buffalo were found to have the most growth during their second year of life. The Watts Bar buffalo population also has the most growth during their second year. These data suggest that smallmouth buffalo characteristically have a higher absolute growth rate during their second year of life, which may be the result of a change in food habits after the first year of life.

Watts Bar buffalo averaged 5 mm less than the Wisconsin fish at the end of the first year, but exceeded the Wisconsin buffalo by 12 mm in the second year, 32 mm in the third, 18 mm in the fourth, and 4 mm in the fifth. Watts Bar buffalo were 8 mm shorter than the Wisconsin fish in year class six. Year class seven Watts Bar fish averaged 19 mm

longer than Wisconsin buffalo. The Wisconsin collection was made up of large numbers of fish in year classes two through four, whereas, the Watts Bar collections were larger for year classes five through seven.

Large numbers of Wisconsin buffalo were found within a single age group which suggests a dominant year class. There also was evidence that the Wisconsin fish had cycles of abundance with good seasons coming every third year. Watts Bar data gave no indication of dominant year classes or a cyclic population.

Lee (1912) reported that estimated fish growth in earlier years of life, as determined from scales of the older fish, often was less than the observed growth. This observation, known as Rosa Lee's phenomenon, has been accepted as a true characteristic of some fish populations (Hile, 1936). A comparison was made of absolute growth in early years for year classes six through fifteen which were assumed to be equally vulnerable to the commercial fishing. There was a steady decrease in the calculated total lengths in early years from year class six through year class nine, but up and down fluctuations followed through year class fifteen. These fluctuations tend to preclude the presence of Lee's phenomenon in the Watts Bar smallmouth buffalo population. However, data on year classes ten through fifteen are questionable because of small numbers of fish of these ages in the sample. Data on year classes six through nine only suggest the presence of Lee's phenomenon. In order to adequately test for the presence of this phenomenon samples of the same year class should be taken in several successive seasons to avoid possible bias introduced by differing growth

rates in different years. Collections in this study were limited to one year.

The term growth compensation has been applied to a phenomenon in fish species where individuals that had grown rapidly in early life were approached in size in succeeding years by individuals which had a relatively slow growth rate in their early years. Growth compensation apparently is produced by a change in the relative rate of increase among the larger and smaller fish in any age group. Scott (1949) pointed out that growth compensation is associated with a decrease in the average yearly increment. Inspection of the average annual growth increments of Watts Bar smallmouth buffalo (Figure 8, page 33) revealed that there was a complete reversal in the relative position of the annual growth increments for year classes four through nine beginning during the fourth year of life and continuing through the fifth year. This reversal indicates that the large fish which had grown rapidly during their first three years of life start slowing down in their growth rate during their fourth year of life and that the small fish with a slow initial growth rate begin to grow at a relatively faster rate. Growth compensation does exist in the Watts Bar smallmouth buffalo population.

CHAPTER VI

STABLE AND RADIOCHEMICAL COMPOSITION OF FISH TISSUES

A. Stable Chemistry

A composite sample was made up of approximately four scales from each individual in the June collection of Watts Bar smallmouth buffalo. This sample was oven dried at 104° C.

An ash sample was sent to the Spectrochemical Laboratory of the Oak Ridge National Laboratory Analytical Chemistry Division for spectrographic analysis. The values reported (Table VIII) were visual estimates taken from a standard plate and using a common graphite matrix. These values are to be interpreted as approximations and are within the range of 1/2 to 2 times the actual concentrations.

One ash sample was put into solution by alternate addition of concentrated HCL, 30 per cent H_2O_2 , concentrated HNO₃, and O.1N HCL, with each step being preceded by complete evaporation. The sample finally was brought to twenty-five milliliter volume with distilled water. This sample was analyzed by flame spectrophotometry by the Oak Ridge National Laboratory Analytical Chemistry Division. Results of these stable chemical analyses are given in Table IX, page 45.

Smallmouth buffalo scales have a mineral residue content of 46.05 per cent by weight. Moisture content of the scales was not determined. Calcium was by far the most abundant element amounting to 0.142 mg/g fresh weight of the scale. There followed in decreasing abundance: sodium, potassium, manganese, zirconium, iron, aluminum, lead, silicon,

TABLE VIII

SPECTROGRAPHIC ANALYSIS FOR STABLE ISOTOPES IN FISH SCALES

Element	Ash Content (ppm)
Sodium	5000 - 10000
Potassium	500 - 1000
Manganese	200 - 300
Zirconium	Less than 200
Iron	50 - 100 ·
Aluminum	20 - 100
Lead	Less than 100
Silicon	20 - 50
Cobalt	Less than 50
Chromium	Less than 50
Tin	Less than 50
Zinc	Less than 50
Molybdenum	Less than 50
Nickel	Less than 50
Rubidium	10 - 20
Strontium	10 - 20
Titanium	Less than 20
Vanadium	Less than 20
Boron	5 - 10
Copper	Trace - 10
Lithium	1 - 5
Silver	Traces

Element	Ash Content (ppm)
Strontium	. 266
Calcium	308,000
Potassium	1,720
Sodium	9,160
Cesium	1
Rubidium	1

FLAME SPECTROPHOTOMETRY ANALYSIS FOR STABLE ISOTOPES IN FISH SCALES

cobalt, chromium, tin, zinc, molybdenum, nickel, strontium, rubidium, cesium, titanium, vanadium, boron, copper, lithium, and silver. A comparison of ash content of strontium (0.266 mg/g) to that of calcium (308 mg/g) shows a stable strontium-calcium ratio of 0.394 x 10^{-3} in fich ccales.

Van Oosten (1957) summarized data on fish scale analyses and reported that fish scales were composed of 41 to 84 per cent organic protein and up to 59 per cent mineral residue in air dry matter. The moisture content of menhaden scales was 20.6 per cent, organic matter content 46.8 per cent, and mineral ash content 32.6 per cent. Chemical compounds and elements present were mainly $Ca_3(PO_4)$ and $CaCO_3$ with lesser amounts of $Mg_3(PO_4)_2$, CaF_2 , Na_2CO_3 , NaCl, Fe, S, As, CaO, MgO, P_2O_5 , and CO_2 .

TABLE IX

Results of stable chemical analyses of smallmouth buffalo scales (Tables VIII, page 44, and IX, page 45) agree with Van Oosten on the importance of calcium and the presence of magnesium, sodium, and iron in fish scales. Van Oosten did not discuss the other elements found in this study.

B. Radiochemistry

Bones and scales of smallmouth buffalo from the Clinch River were analyzed by gamma spectrometry using the ORNL Low-level Radiochemical Laboratory. Bone samples were prepared by removing the flesh, cleaning in tap water, oven drying at 104° C for twenty-four hours, and pulverizing. Scale samples were prepared by scrubbing them in tap water to remove epidermal tissues and drying at 104° C for twenty-four hours. The samples were analyzed for gamma emitters; ruthenium-106, cesium-137, and cobalt-60 were found to be present (Table X).

TABLE X

		х 10 ⁻⁷ µc/	g
Tissue	Ru ¹⁰⁶	Cs ¹³⁷	co ⁶⁰
Bone		135	·
Bone		108	108
Scales	347		198

RADIOCHEMICAL COMPOSITION OF SMALLMOUTH BUFFALO BONES AND SCALES

Of the four major radionuclide contaminants in the Clinch River, strontium-90, cesium-137, cobalt-60, and ruthenium-106, only strontium-90 can be considered a bone seeker. Nelson and Griffith (1962) in analyzing white crappie from the Clinch River found an average accumulation of strontium-90 of 120 $\mu\mu$ c/g in bone. However, strontium-90 concentrations in bone were found to vary from 3.0 $\mu\mu$ c/g to 297.0 $\mu\mu$ c/g in white crappie bone. It can be assumed that strontium-90 was present in the bone and scales of smallmouth buffalo, but no analyses were made for this radionuclide.

Scales and bony tissues of fish analyzed in this study were found to contain radionuclides of ruthenium, cesium, and cobalt. These elements are not bone-seekers and it would not be expected that they should be found in large quantities in bony tissues. Analyses of other tissues probably would have revealed higher concentrations of these radionuclides, but this study was concerned only with those radionuclides accumulated in bony tissues except for strontium-90. Few of the fish taken in this study contained enough accumulated radionuclides in their scales and bones for accurate analysis.

C. Radiometric Surveys

Radiometric surveys were made of fish tissues to determine the quantity of activity from accumulated radionuclides. Scales were prepared by scrubbing them in tap water and drying at 104° C. Bones were scraped clean, scrubbed in tap water, and dried at 104° C for twenty-four hours. Gross gamma counts were made of the dried samples

using a gamma spectrometer equipped with a 3 by 3 inch sodium-iodide crystal with a 1 by 1 inch well. Gross beta counts were made of the same samples using a counter equipped with a Geiger-muller tube. A comparison of the sensitivity of these two counting methods is made in Table XI.

Beta surveys revealed the presence of accumulated radionuclides in tissues which showed no gamma activity. The high sensitivity of beta counting results from the fact that \cos^{60} , Sr^{90} -Y⁹⁰, Zr^{95} -Nb⁹⁵, Ru¹⁰⁶-Rh¹⁰⁶, Cs^{137} , and Ce^{144} -Pr¹⁴⁴ decay primarily by negative beta particle emission. Of the radionuclides found in fish scales from the Clinch River, only Zn^{65} with 98.5 per cent decay by orbital electron capture and 1.5 per cent decay by positive beta particle emission does not decay primarily by negative beta particle emission.

The primary purpose of the radiometric surveys was to determine from which fish the scales would be autoradiographed. Most of the autoradiographic exposure of No-screen X-ray film is produced by beta particles, therefore, gross beta counting was selected as the best method of screening the scales. Radiometric surveys were made with a Model D47 Gas Flow Counter manufactured by Nuclear-Chicago Company. The counter was equipped with a "Micromil" window and automatic sample changer. Results of this counting were grouped by capture location and month of capture. Frequency distribution of the counting results of all the Clinch River smallmouth buffalo appear in Figure 9. Figures 10 through 13, pages 50 through 51, show the frequency distribution of beta counts of scale samples from the Watts Bar smallmouth buffalo for the months of June through September respectively.

TABLE XI

		Gross Be	ta cpm	Gross Ga	amma cpm
Species	Capture Location	Scales	Bone	Scales	Bone
Carpsucker	White Oak Creek	45-111	0	63	0
Ťt.	11	40	184	ŏ	191
11	11	28	117	105	· _ 0
11	11	39-114	111	ó	0
11	11	23- 38	66	· 0	0
11	Watts Bar	້ 2	-	44	-
White Bass	CRM 19.0	0	0	0	0
Gizzard Shad	11	0	Ó	0	0
Sunfish Hybrid	White Oak Lake	6- 13	7	0	0
11	31	Õ	14	0	0
Flat Bullhead	11,	-	41	-	0
	tr	-	32	-	210
1f	TT	-	50	· _	0
Warmouth	11	9	10	0	0
Bluegill	t i	ĺ4	4	0	21
	11	8	10	0	0
11	11	0	16	0	0
White Crappie	White Oak Creek	0	6	0	0
	11	0	27	0	0
11	11	0	2	0	0
11	17	0	6	0	0
ff .	11	0	0	0	0
f #	White Oak Lake	0	8	0	0
Black Crappie	White Oak Creek	0	6-10	0	0
Smallmouth Buffalo	11	43	200	55	0
11	11	9 5	208	Ō	0
н.	11	75	132	0	0
11	Watts Bar	0	-	31	-
Yellow Bullhead	White Oak Creek	-	55	-	0
11	11	-	21	-	0
Channel Catfish	11	-	380	-	0
Golden Redhorse	. 11	4	5	0	0
11	**	2	6	0	0
11	11	2	6	0	0
Goldfish	White Uak Lake	21- 37	58	-	595
Carp	CRM 20.0	4- 5	0.	6	0
ū	11	5	0	0	0
Largemouth Bass	White Oak Creek	Ó	0	0	0
5	· · ·		-		ŕ

COMPARISON OF BETA AND GAMMA SURVEYS OF FISH TISSUES







Fig. 10. Frequency Distribution of Gross Beta Counts of Scales from 509 Watts Bar Smallmouth Buffalo Caught in June 1962.



Fig. 11. Frequency Distribution of Gross Beta Counts of Scalcs from 293 Watts Bar Smallmouth Buffalo Caught in July 1962.



Fig. 12. Frequency Distribution of Gross Beta Counts of Scales from 257 Watts Bar Smallmouth Buffalo Caught in August 1962.



Fig. 13. Frequency Distribution of Gross Beta Counts of Scales from 212 Watts Bar Smallmouth Buffalo Caught in September 1962.

Thirty-two scale samples and three background counts were in each counting group. A preset count of one hundred was reached for each sample and background. The background varied from day to day by as much as two counts per minute. Counting data were converted to counts per minute. The highest background measurement in each counting group was used as the background for that particular group. When the counts per minute for any single scale exceeded the highest background for that group the sample was selected for autoradiography. A total of 1,271 smallmouth buffalo scales from Watts Bar were surveyed and 342 of these individuals were selected for scale autoradiography. All of the 146 Clinch River smallmouth buffalo were scale autoradiographed.

A comparison was made of the number of individuals in each monthly sample of smallmouth buffalo from Watts Bar Reservoir which exceeded the average background for that counting group. In the June group from Watts Bar 49.9 per cent of the fish exceeded the average background. The percentage increased in the July group to 51.8. In August the percentage again increased to 59.2. The September group was the highest with 61.4 per cent of the samples exceeding the average background for the group. This may mean that the radionuclide content of smallmouth buffalo scales in Watts Bar Reservoir increased during the summer of 1962, but a thorough investigation is needed to test this supposition. Smallmouth buffalo from the Clinch River would be expected to have a higher percentage exceeding the average background because the group is much closer to the source of contamination. Of all the smallmouth buffalo taken from the Clinch River in 1961 and 1962, 60.4 per cent of the samples exceeded the average background for the group.

Comparison of the counting results was questioned because of operational difficulties encountered during counting the samples. The "Micromil" window of the gas flow counter was damaged and had to be replaced with an aluminum foil window (1 mg/cm^2) . This changed the efficiency of the counter and caused a noncorrectable variation in background readings. With the "Micromil" window the average background was 12.66 ± 3.245 cpm at the 95 per cent level of significance. However, with the aluminum foil window the average background was 13.97 ± 0.965 cpm at the same level of significance. When the scale counts are at

such a low level that few exceed background, the confidence interval becomes critical and must be very exact for the comparisons to have meaning.

D. Scale Autoradiography

1. Scale cleaning and mounting

Several different methods were tested for cleaning the scales to remove epidermal tissues. Scales were placed in a solution of pepsin and HCL at various concentrations to digest the epidermis. This method proved to be unsatisfactory because there was some breakdown of the bony structures when the solution was highly acid. Solutions of low acidity had no apparent advantage over tap water in removing the epidermis. Scales were soaked in tap water for several hours and then scrubbed by hand. This method was effective, but too time consuming.

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The most efficient method of cleaning scales was found to be placing them in tap water and allowing the epidermal tissues to decay at room temperature. During this process the scales were placed on a shaker which provided continuous, slow agitation. Usually less than two weeks were required for the epidermis to disintegrate. The scales then were rinsed several times in tap water. Then they were placed between sheets of blotting paper and weighted and allowed to dry at room temperatures for about two to three weeks. This method was adequate for removing the epidermis and flattening the scales. However, some radioactivity was lost into the water during soaking. Origin of the radioactivity was not determined. It is probable that most came from the radionuclides within the epidermis, rather than from the bony parts of the scales.

Dried, flattened scales were mounted on glass microscope slides for autoradiographic exposure. The inner or fibrillary plate surface was fixed to the glass. Fish scales have the shape of flattened cones and have a tendency to bend or buckle away from the slide when the cement dries. Several different types of cement were tested in fixing the scales to the slides. The most successful method of mounting the scales involved the use of subbed slides. Dipping the slide into a subbing solution coats the surface of the slide with a substance which is more easily adhered to than clean glass. Slides were subbed in a solution of five grams of gelatin, one-half gram of chrome alum $(Cr_2(SO_h)_3)$ and 1,000 cc of distilled water. Slides were dried for twenty-four hours after subbing before scales were mounted on them. Scales were held on the slides by a small drop of Eastman 910 cement and pressed flat for several minutes, then allowed to air dry at room temperature. This method generally was successful, but in several instances the scales buckled away from the slide during drying. The slides were labeled and mounted on 8 by 10 inch sheets of carboard for exposure.

2. Exposure and development

Scales of sufficient activity were exposed in 10 by 12 inch cassettes and weighted to prevent the slides from shifting position on the film. The outer sculptured surface of the scale was placed toward the film. At first a layer of Saran Wrap was placed between the scale and the film to prevent any chemical reactions from moisture diffusing out of the scale. Saran Wrap effectively prevented any moisture from reaching the surface of the film from the scale. However, sufficient

drying eliminated the need for the protective layer between the scale and film.

Several different types of autoradiographic film were tested to find the fastest and clearest method. NTB-2 and NTB-3 liquid emulsions were painted directly on the outer surface of mounted scales. Liquid emulsions were highly unsatisfactory because the scales bent and buckled away from the slide under the shrinking influence of the drying emulsion. Buckling occurred during development and fixing also. This caused some difficulty in the preparation of permanent slides. Distortion caused by the scale buckling rendered the autoradiograms unreadable.

Stripping film was placed directly on the sculptured surface of the scales which were mounted on glass slides. Type AR .10 and AR .50 stripping films were tested. The AR .10 was unsatisfactory because of its low sensitivity which required a lengthy exposure period at the low activity exhibited by most fish scales. Type AR .50 stripping film which is approximately ten times more sensitive than AR .10 proved to be partially satisfactory and was used for preliminary analyses and for the laboratory tagging experiment. Both types of stripping film caused considerable buckling of the scales during drying and development and were not suitable for permanent records.

No-screen X-ray film was the best material for scale autoradiography. However, this film is not particularly sensitive. A total exposure of about 2,000,000 counts over background was needed to produce a readable pattern or image. Exposure times for the fish scales ranged from ten days to nine months. Autoradiograms exposed for a long period of time were expected to show evidence of some exposure from

naturally occurring radionuclides. Robeck, Henderson, and Palange (1954) reported that the natural radioactivity in fresh water is extremely low and that the radioactivity in aquatic organisms is at or below 2 x 10^{-2} dpm/g. There were no obvious differences in the number or distribution of exposed photographic grains in the background areas between scales and the number or distribution of exposed grains in those areas of scales where there were no accumulations of radionuclides.

Development and fixing methods were the same for all films used. They were developed for five to ten minutes in Kodak D-19 Developer at 20° C. As soon as the image started to appear on the film the development was stopped by placing the film in tap water at 20° C for about thirty seconds. Leaving the film too long in the developer resulted in over-development causing the background areas of the film to become darkened. Film was cleared and fixed immediately in DuPont X-ray Fixer and Hardener at 20° C for at least ten minutes. Film was then washed for at least fifteen minutes in running tap water and dried in a dustfree drier with circulating air at room temperature. Photographic negatives were made of the scale autoradiograms and these negatives were used in producing prints for permanent records.

Prosser, et al. (1945) autoradiographed scales of goldfish which had been immersed in a pond water solution of strontium-89 at 0.6 μ c/ml for six hours. Examination of these scales revealed concentric rings and greater activity in the thick area at the base of the scale than in the thinner areas. It was concluded that the concentric bands did not correspond to growth rings, but rather to areas of different thickness.

Micrometer measurements were made of the thickness of smallmouth buffalo scales in this study. All scales were found to increase in thickness from the margin to the focus. Many autoradiographed scales showed the greatest ratioactivity was in the thin marginal areas. In comparing results of this study to those of Prosser, <u>et al.</u>, it is significant to note that fish in this study had lived in contaminated areas and actually incorporated radionuclides into structural material in the scales, whereas, fish in the other study were simply immersed in the tagged solution for a few hours where it was impossible for growth to occur. The presence of any radionuclides in the goldfish scales must have been due to imperfect cleaning methods prior to autoradiography.

The first step taken when fish scales containing radionuclide accumulations were found to produce autoradiographic patterns of concentric circles was to determine if all the scales from an individual would produce the same pattern. One smallmouth buffalo was taken from White Oak Creek with scales which counted over 440 beta counts over background. All the scales, more than one thousand, were removed from one side of this fish and labeled on the inner surface with india ink. These scales were cleaned, pressed, mounted in order, and autoradiographed with No-screen X-ray film.

Subsequent development of the film showed that all the normal scales of the fish had the same pattern of concentric circles (Figure $1^{j_{+}}$). Some regenerated scales produced an exposure over the entire regenerated portion of the scale with the concentric circle pattern being resumed at the point where normal growth resumed. Some of the regenerated scales produced no exposure at all. It was concluded that scales



Fig. 14. Autoradiogram of Scales from a Single Smallmouth Buffalo from White Oak Creek.

which grew while the fish was in a contaminated area accumulated radionuclides in the region of scale growth. Scales which were regenerated while the animal was in a contaminated area contained accumulated radionuclides in the regenerated portion of the scale. However, scales which were regenerated while the animal was in a noncontaminated area exhibited no accumulated radionuclides in the regenerated portion. These data tend to deny the translocation of radionuclides from one portion of the scale to another.

Autoradiographic examination of fish scales was established as a valid method of determining the distribution of accumulated radionuclides in the bony surface layer of the scale. There are several problems yet to be solved in the perfection of this technique. Most important is the availability of a film sensitive enough for the low activity in the scale to produce an exposure within two or three weeks. Films currently in use require up to 2,000,000 counts over background irradiation to produce an adequate image. On this basis, the most active scales would produce a readable image on No-screen X-ray film in three to seven days. However, this high degree of activity was unusual and the most active scales from the Watts Bar collection exhibited only 21.5 beta cpm, which required over two months of exposure time to produce an acceptable image. Scales counting less than 20 beta cpm produced no readable images because the time required for exposure was so long that natural background irradiation and chemical reactions produced fogging of the film and eliminated the scale image.

E. Cesium-134 in Scale Tagging

In the early autoradiographic examinations of scales from fish caught in contaminated areas the patterns of concentric circles led to the idea that radionuclides are accumulated in scale structures as growth occurs. If these rings could be identified with residence in a contaminated area it would be possible by back calculation to trace the movements of fish in relation to contaminated and noncontaminated areas. A laboratory experiment was designed to test the feasibility of tagging fish scales with radionuclides and using the accumulations to identify the fish.

Bluegill, <u>Lepomis</u> <u>macrochirus</u> Rafinesque, and warmouth, <u>Chaenobryttus</u> coronarius (Bartram), were selected for the tagging attempt because of

their small size and ease of feeding and maintaining in aquaria. Fish were maintained individually in ten gallon aquaria which were submerged in a water bath for temperature control. Aeration was provided to each aquarium. Periodic weights, measurements, and whole body gamma counts of the fish were taken. Three scales were taken from each fish at the start of the experiment and periodically during the course of the experiment. Mounted scales were autoradiographed with Kodak AR .50 stripping film. The fish were fed earthworms. Worms were washed in tap water prior to tagging. They were tagged by placing them in 25 ml of a solution of cesium-134 for three to eight hours at a concentration of approximately 1.1 x 10^6 dpm.

Fish were divided into three groups. Experimental fish from noncontaminated areas were fed only tagged food during the experiment. Reciprocal fish from contaminated areas were fed only noncontaminated food. Control fish from noncontaminated areas received noncontaminated food.

The tagging experiment was only partially successful. There were two reasons for lack of success. Growth was evident in only one of the fish, therefore, the others did not deposit new scale material. Cesium-134 is not a bone-seeker and only a small percentage of the accumulated radionuclide was deposited in the scales of the fish that did grow.

Analyses of autoradiograms of scales from the warmouth which grew showed that fish scales can be marked with an accumulation of radionuclides for use in identifying the animal (Figure 15). The margin of the scale appears in the left side of the picture. A narrow line of exposed photographic grains was evident extending from the top to the


Fig. 15. Autoradiogram of the Posterior Margin of a Scale from a Warmouth Tagged with Cesium-134.

bottom of the picture along the margin of the scale in the area where growth has taken place. This line of exposed grains indicated the presence of accumulated cesium-134. Widely scattered exposed photographic grains were observed over the entire surface of the autoradiogram. These were caused by background irradiation. There were no radionuclide accumulations in scales of the experimental fish which were fed tagged food, but did not grow. The lack of accumulation in these scales indicates that radionuclides are accumulated only in those portions of the scales which actually are grown in the contaminated area.

Experimental fish which had been fed only tagged food were dissected upon completion of the experiment. Tissues were separated and oven dried for twenty-four hours at 10^{10} C. Samples were counted in 25 by 150 mm glass tubes in a gamma spectrometer equipped with a 3 by 3 inch sodium-iodide well detector. The counter was calibrated with cesium-137. All samples and backgrounds were counted for five minutes each in the 0.555 to 0.844 Mev portion of the gamma spectrum where cesium-134 exhibits characteristic photopeaks. Results of the radio chemical analysis of these tissues are shown in Table XII.

Distribution of cesium-134 in the fish's body was compared to the work of Boroughs, Chipman, and Rice (1957) who found that an ingested dose of radiocesium in small tuna accumulated rapidly in the liver, heart, spleen, and kidneys, but was lost rapidly from these organs. Muscle, gonads, and skin continued to accumulate cesium-137 faster than they lost it. The largest accumulations of cesium-134 in this experiment were in the testes, muscle, and liver and spleen. Generally the gills, gastrointestinal tract, and eyes were intermediate. Bone, skin and scales, and fins had the lowest accumulation of cesium-134 of any tissue Lested.

TABLE XII

CESIUM-134 ACCUMULATION IN FISH TISSUES

Tissue	Cesium-134 Accumulation $(x \ 10^{-2} \ \mu c/g \ dry \ weight)$			
Bluegill				
Gills (including bony element)	- 3.20 ± 0.06			
Muscle	8.58 ± 0.04			
Testes	8.75 ± 0.42			
Bone	1.11 ± 0.02			
Gastrointestinal tract (cleaned)	1.84 ± 0.04			
Skin and scales	1.21 ± 0.01			
Liver and spleen	4.17 ± 0.08			
Warmouth				
Gills (including bony element)	1.56 ± 0.02			
Muscle	3.75 ± 0.02			
Testes	8.71 ± 0.42			
Bone	0.65 ± 0.03			
Gastrointestinal tract (cleaned)	3.21 ± 0.05			
Skin and scales	1.24 ± 0.02			
Liver and spleen	4.08 ± 0.08			
Finc	0.70 ± 0.02			
Еуөс	2.14 ± 0.05			

CHAPTER VII

DISPERSION OF SMALLMOUTH BUFFALO

A. Conventional Tagging

There are several methods of marking living fish for future recognition. Fish may be marked by mutilation, such as fin-clipping, branding, or tattooing. The most common marking method is the attachment of tags. In the tagging operations of the Radiation Ecology Section, Oak Ridge National Laboratory, Atkins type plastic tags were used. These tags were numbered and labeled for return through the TVA Fish and Game Section. The tags were attached by monofilament polyethylene line inserted through the muscles ventral to the posterior portion of the fish's dorsal fin. These tags were used in the tagging operations of 1960 and 1961.

In 1960, 347 smallmouth buffalo were tagged. There were ten tag returns from this group. Five of these returns were from commercial fishermen and five were in Radiation Ecology Section hoop nets. A total of 309 smallmouth buffalo were tagged in 1961. There were three returns from this group: two in Radiation Ecology Section hoop nets and one from commerical fishermen. Table XIII shows data on smallmouth buffalo movements as revealed by examination of tag return records. Tag returns represent 2.8 per cent of the fish tagged in 1960 and 1 per cent of those tagged in 1961.

A comparison was made of the length and weight changes between capture and recapture of rough fish species tagged during 1960 and 1961.

TABLE XIII

Tagging Date	Tagging Location	Time Lapse (Days)	Distance Moved (Miles)	Direction Moved
7-6-60	CRM 19.5	129	15.1	Downstream
7-9-60	CRM 20.8	154	16.4	**
7 - 13-60	CRM 21.8	287	1.0	11
7 - 15-60	CRM 21.8	306	2.4	**
8-10-60	CRM 21.8	365+	450.0+	11
8-15-60	CRM 21.8	259	0.7	11
8-26-60	CRM 18.5	298	4.9	11
9-7-60	CRM 17.5	109	13.1	Ħ .
9 - 14-60	CRM 17.5	275	2.9	Upstream
8-11-60	CRM 17.5	. 577	35.5	Downstream
4 - 17-61	CRM 19.4	50	0	
5 -19 - 61	CRM 20.8	38	0	
6-16-61	CRM 20.6	199	43.0	Downstream

SMALLMOUTH BUFFALO MOVEMENT AFTER TAGGING

^aThis individual moved 17.5 miles down the Clinch River and 18 miles upstream in the Tennessee River.

This comparison was made in an attempt to determine if tagging exerted a detrimental influence on the growth of individuals. Adequate data for this comparison were available on eleven fish (Table XIV).

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TABLE XIV

Tagging		Reca	Recapture	
Length (mm)	Weight (g)	Length (mm)	Weight (g)	Lapse (Days)
520	1890	500	1700	287
400	850	385	710	306
395	980	385	850	259
. 440	1280	430	1210	275
420	1040	415	1050	50
530	1930	530	2000	286
505	1560	490	1600	263
490	1410	470	1300	279
500	2090	500	1700	159
395	79Ô	390	800	305
605	2120	, 585	2000	403
	Tagg Length (mm) 520 400 395 440 420 530 505 490 500 395 605	TaggingLength (mm)Weight (g)52018904008503959804401280420104053019305051560490141050020903957906052120	TaggingRecaLength (mm)Weight (g)Length (mm)52018905004008503853959803854401280430420104041553019305305051560490490141047050020905003957903906052120585	$\begin{array}{c c c c c c c c c c c c c c c c c c c $

LENGTH AND WEIGHT CHANGES BETWEEN TAGGING AND RECAPTURE

Tag returns from the 1960 and 1961 tagging operations revealed great variations in movements of smallmouth buffalo between tagging and recapture. From a total of thirteen tag returns the fish were determined to have moved distances ranging from 0 to over 450 miles during a time lapse between tagging and recapture which ranged from 38 to 577 days. The speed of movement ranged from 0.01 to 0.22 river miles per day for the eleven fish which moved. One fish had moved 2.9 river miles upstream. One had moved 17.5 miles down the Clinch River and 18 miles upstream in the Tennessee River. The remainder of fish which moved went downstream. It is possible that capture, handling, and attachment of the tag reduced the vigor of the animal giving the fish a greater tendency to move downstream with the current rather than exerting the energy necessary to move upstream against the current. Commercial fishing operations occur in the areas downstream from the tagging area. Sampling upstream might have revealed that some of the individuals moved upstream after tagging.

The study of plants and animals is designed primarily to obtain information about their operation under natural conditions and studies of physiology or behavior of organisms held under abnormal ecological conditions may be misleading (Woodbury, 1956). When a tag is attached to a fish's body abnormal conditions are created which decidedly affect the animal's physiology and behavior. Ricker (1942) concluded that trapping, handling, removing the fins, and even the presence of a tag resulted in little or no mortality; but that the tag, presumably by interfering with feeding, vitiated estimates of populations made from recoveries of line-caught fish. Rousefell and Everhart (1953) reported that the chief drawback of the mark-recapture method of population estimates lies in the assumptions that the tagged fish do not suffer any increased mortality and that the recaptured fish are observed and recorded. Black (1957) demonstrated that some physiological difficulties were imposed on fish in the process of capturing and marking it. DeRoche (1963) presented data indicating that monel metal jaw tags on adult lake trout produced a reduction in growth rate which continued with increasing effect throughout the life of a tagged fish. Ricker (1958) reversed

his opinion that marking imposes no increased mortality on fish and reported a frequent effect of marking is extra mortality among marked fish, either as a direct result of the mark or tag, or indirectly from the exertion and handling incidental to marking operations. In either event recoveries will be too few to be representative; hence population estimates made from them will be too great and rates of exploitation will be too small.

Conventional tagging methods normally are used to determine the movement of fishes between the time of tagging and recapture. However, many instances of abnormal behavior of tagged fish have been reported. Ricker (1958) reported that tagged sunfish usually swim to the bottom and burrow into vegetation immediately after being released. This behavior might make them more apt to remain in the same area and be recaptured than untouched fish. Marking may cause less feeding or less moving and reduce the chance of being caught. Tagging of some fish resulted in increased or more erratic movement for some time.

Results of the ORNL Radiation Ecology Section tagging operations of 1960 and 1961 indicate that the presence of tags on smallmouth buffalo has a detrimental effect on the animal. Only five smallmouth buffalo tag returns were accompanied by accurate length and weight measurements. These fish had experienced length losses of from 5 to 20 mm. Four of the fish had had weight losses of from 70 to 190 g and the other had gained 10 g. The time lapse between tagging and recapture ranged from 50 to 306 days. Of a total of eleven rough fish tag returns accompanied by accurate length and weight measurements nine fish lost length and two had no length change between tagging and recapture. Seven of the eleven fish lost weight and four gained weight during the time lapse. There

were many observations of open wounds where the monofilament line passed through the dorsal muscles of the tagged fish. Such wounds undoubtedly would be a drain on the vitality of the animal.

B. Autoradiogram Analyses

Autoradiograms were made of scale samples from 146 Clinch River smallmouth buffalo. Ten of these samples (7 per cent) contained sufficient radioactivity to produce readable autoradiograms. Scale samples from 342 Watts Bar smallmouth buffalo were autoradiographed. Only one of the samples (0.3 per cent) produced a readable image. The autoradiograms were compared to impressions of the same scales in order to determine movements of the fish in and out of contaminated areas.

Smallmouth buffalo 1 (Figure 16) hatched in the spring of 1955 in a noncontaminated area. It lived until the spring of 1959, four complete seasons, in the noncontaminated area. This fish formed its fourth annulus in the spring of 1959 at a total length of 422 mm. It entered the contaminated area at the start of its fifth growing season immediately after formation of its fourth annulus. The fish was captured at CRM 21.7, 0.9 mile upstream from the mouth of White Oak Creek, on April 1, 1960. It had remained in the contaminated area since the spring of 1959 and had added 18 mm in length during that time. The fish was 440 mm long and weighed 1,222 g at capture. Scales from this specimen averaged 13.4 beta counts per minute exceeding background at capture.

Smallmouth buffalo 2 (Figure 17, page 70) hatched in the spring of 1956 in a noncontaminated area. It lived until the spring of 1960, four complete seasons, in the noncontaminated area. The fourth annulus



Fig. 16. Autoradiogram of Scales from Smallmouth Buffalo 1.



Fig. 17. Autoradiogram of Scales from Smallmouth Buffalo 2.

was formed in the spring of 1960 when the fish had a length of 365 mm. At this time the animal moved into a contaminated area and remained through its fifth growing season. The fifth annulus was formed in the spring of 1961 when the fish was 418 mm long. It remained in the contaminated area until capture in White Oak Creek on November 6, 1961. It was 440 mm long and weighed 1,345 g at capture and scale samples averaged 144 beta cpm over background. This specimen had been in a contaminated area for over one year and had added 75 mm length during this time.

Smallmouth buffalo 3 (Figure 18) hatched in the spring of 1956 in a noncontaminated area. It lived five completed years in the noncontaminated area until the spring of 1961. Just prior to annulus formation in the early spring it entered a contaminated area at a length of 412 mm.



Fig. 18. Autoradiogram of Scales from Smallmouth Buffalo 3.

This fish remained in the contaminated area from the spring of 1961 until its capture in White Oak Creek on November 13, 1961. It was 420 mm long and weighed 1,465 g at capture and its scales averaged 300 beta cpm per scale over background.

Smallmouth buffalo 4 (Figure 19) hatched in the spring of 1956 in a noncontaminated area. It lived five completed years in the noncontaminated area until the summer of 1961, at which time it moved into a contaminated area at a length of 414 mm. It remained in the contaminated area until its capture on January 23, 1962, in White Oak Creek. At capture this fish was 440 mm long and weighed 1,045 g. Its scales averaged 2 beta cpm per scale over background.



Fig. 19. Autoradiogram of Scales from Smallmouth Buffalo 4.

Smallmouth buffalo 5 (Figure 20) hatched in the spring of 1957 in a noncontaminated area. It lived three complete years in the noncontaminated area until the summer of 1960, at which time it moved into a contaminated area at a length of 304 mm. This fish remained in the contaminated area through formation of its fourth annulus, summer of 1961, and until its capture on November 13, 1961, in White Oak Creek. It was 400 mm long and weighed 975 g at capture. Scale samples averaged 205 beta cpm per scale over background.

Smallmouth buffalo 6 (Figure 21) hatched in the spring of 1957 in a noncontaminated area. In the summer of 1961, after four complete growing seasons, this fish moved into the contaminated area at a length



Fig. 20. Autoradiogram of Scales from Smallmouth Buffalo 5.



Fig. 21. Autoradiogram of Scales from Smallmouth Buffalo 6.

of 361 mm. It remained in the contaminated area until capture on May 10, 1962, in White Oak Creek. At capture it was 410 mm long and weighed 953 g. Scales averaged 8 beta cpm per scale over background.

Smallmouth buffalo 7 (Figure 22) hatched in the spring of 1956 in a noncontaminated area. It remained in the noncontaminated area for over three years. This fish moved into the contaminated area in the winter of 1959, during its fourth growing season, at a length of 359 mm. It remained in the contaminated area during formation of its fourth annulus, spring of 1960, and through formation of its fifth annulus, spring of 1961. It was captured on May 10, 1962, in White Oak Creek at a length of 520 mm and weight of 1,969 g. Scale samples averaged 82 beta cpm per scale over background.



Fig. 22. Autoradiogram of Scales from Smallmouth Buffalo 7.

Smallmouth buffalo (Figure 23) hatched in the spring of 1957 in a noncontaminated area. It entered a contaminated area immediately after formation of its second annulus, probably the spring of 1959. This fish was 271 mm long at formation of the second annulus. It remained in the contaminated area until some time during the winter of 1959-1960 when it left the contaminated area at a length of 304 mm. The animal was in a noncontaminated area until its capture on June 29, 1962, just prior to the formation of its fifth annulus. This fish was captured at CRM 16.0 at a length of 460 mm and weight of 1,410 g. Scales averaged 6 beta cpm per scale over background at capture.

Smallmouth buffalo 9 (Figure 24) hatched in the spring of 1957 in a noncontaminated area. This fish remained in the noncontaminated



Fig. 23. Autoradiogram of Scales from Smallmouth Buffalo 8.



Fig. 24. Autoradiogram of Scales from Smallmouth Buffalo 9.

area for over two years. During the winter of 1959-1960 it entered a contaminated area at a length of 294 mm. This animal remained in the contaminated area through the formation of its third annulus and until the formation of its fourth annulus. It left the contaminated area in the spring of 1961 at a length of 368 mm. This fish was caught on August 4, 1962, at approximately mile 542 in the Tennessee River. At capture it was 460 mm long and scales averaged 21.5 beta cpm per scale over background.

Smallmouth buffalo 10 (Figure 25) hatched in the spring of 1956 in a contaminated area. It remained in the contaminated area for two full growing seasons until it left in the summer of 1958. This individual moved into a noncontaminated area and remained through the summer of 1959. In the fall of 1959 it returned to the area of contamination and



Fig. 25. Autoradiogram of Scales from Smallmouth Buffalo 10.

remained for over one year, until the summer of 1961. It again left the contaminated area and remained away until its capture on December 19, 1961, at the mouth of White Oak Creek. Apparently this animal had just returned to the area of contamination at the time of its capture.

Smallmouth buffalo 11 (Figure 26) hatched in a contaminated area in the spring of 1957. It remained two full growing seasons until the summer of 1959, at which time it entered a noncontaminated area. This fish was 201 mm long at the time it left the area of contamination. It remained in the noncontaminated area for two complete growing seasons until the summer of 1961, at which time it returned to the contaminated area at a length of 326 mm. It was caught on December 28, 1961, in White Oak Creek at a length of 340 mm.



Fig. 26. Autoradiogram of Scales from Smallmouth Buffalo 11.

In defining the contaminated area only the immediate vicinity of White Oak Creek can be considered. Of the eleven smallmouth buffalo with readable autoradiograms eight were captured in White Oak Creek, one at CRM 21.7, one at CRM 16.0, and one at TRM 542, approximately fortyseven miles below the mouth of White Oak Creek. A high concentration of radionuclides is assumed to be necessary in order for an animal to accumulate sufficient quantities in the scales for autoradiogram exposure and these high concentrations are present only in White Oak Creek. One hundred and forty-six smallmouth buffalo from the Clinch River were subjected to scale autoradiography. Only ten of these fish had scales containing sufficient activity for autoradiogram exposure indicating the contaminated area is rather small. If surface area is considered as a measurement of available fish habitat, White Oak Creek (estimated surface area of five acres) comprises less than 0.02 per cent of Watts Bar Reservoir (38,660 acres) at full pool. If smallmouth buffalo were equally dispersed over the entire area of Watts Bar Reservoir approximately 0.02 per cent of the animals could be expected to enter White Oak Creek or reside there if the species were not wide ranging. One individual out of 1,271 captured from the Watts Bar area (0.08 per cent) showed autoradiographic evidence of residence in White Oak Creek. Small numbers in the sample prevent conclusions as to the percentage of the smallmouth huffalo population in Watts Bar which actually enters White Oak Creek.

Movements of individuals which were determined by autoradiographic analyses in this study can be considered accurate. However, generalizations made concerning the smallmouth buffalo population as a whole

are questionable because of the small number of autoradiographed individuals involved. Age of the individual seemed to have some influence on movement. None of the fish apparently left the area of hatching before it was two years old. The area of hatching here is defined as being either a noncontaminated or a contaminated area. The two fish hatched in a contaminated area left at the end of their second year of life. One of these (Figure 26, page 78) returned to the contaminated area at the start of its fifth year of life. The other (Figure 25, page 77) returned to the contaminated area during the fall of its fourth year of life. All the fish hatched in noncontaminated areas moved into the contaminated area no earlier than two years and no later than five years after hatching. This may mean that smallmouth buffalo are relatively sedentary for two years after hatching, then move upstream into the tributary areas, possibly maturing sexually and entering the upstream areas to spawn. Age of sexual maturity is not known for this species.

Total length of the individuals at the time of movement into or out of a contaminated area was examined. There was no apparent correlation between size and movement. Total length at the time of such movement varied from 2/1 to 422 mm.

In the eleven fish examined autoradiographically there were sixteen instances where movement occurred between noncontaminated and contaminated areas. Twelve of these moves coincided with resumption of growth at the time of annulus formation. This fact would indicate that the majority of the moves occurred during the late winter or early spring. There have been no recorded mass movements or migrations of this species in Watts Bar, the Clinch River, or anywhere else.

A large number of regenerated scales were observed in the smallmouth buffalo scale samples. Autoradiograms revealed that when scales were regenerated while the fish was in a contaminated area there was an even distribution of accumulated radionuclides over the entire regenerated portion of the scale. Figure 19, page 72) indicates the rapidity with which scales are regenerated. Autoradiograms of the three normal scales indicate that this individual was in the contaminated area from annulus formation in the spring of 1961 until its capture on January 23, 1962. During this period of time the regenerated scale was formed.

Two regenerated scales shown in Figure 16, page 70) were formed prior to the individual's entry into the contaminated area. These scales had resumed the normal growth pattern of circuli formation by the time the fish started to accumulate radionuclides, therefore, there was an accumulation only in those parts of the scale which were formed while the animal was actually in the area of contamination.

The classic concept of scale growth advanced by Creaser (1926) and Van Oosten (1957) is that growth is not equal around the entire margin of the scale at the same time, but that detached portions may be forming at the same time. These portions usually unite to form a continuous circulus. The lateral fields of the scale are limited in size by the proximity of the adjacent scales in vertical rows and the anterior field is limited by the density of the lower layers of dermis into which the scale penetrates. The position of cutting-over of the circuli which usually is in the posterior region of the lateral scale fields indicates

growth commences in the anterior field and progresses around the margin of the lateral fields, thus giving the ridges the appearance of extending from the anterior field laterally around both sides of the scale in a posterior direction.

Analyses of the autoradiograms revealed that growth of scales of year class four smallmouth buffalo and older begins in the lateral fields (Figure 16). Growth next occurs in the posterior field (Figures 19, 21, and 22) and finally, occurs around the entire margin of the scale (Figures 17, 18, 20, 23, 24, 25, and 26). These data indicate that smallmouth buffalo scales commence growth in the lateral fields, followed by growth in the posterior and anterior fields respectively. This information on the progress of scale growth is in contrast to the classic concept and may give an indication of the reason for incomplete annulus formation in the cycloid scales of some fish species.

Examination of preliminary autoradiograms of scales from several different species of fish revealed that accumulated radionuclides were evenly distributed throughout the fibrillary plate layer of the scale. This distribution was evident when scales were exposed with the lower surface of the scale next to the film. These same scales when exposed with the bony layer next to the film showed the characteristic concentric circle pattern of other scales from the same fish. One scale in Figure 23, shows a spot of exposure in the center. The "hot spot" resulted when the bony surface layer of the scale was broken allowing the underlying fibrillary plate's accumulated radionuclides to expose the film. These data suggest that the bony layer of the scale acts as a shield which prevents beta particles emitting from the radionuclides

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accumulated in the fibrillary plate from passing through to expose the film.

CHAPTER VIII

DISCUSSION

An analysis of the Watts Bar smallmouth buffalo population can be made by applying data obtained in this study to a catch curve calculated from the 1962 Watts Bar Collection (Figure 27). The catch





curve is based on the log frequency of the number of individuals in each year class in the catch plotted against age. The ascending left limb of the curve represents the age groups which are incompletely vulnerable. The descending right limb represents those year classes which were completely vulnerable. Small numbers of individuals in year classes ten and older invalidate any assumptions made in that portion of the curve.

The rate of commercial fishing on Watts Bar Reservoir has changed considerably over the past five years. Fishing was negligible during 1957 because there were no organized commercial fishing operations on Watts Bar and sport fishermen rarely take this species. Commerical fishing commenced on Watts Bar on a limited scale in 1958, when 15,687 pounds (dressed weight) of smallmouth buffalo were removed from the lake. This catch represented a catch per unit effort of 7.34 pounds/yard of net/year. In 1958 the nets used were made of 4 and 5 inch mesh which selected for larger fish than the 3 inch nets used later. Fishing pressure increased considerably in 1959, when 54,035 pounds of smallmouth buffalo were taken from the lake. This represents an increase of 345 per cent of the 1958 catch. Catch pcr unit effort was 12.01 lbs/yd/year. In 1960, 63,705 pounds of smallmouth buffalo were removed from Watts Bar. This was 118 per cent of the 1959 catch and represented a catch per unit effort of 14.16 lbs/yd/year. In 1961, 59,328 pounds were caught. This was 93 per cent of the 1960 catch and represented a catch per unit effort of 13.18 lbs/yd/year. During these years the fishing mortality was relatively constant and the smallmouth buffalo population apparently did not suffer depletion as was indicated by the catch per unit effort.

There was a heavy influx of commercial fishermen in 1962, when 161,303 pounds of smallmouth buffalo were taken from Watts Bar. This was 272 per cent of the 1961 catch. A catch per unit effort could not be determined for 1962 because of irregular fishing and varying numbers of fishermen working the lake. However, the catch during the first three months of 1963 has been considerably less than in previous years and the smallmouth buffalo population in Watts Bar apparently has been somewhat depleted by the heavy fishing pressure of 1962. If this depletion proves to be true it will become more apparent in later catches and population studies in Watts Bar.

A catch curve with a convex right limb can be produced by any one of three conditions within a population (Ricker, 1958). Continued recruitment at later years can produce a convex curve. However, data from this study indicate that recruitment is completed by age six and that there is no continuation in later years. Table II (page 25) indicates there is a definite trend toward younger fish being recruited, but an adequate examination of recruitment trends can be made only by continued samples over a period of years.

A steady increase in rate of fishing with age can produce a convex catch curve. The rate of fishing in Watts Bar could not be accurately determined from available data, but it can be assumed that the population is sampled representatively since recruitment appears to occur abruptly. There are no indications that fishing pressure increases with age for the Watts Bar smallmouth buffalo.

An increase in the rate of natural mortality with age of the fish can produce a convex catch curve. Since recruitment appears to be

abrupt and the older fish are not subjected to increased fishing pressure, it can be assumed that an increase in the rate of natural mortality as the fish become older is responsible for the convexity of the Watts Bar smallmouth buffalo catch curve.

No matter when year classes t and t-l are sampled, the ratio of their abundance is a measure of the survival rate which existed during the first year that the younger year class became vulnerable to fishing. Therefore, survival rates pertain to past years. The slope of the catch curve in any given part will represent the survival rate at the time the fish in question were being recruited into the fishery.

Data from the age distribution of Watts Bar smallmouth buffalo (Table 1, page 23) give some indication that a segmented population may exist in that reservoir. The higher percentages of scales exceeding background counts in August and September might suggest the movement of an increasing number of Clinch River smallmouth buffalo into the fishing area. However, additional investigation would be necessary to establish the Clinch River fish as a segment of the Watts Bar population.

Broad generalizations on the relative importance of the smallmouth buffalo as an accumulator of radionuclides would be speculative if based on the available data. Major radionuclides in White Oak Creek are accumulated by the species in quantities which generally have varied with the degree of exposure by residence in White Oak Creek. The body burdens should vary with the concentration in the environment of both stable and radioisotopes of the particular element, essentiality of the element, and the physical and chemical state of the element.

Smallmouth buffalo undoubtedly take up radionuclides both by ingestion and absorption, depending on the element.

When the population is considered as a whole, the smallmouth buffalo is a relatively minor accumulator of radionuclides. Only 0.08 per cent of the Watts Bar smallmouth buffalo definitely showed an accumulation by scale analyses. Approximately 6 per cent of the Clinch River smallmouth buffalo definitely showed an accumulation. Approximately 77 per cent of the White Oak Creek smallmouth buffalo contained large accumulations of radionuclides in their scales. These data emphasize the importance of distance from the source of contamination as a factor in the accumulation of radionuclides within a specific population. The small percentage of Clinch River fish showing accumulated radionuclides probably is due to the limited size of White Oak Creek, the only area where the concentration of radionuclides is great enough for accumulation to occur in measurable quantities. This limitation of habitat means that only a small percentage of the total population can remain in the contaminated area for any length of time.

A comparison was made of the total length of smallmouth buffalo which had resided in contaminated areas to the total length range of Watts Bar smallmouth buffalo. Watts Bar fish in year class five ranged from 420 mm to 540 mm in total length. Year class six fish ranged from 400 mm to 570 mm. Evidently, net selectivity prevented the capture of the smaller year class five fish. There were five (5) fish of year class five from the contaminated areas with total lengths of 340, 400, 410, 440, and 460 mm. All these fish would fall well within the range for their year class with the possible exception of the flsh measuring

340 mm. This smaller fish was one of those which had hatched in the contaminated area, left the area for two years, and returned to the contaminated area approximately six months prior to capture. There were five (5) year class six fish from the contaminated areas with total lengths of 420, 440, 440, 460, and 520 mm. All these fish fell well within the total length range for their year class. These data tend to suggest that growth of the smallmouth buffalo is not affected by periodic residence in an area contaminated with radionuclide wastes.

The fact that fish have definite patterns of radionuclide accumulation in their scales results in the possibility of a new technique for studying populations. Identification of resident and transient individuals in a population has long been a problem. Capture-recapture methods of population estimates have no provision for separating these population segments into their relative numbers. When capture-recapture estimates are made there is always the possibility that transient individuals are caught and tagged. When these are released they resume their movements and may leave the area. This loss of tagged individuals from a study area can result in bias causing the population to be overestimated.

When individuals reside in a contaminated area they have definite patterns of radionuclide accumulation in their scales. All the members of any particular year class would exhibit the same pattern. These patterns can be used as identifying marks.

Autoradiographic examination of radionuclide accumulation patterns in fish scales can be used in conjunction with capture-recapture estimates of population estimates. This application would follow a definite series of steps: (1) The selection of an area of study would

be limited to an area where an adequate concentration of radionuclides existed for the resident individuals to accumulate them in the scales. (2) The area should be effectively blocked off with nets to prevent the escape of tagged individuals during the capture-recapture phase of the study. (3) A conventional capture-recapture estimate should be made of the total number of fish within the area. This method is based on the assumption that the ratio of the number of fish captured and marked during the first collection to the total number of fish in the area is the same as the ratio of marked recaptures to the total catch during the second collection. (4) Scale samples would be taken from all individuals in both collections. The scales would be radiometrically surveyed and those with sufficient activity would be autoradiographed. Autoradiographs would be analyzed to identify the resident individuals in the sample. Then, only the numbers of resident individuals from the capture-recapture operation would be considered in estimating the size of the resident population. The number of transient individuals within the area at the time of study also could be estimated from the numbers of nonresident fish.

Tagging of fish with radionuclides is a definite possibility at the present time. Large numbers of fish could be tagged in holding ponds with little effort and later released into natural habitats for population studies. Systematic use of scale autoradiography could be used in identifying these tagged individuals. Scales were observed to regenerate rapidly and to accumulate large quantities of radionuclides during regeneration in contaminated areas. Removal of a key scale or small group of scales from all individuals to be tagged, then holding the fish in a pond with a sufficient concentration of bone-seeking

radionuclides, would result in a group of fish tagged in a consistent manner. Tagged fish could be identified later by removal of the key scales from all recaptured individuals and either radiometrically surveying or autoradiographing them.

In radionuclide tagging, selection of the radionuclide is of primary importance. Effective half-life (T_{eff}) of the radionuclide must be considered. The T_{eff} of an element is the time required for the radioactive element fixed in tissue of the animal's body to be diminished 50 per cent as a result of the combined action of radioactive decay and biological elimination:

$$\mathbf{T}_{eff} = \frac{(\mathbf{T}_r)(\mathbf{T}_b)}{\mathbf{T}_r + \mathbf{T}_b}$$

where T_r = physical half-life, and T_b = biological half-life. Strontium-90 appears to be an excellent radionuclide for tagging purposes because of its affinity for bone and its physical half-life of 27.7 years. However, in the selection of a radionuclide for tagging the health hazards must be carefully analyzed and the application must be kept under strict control.

Any population study is biologically significant in that it increases our knowledge of the organism and the characteristics of the population. The age distribution and growth of smallmouth buffalo in Watts Bar has been described. This information is basic and may be used in conjunction with later studies of a similar nature in determining the history of this commercially important species in Watts Bar as a management tool for the regulation of this fishery.

The dispersal study is especially significant, in that an entirely new technique of study was developed and compared to a conventional tagging study. Even though the numbers of individuals involved in the study were small, considerably more data were derived from the autoradiographic analyses of scales than from the tagging returns because conventional tagging and recovery can only locate fish at single points in time while autoradiographic records of scales provide a continuously recorded history. When large numbers of fish with sufficient radionuclide accumulations in their scales are available for autoradiography, the natural dispersion of these fish without the detrimental effects of conventional tags may be determined.

CHAPTER IX

SUMMARY AND CONCLUSIONS

The smallmouth buffalo, <u>Ictiobus bubalus</u> (Rafinesque), population of Watts Bar Reservoir, Tennessee, was investigated in order to describe its age distribution, growth rates, dispersion, and importance as an accumulator of radionuclides. Measurements and scale samples were taken from commercially-caught fich and fish caught in the ORNL tagging operations. Scale impressions were analyzed for age and growth phenomena. Dispersion of smallmouth buffalo was investigated by conventional tagging methods and by autoradiographic analyses of scales. Stable and radiochemical composition of scales was determined by

spectrographic analysis, flame spectrophotometry, radiometric surveys, and gamma spectrometry.

Watts Bar smallmouth buffalo were found to correspond to the theoretical distribution for stable fish populations where large numbers are present in the younger year classes and succeeding year classes become less numerous as a result of mortality. The largest number of fish in the commerical catch was in year class six, the youngest year class which was completely vulnerable to the commercial fishing gear. No indications were found that a dominant year class existed in the Watts Bar population.

Survival rates were calculated to be 49 per cent for year class six, 35 per cent for year class seven, 26 per cent for year class eight, and 19 per cent for year class nine. Annulus formation was concluded to be prior to June for Watts Bar smallmouth buffalo. There were some indications that the Watts Bar population is made up of segments which have different growth rates associated with tributary habitat differences. Recruitment was found to be complete at age six and commercial fishing pressure was equal on all fish from year class six upward.

The calculated length-weight relationship of Clinch River smallmouth buffalo revealed that the fish had isometric growth which is characterized by an unchanging body form and specific gravity. The fish were found to increase 100 g in weight for every 1 cm increase in length for fish in excess of 31 cm total length.

Absolute growth of Watts Bar smallmouth burralo averaged 422 cm at the end of the third growth year, 441 mm for the fourth, 453 mm for the fifth, 465 mm for the sixth, 487 mm for the seventh, 522 mm for the

eighth, and 609 mm for the ninth. Absolute growth rates were found to have increased with each succeeding year for year classes nine through four probably as a result of increased food availability accompanying increased fishing pressure. Calculated annual total length increments indicated this species characteristically had the largest increment during the second year of life. This fact was confirmed by data from other study areas and may be the result of a change in food habits after the first year of life. Growth compensation was evident during the fourth and fifth years of life.

Smallmouth buffalo scales were found to have a mineral residue content of 46.05 per cent by weight. Calcium was the most abundant element amounting to 0.142 mg/g fresh weight with at least twenty-three other elements present in lesser quantities. The strontium-calcium ratio was found to be 0.394×10^{-3} in scales. Smallmouth buffalo scales were found to contain radionuclides of ruthenium, cesium, zirconium, zinc, and cobalt.

The Watts Bar smallmouth buffalo population was concluded to be of minor importance as an accumulator of radionuclides. Only 0.08 per cent of the Watts Bar population in radiometric surveys showed accumulations of artificially produced radionuclides. Samples from areas closer to the source of contamination showed greater concentrations. Approximately 6 per cent of the Clinch River smallmouth buffalo had measurable accumulations of radionuclides and White Oak Creek fish had 77 per cent.

Autoradiographic examinations of smallmouth buffalo scales revealed that radionuclides were accumulated in patterns of concentric circles. These patterns were found to be consistent in all the norma

scales from any individual and were associated with growth in a contaminated area. A new technique was proposed by which scale autoradiography could be used in conjunction with a conventional capturerecapture population estimate to divide a fish population within a contaminated area into the sedentary and mobile segments if such existed.

Scale autoradiography and conventional tagging methods were used to study the movements of Watts Bar smallmouth buffalo. Conventional methods revealed these fish traveled 0 to 450 miles during time lapses ranging from 38 to 577 days. Evidence was presented that the presence of a tag on the animal's body is detrimental, resulting in a loss of length and/or weight. This fact supported the opinions of many investigators that tagged animals suffer physiological and behavioral difficulties imposed by the presence of the tag.

Autoradiographic examinations of smallmouth buffalo scales revealed considerably more information on movements than conventional tagging methods. The movements of individuals between noncontaminated areas and White Oak Creek, the only area of considerable contamination, were determined, as well as the age and size of the fish at the time it entered or left White Oak Creek. Smallmouth buffalo were concluded to be relatively sedentary for two years after hatching, then to have moved upstream into the tributary areas. The majority of the moves occurred during the late winter or early spring, but no mass movements or migrations were recorded on Watts Bar. Growth was not affected by residence in contaminated areas.

Laboratory experiments showed that fish scales could be tagged with cesium-134 for autoradiographic identification of the tagged individual. However, much larger concentrations of the cesium-134 occurred

in the soft tissues than in the scales and bony tissues leading to the conclusion that this radionuclide was not suitable as a scale tag. Selection of a suitable radionuclide for scale tagging and methods of application were discussed.

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