LIBRARY
OF THE
UNIVERSITY
OF ILLINOIS

cop.2

NATURAL HISTORY
SURVEY

STATE OF ILLINOIS
Otto Kerner, Governor

DEPARTMENT OF REGISTRATION AND EDUCATION
William Sylvester White, Director

HYBRIDIZATION BETWEEN THREE SPECIES OF SUNFISH (*Lepomis*)

WILLIAM F. CHILDERS
GEORGE W. BENNETT

NATURAL HISTORY
SURVEY DIVISION

Harlow B. Mills, Chief
Urbana, Illinois

Biological Notes No. 46

Printed by Authority of the State of Illinois

November, 1961

Fig. 1. — The three parent species of sunfishes and three of the six possible F₁ hybrids used in the experiment reported here. The reciprocal F₁ hybrids are not shown because they are morphologically similar to those pictured.

HYBRIDIZATION BETWEEN THREE SPECIES OF SUNFISH (*Lepomis*)

William F. Childers
George W. Bennett*

American agriculturists have produced many hybrids for increasing food production or for improving the quality of food products. Corn and poultry hybrids are so commonplace that they are considered normal crops for progressive farmers. The use of other types of plant and animal hybrids on the farm is spreading, and it would be no great step for farmers to accept the idea of hybrid sunfish in their pasture ponds.

The need for a hybrid fish in pond culture relates largely to the problem of population control: many kinds of warm-water fishes reproduce so successfully that they create conditions of overpopulation and become severely stunted. Stunted populations of fishes are useless for recreation or food. A hybrid fish with a reduced reproductive potential would be a great improvement over the fishes now commonly used in warm-water ponds, particularly if it combined rapid growth to a large size with other characteristics desirable for angling.

Naturally produced hybrids of fresh-water fishes are not uncommon and have been reported in the salmon family (Salmonidae), sucker family (Catostomidae), minnow family (Cyprinidae), pike family (Esocidae), sunfish family (Centrarchidae), and some others. Moenkhaus (1911) experimentally hybridized many species of teleosts and found that in the species he tested the eggs of each species could be impregnated by the sperm of any other species. He also determined that the stage to which any given hybrid would develop was correlated with the nearness of the taxonomic relationship of the two species used. Thompson (1935) reported that within certain groups of fishes hybrids were common and ranged from a few to as high as 10 per cent of the populations.

Hybrid sunfish have received the attention of a number of biologists. Hubbs (1920) expressed the opinion that *Lepomis euryurus* McKay was the hybrid of the green sunfish, *L. cyanellus* Raf., and the pumpkinseed sunfish, *L. gibbosus* (L.). Hubbs & Hubbs (1931) validated this opinion by successfully hybridizing green and pumpkinseed sunfishes in aquaria in the laboratory. These authors (1932) also established that the name *L. ischyurus* (Jordan & Nelson) was apparently based on the hybrid of the bluegill, *L. macrochirus* Raf., and

the green sunfish. Hubbs & Hubbs (1933) reported that "aquarium-reared" and "natural" hybrid sunfishes grew more rapidly than their parent species, were predominately of the male sex, and were apparently sterile. Luce (1937) successfully produced hybrid sunfish by manually stripping eggs from ripe females and milt from ripe males into fingerbowls containing small amounts of water. After the eggs hatched, Luce raised some of these hybrids to sexual maturity in aquaria in the laboratory. Ricker (1948) and Krumholz (1950) produced large numbers of hybrids by placing adult males of the bluegill and adult females of the red-ear, *L. microlophus* (Gunther), in ponds containing no other fish. Lagler & Steinmetz (1957) produced hybrids by placing ripe adult males of one species of sunfish with ripe adult females of another species in ponds which contained no other fish (pumpkinseed males with bluegill females and bluegill males with pumpkinseed females).

The intrageneric *Lepomis* crosses investigated by the authors mentioned above and the results of these investigations are summarized in table 1. Each of the *Lepomis* hybrids except, of course, the "natural" hybrids of Hubbs & Hubbs (1933) was produced by one of three methods: (1) isolating a ripe male of one species and a ripe female of a different species in an aquarium, (2) manually stripping eggs from ripe females and milt from ripe males into fingerbowls, and (3) isolating adult males of one species and adult females of a different species in a pond containing no other fish. The environments of the hybrids in the embryonic and larval stages probably differed considerably with the method used and may have affected the viability, morphology, and rate of growth of the hybrids.

METHODS OF PRODUCING HYBRIDS

In this paper, *G* refers to green sunfish, *B* to bluegill, and *R* to red-ear sunfish. Matings between individuals of different species are designated as P_1 crosses, and the resultant hybrids are designated as F_1 hybrids, fig. 1. F_2 hybrids are those produced by mating an F_1 male with an F_1 female. The kind of fish that furnished the sperm is always given first; thus, the P_1 cross of a male bluegill and female green sunfish is designated $B \times G$, and the resultant hybrids are des-

*William F. Childers is Assistant Aquatic Biologist and George W. Bennett is Aquatic Biologist and Head of the Section of Aquatic Biology, Illinois Natural History Survey.

ignated BG F₁ hybrids; GB F₁ designates the reciprocal hybrids.

In the experiments reported here, two methods were employed in an attempt to produce each of the six possible F₁ hybrids between bluegill, green sunfish, and

each species were kept separated. Eggs from one or more ripe females of one species were stripped into a damp petri dish, fig. 3A, and milt from one or more males of another species was stripped onto the eggs. The petri dish was then shaken vigorously in order to

Table 1. — Intrageneric *Lepomis* P₁ crosses attempted experimentally by various biologists and the results of these attempts.

P ₁ Cross (Male x Female)	Reference	Result
Bluegill x green	Hubbs & Hubbs (1932)	61 raised to sexual maturity
Green x bluegill	Luce (1937)	Many raised to free-swimming stage*
Bluegill x red-ear	Ricker (1948)	Many raised to sexual maturity
	Krumholz (1950)	Many raised to sexual maturity
	Krumholz (1950)	No hybrids produced
Red-ear x bluegill	Hubbs & Hubbs (1932)	11 raised to sexual maturity
Green x pumpkinseed	Hubbs & Hubbs (1932)	41 raised to sexual maturity
Pumpkinseed x green	Luce (1937)	Some raised to sexual maturity
Pumpkinseed x orangespotted†	Luce (1937)	Some raised to sexual maturity
Orangespotted x pumpkinseed	Luce (1937)	Some raised to sexual maturity
Green x orangespotted	Hubbs & Hubbs (1932)	No spawning occurred
Pumpkinseed x bluegill	Luce (1937)	Many raised to free-swimming stage *
	Lagler & Steinmetz (1957)	Many raised to sexual maturity
Bluegill x pumpkinseed	Hubbs & Hubbs (1932)	No spawning occurred
	Luce (1937)	Many raised to free-swimming stage*
	Lagler & Steinmetz (1957)	Many raised to sexual maturity
Longear‡ x green	Hubbs & Hubbs (1932)	Spawning occurred, but all eggs fungused
Green x longear	Hubbs & Hubbs (1932)	No spawning occurred
Pumpkinseed x longear	Hubbs & Hubbs (1932)	No spawning occurred
Longear x pumpkinseed	Hubbs & Hubbs (1932)	No spawning occurred
Longear x bluegill	Hubbs & Hubbs (1932)	No spawning occurred
Bluegill x longear	Hubbs & Hubbs (1932)	No spawning occurred

*Information from author.

†Orangespotted sunfish, *L. humilis* (Girard).

‡Longear sunfish, *L. megalotis* (Raf.)

red-ear sunfish. Each P₁ cross was made in the laboratory by using Luce's method (referred to hereafter as *laboratory cross*) and each was arranged for in outdoor ponds by placing males of one species with females of another species (referred to hereafter as *field cross*).

Adults used in making the P₁ crosses were obtained from several lakes and ponds in central Illinois, fig. 2. Bluegills were obtained from Lake Italy, Fairmount Quarry, Fairmount; from Farmer City Country Club Pond, Farmer City; and from Big Pond, Utterback's Farm, 5 miles southeast of Gibson City. Green sunfish were obtained from Allerton Lake, 4-H Memorial Camp of the University of Illinois at Robert Allerton Park near Monticello, and from Pond E2, Fairmount Quarry, Fairmount. Red-ear sunfish were obtained from Taylor's Pond, Taylor's Farm, 3 miles southwest of Fairmount; from Lake Italy; and from Big Pond.

Laboratory Crosses. — Ripe males and females of the bluegill, green sunfish, and red-ear sunfish in the lakes and ponds named above were trapped and moved into aquaria in the laboratory. Males and females of

mix milt and eggs. Two minutes were allowed for fertilization to take place, and then 200 to 300 fertilized eggs, fig. 3B, were poured into each of a number of clean petri dishes filled with aged tap water. As soon as the egg fluid, which had prevented the eggs from sticking, became diluted, the eggs adhered to the bottoms of the petri dishes. The fertilized eggs or zygotes were left undisturbed for approximately 15 minutes to allow them to become water hardened. The zygotes were then washed by lowering and raising the petri dishes four or five times in a large beaker filled with aged tap water. After the zygotes had been washed, enough water was added to each petri dish to cover the zygotes. This water was poured off and replaced approximately six times each day during the incubation period. After the embryos hatched, the larvae were placed in 10-gallon aquaria. The water in these aquaria was kept in constant gentle motion by releasing air from air stones placed close to the bottom. As soon as the larvae became free-swimming fry, they were placed in ponds containing no other fish and held for one or more growing seasons.

Fig. 2. — Hardware cloth traps used to collect parent species from central Illinois lakes and ponds. These traps were used also to sample hybrid populations. *A*, traps are shown here being assembled. *B*, a trap is being set in a small gravel pit pond.

Fig. 3. — Procedures used in making laboratory crosses. *A*, eggs from a female red-ear sunfish are being stripped into a damp petri dish, where they are immediately fertilized with sperm from a male bluegill. *B*, after the fertilized eggs have been allowed to set for 2 minutes, they are placed in other petri dishes; they adhere to the bottoms of the dishes.

Field Crosses. — Each of 16 small ponds (0.02 to 0.9 acre) containing no fish was stocked with sexually mature fish: females of one species and males of another, table 2. At frequent intervals, the ponds were checked, and any nests that were found were examined to see if they contained eggs or fry. The ponds were also seined and, when young F_1 hybrids were present, some of the hybrids were removed and used for stocking other ponds. F_1 hybrid sunfishes produced by field crosses, like those produced by laboratory crosses, were held in ponds for one or more growing seasons.

RESULTS OF HYBRIDIZATION

In this paper, direct genetic isolation refers to species isolation resulting from incompatibility between the gametes of different species, while indirect genetic isolation refers to species isolation resulting from such differences as time of spawning, habitat requirements for spawning, and mating behavior patterns.

Laboratory Crosses. — Several thousand free-swimming F_1 hybrids were produced in the laboratory from each of the six possible P_1 crosses; these hybrids

were proof that direct genetic isolation did not exist between the bluegill, green sunfish, and red-ear sunfish. Incubation times of fertilized eggs from all P₁ crosses were similar when incubation temperatures were similar. Eggs incubated at 70 degrees F. began hatching 47 hours after fertilization, and none hatched later than 61 hours after fertilization. The larvae be-

one of these ponds became contaminated with female bluegills and the other two ponds dried up during the summer. The B x R cross has been made experimentally with success six times in Indiana (Krumholz 1950).

The R x B cross was tried in four ponds. In three of the four ponds, no hybrids were produced. In the fourth, 11 young sunfish were found when the pond was

Table 2. — Field crosses attempted by placing adult males of one species and females of another species in ponds containing no other fish. Ponds were stocked in May or early June and were censused by being drained, seined, or chemically treated the following August or September. The species used were bluegill (B), green sunfish (G), and red-ear sunfish (R).

P ₁ Cross (Male x Female)	Size of Pond (Surface Acres)	Number of Adults Used		F ₁ Hybrid Generation	Remarks
		Males	Females		
R x G	0.02	4	7	Yes	Hybrids abundant
R x G	0.04	8	3	Yes	Hybrids abundant
G x R	0.10	2	8	No	----
G x R	0.02	4	6	No	----
G x R	0.10*	8	11	No	----
G x B	0.02	4	6	Yes	Hybrids abundant
G x B	0.90	30	35	Yes	Hybrids abundant
B x G	0.02	4	5	----	Contaminated
B x G	0.03	7	10	----	Contaminated
B x R	0.10	7	6	----	Contaminated
B x R	0.02	4	6	----	Pond went dry
B x R	0.20*	17	15	----	Pond went dry
R x B	0.02	4	6	?	11 young found†
R x B	0.10*	15	10	No	----
R x B	0.10*	13	15	No	----
R x B	0.80*	16	11	No	----

*Size of pond estimated.

†The 11 young sunfish found were too small to be identified as hybrids or as young of parent species.

came free-swimming fry during the fifth or sixth day after hatching.

One group of green sunfish eggs fertilized with bluegill sperm was accidentally incubated at temperatures ranging from 70 to 95 degrees F. Many of the BG F₁ hybrids produced from this group of eggs had very short caudal peduncles. In a few of these fish, the posterior end of the caudal fin did not extend beyond the posterior edges of the soft dorsal and soft anal fins.

Field Crosses. — Hybrids were produced in 4 and possibly 5 of the 16 ponds used in the experiment reported here, table 2.

The R x G cross and the G x B cross, each cross tried in two ponds, resulted in the production of large numbers of F₁ hybrids.

The B x R cross was attempted without success in three ponds. These were all improper tests because

drained. These young sunfish were so small (0.25-0.50 inch total length) that it was impossible to identify them as hybrids. An attempt to raise them in the laboratory failed. However, circumstantial evidence indicates that they were RB F₁ hybrids. When the pond was drained, the parent fish were sexed by dissection and only male red-ears and female bluegills were found. If this pond had been contaminated with male bluegills or female red-ears, it is probable that several thousand young would have been produced.

The B x G cross was attempted in two ponds. Both ponds became contaminated with male green sunfish, and in each pond large numbers of young greens were produced. Consequently, these ponds gave no real tests of hybridization between bluegill males and green females.

The G x R cross was set up in three ponds. No

Table 3. — Sex ratios of F₁ hybrid sunfishes expressed as per cent males in individual samples. Parent species were bluegill (B), green sunfish (G), and red-ear sunfish (R); in name of hybrid, male parent is given first.

F ₁ Hybrid	Number Sexed by Dissection	Number Sexed by Observation	Total Number Sexed	Per Cent Males
GR	252	----	252	48
RG	229	228	457	70
GB	44	----	44	70
BG	142	----	142	97
BR	45	65	110	97
RB	175	38	213	100

evidence of hybridization was found in any of these ponds, even though the parent fish appeared to have had every opportunity to produce young.

SEX RATIOS OF F₁ HYBRID SUNFISH

Sex was determined for varying numbers of individuals of each of the six F₁ hybrid types. Most of the F₁ hybrids were sexed by dissection; however, some were sexed by reference to color and other dimorphisms during their reproductive period when these differences were pronounced. Table 3 lists numbers sexed by dissection and observation, and the male percentage in each sample of F₁ hybrids. Of the hybrids sexed by observation, it was possible to strip sperm or eggs from some. Most of the rest could be sexed on the basis of (1) color (male sunfish are usually darker and more vividly colored than females), (2) size of abdomen (early in the spawning season, the abdomens of female sunfishes often are greatly distended with eggs), and (3) the relative diameters of the anal and urogenital openings (during the breeding season, in the sunfish male the urogenital opening is much smaller than the anal opening, while in the female the reverse is true). If there was still doubt as to the sex of a particular individual, that individual was sexed by dissection. The accuracy of determining sex by observation during the reproductive season was sometimes tested by dissection. Tests made of the accuracy of determinations based on the characters described above revealed no errors.

Of the six F₁ hybrid types, only the GR hybrids exhibited an approximate 50:50 sex ratio. As table 3 shows, the other five F₁ hybrid types were predominantly males. Of the RG and GB hybrids, 70 per cent were males; of the BG and BR hybrids, 97 per cent were males; and of the RB hybrids, 100 per cent were males.

Hubbs & Hubbs (1933) reported that 81 per cent of 43 aquarium-reared BG F₁ hybrids they examined were males. In the study reported here, 97 per cent of 142 BG F₁ fish were males, table 3.

Ricker (1948) sexed 428 BR F₁ hybrids from field

crosses and found that 98 per cent were males. In this study, 97 per cent of 110 BR F₁ hybrids were males.

REPRODUCTIVE SUCCESS OF F₁ HYBRIDS

Each of the six types of F₁ hybrids was produced at various times during the summers (May through August) of 1956-1959 and, to demonstrate its reproductive capacity, was placed in one or more ponds containing no other fish, table 4. The males of all six types constructed and vigorously guarded nests. Hubbs (1955) noted that male F₁ hybrid sunfish constructed, fanned, and guarded their nests with unusual vigor over a prolonged period.

The F₁ hybrids RG and GB, each group with a ratio of 70 males to 30 females, produced large numbers of F₂ hybrids, table 4.

Neither the BG F₁ nor the BR F₁ hybrids, each group with a ratio of 97 males to 3 females, produced an F₂ generation in any of our ponds. In Indiana, Ricker (1948) reported that BR F₁ hybrids successfully reproduced in two ponds. It is impossible at this time to explain why our BG and BR hybrids did not reproduce. The lack of reproduction was not caused by an absence of females; in each pond a few F₁ females were captured and released unharmed. The possibility of incompatibility of mating behavior between F₁ males and females should not be overlooked as an explanation for the lack of production of an F₂ generation in our

Table 4. — Reproductive success of six F₁ hybrid types when placed in ponds containing no other fish. Parent species were bluegill (B), green sunfish (G), and red-ear sunfish (R); in name of hybrid, male parent is given first.

F ₁ Hybrid	F ₂ Generation	Remarks
GR	Yes	Only a few F ₂ hybrids produced
RG	Yes	F ₂ hybrids very abundant
GB	Yes	F ₂ hybrids very abundant
BG	No	----
BR	No*	----
RB	No	----

*Ricker (1948) reported F₂ hybrids produced in two ponds containing F₁ hybrids and no other fish.

ponds. This incompatibility may be of a type that is expressed in some environments but not in others.

The RB F₁ hybrids were all males; so it was impossible for these hybrids to produce an F₂ generation.

The GR F₁ hybrids, with an approximate 50:50 sex ratio, produced a very small F₂ generation in a gravel pit pond, fig. 4. Field observations revealed that their reproductive behavior was slightly abnormal. Males constructed nests and were extremely aggressive in guarding their territories. On two separate occasions a female was observed to approach a nest guarded by

a male. The male faced the female and challenged her by flaring his opercular flaps. The female, which was greatly enlarged with eggs, swam slowly toward the male. The male left the nest and appeared to lead the female into it. The two fish started slowly swimming in small circles over the nest, with the female closer to the center. The male maintained a position slightly in advance of the female and, by constantly turning toward her, appeared to force her to swim within the limits of the nest. Before the spawning pair had time to make more than a few complete circles, nearby males

Fig. 4. -- A 0.1-acre gravel pit pond that contained GR F₁ hybrids.

deserted their nests and rushed into the nest occupied by the spawning pair. A wild melee followed, during which males fought with each other and attempted to gain control of the female. The female hastily retreated toward deep water. A short time after the female had gone, the invading males returned to their own nests.

When the nests in this pond were examined, approximately 9 out of 10 of those containing eggs had eggs scattered outside as well as inside the nests. All of the eggs which were outside the nests appeared to be covered with fungi, and most of the eggs in the nests were similarly infected. Noninfected eggs were collected from several nests and moved into the laboratory; none of these eggs hatched.

The attacks on spawning pairs by males could limit the production of an F_1 generation in one or more of the following ways:

1. Females could be prevented from spawning until the eggs which they contained were overripe and hence nonviable.

2. There might be lack of synchronization in the release of spermatozoa and eggs. Sunfish spermatozoa are capable of fertilizing eggs for a very short period of time, probably less than 60 seconds under natural conditions, after they are released into the water.

3. The presence of large numbers of fungus-infected eggs outside of the nest could lead to the production of so many fungus spores that a high incidence of fungus-infected eggs within the nest would result.

It is interesting to note that there is a fairly close relationship between the success of certain P_1 field crosses in producing an F_1 generation and their F_1 hybrids in producing an F_2 generation. Both $R \times G$ and $G \times B$ crosses produced large numbers of F_1 hybrids each time these crosses were tried. The RG and GB hybrids produced large F_2 generations. The production of a large F_2 generation was not related to whether the F_1 hybrids were produced from field crosses or laboratory crosses, as both crosses produced large F_2 generations.

GROWTH OF F_1 HYBRID SUNFISH

Growth was recorded for each of the six types of F_1 hybrids that were produced at various times during the summers of 1956-1959 and placed in ponds containing no other fish. Varying numbers of fry had been placed in the ponds, which differed in size, depth, shape of basin, and fertility. Population densities of F_1 hybrids usually were considerably below those in ponds occupied by their parent species. Thus, no valid comparisons in growth rates could be made between the various hybrids in these ponds, or between the hybrids

and their parent species. The observed growth of one population of each hybrid sunfish type has been summarized below.

RB F_1 Hybrids. — Approximately 2,500 (1,000 per surface acre) 1-day-old free-swimming fry were placed in a 2.6-acre pond on August 14, 1956. When this pond was drained on June 11, 1957, after 10 months (about 4 of which were warm enough for fish growth), it contained 463 hybrids ranging in length from 3.4 to 4.4 inches and averaging 4.0 inches. On that date, 102 of these hybrids were placed in a 0.05-acre pond. This pond was drained on July 2, 1958, after a little more than a complete growing season, and 92 hybrids were collected; they averaged 5.3 inches and ranged from 3.8 to 5.9 inches in total length.

BR F_1 Hybrids. — Approximately 1,100 (1,200 per surface acre) 1-day-old free-swimming fry were placed in a 0.9-acre pond on May 24, 1957. On October 12, 1957, after about 4.5 growing months, hardware cloth wire traps were set in this pond, and 40 hybrids were captured. These hybrids averaged 4.7 inches and ranged in length from 3.6 to 5.4 inches. They averaged 0.082 pound in weight and ranged from 0.035 to 0.100 pound. All 40 fish were returned to the pond. On July 28, 1958 (after 1 year and 2 months), 67 hybrids, which ranged in length from 5.4 to 6.9 inches and averaged 6.4 inches, were trapped and removed from this pond. The pond was treated with rotenone on September 23, 1958, and 43 additional hybrids were collected. After about two growing seasons, these hybrids averaged 7.4 inches and ranged in length from 6.2 to 8.3 inches. Average weight was 0.35 pound.

GR F_1 Hybrids. — A 0.1-acre pond was stocked on May 17, 1957, with approximately 1,000 (10,000 per surface acre) 1-day-old free-swimming hybrid fry. One hundred twenty-eight hybrids were trapped and removed from this pond on July 7, 1958, after 1 year and 2 months. At that time these hybrids ranged in length from 3.8 to 4.9 inches and averaged 4.4 inches. On August 15, 1958, 200 additional hybrids, which ranged in length from 3.9 to 5.1 inches and averaged 4.7 inches, were removed from this pond. On July 29, 1959, when the fish were about 2 years old, the population was again sampled by trapping. Fifteen hybrids were captured and returned to the pond. These fish averaged 7.7 inches and ranged in length from 7.3 to 8.2 inches. They averaged 0.41 pound in weight and ranged from 0.30 to 0.47 pound.

RG F_1 Hybrids. — A 0.9-acre pond was stocked on June 11, 1956, with approximately 1,300 (1,400 per surface acre) free-swimming hybrid fry. On June 27, 1957, 79 hybrids, which averaged 5.0 inches and ranged in length from 4.1 to 5.8 inches, were captured in traps and then returned to the pond. During the period

May 13-16, 1958, 327 hybrids, which averaged 6.8 inches and ranged in length from 5.2 to 8.1 inches, were trapped and removed from the pond.

BG F₁ Hybrids. — Sixty-one (200 per surface acre) hybrid fry were placed in a 0.3-acre pond on July 3, 1956. Less than 3 years later, on May 8, 1959, this pond was treated with rotenone, and 10 F₁ hybrids were collected, fig. 5; they averaged 8.6 inches and

the hybrids were recaptured. After 2 growing seasons, these 44 hybrids averaged 6.9 inches and ranged in length from 5.2 to 7.5 inches.

GROWTH OF PARENT SPECIES AND F₁ HYBRIDS

Two experiments were conducted to directly compare growth rates of hybrids with those of their parent spe-

Fig. 5. — Seven of 10 BG F₁ hybrids collected from a 0.3-acre pond after approximately 3 years of growth. These hybrids averaged 8.6 inches in total length and 0.74 pound in body weight.

ranged in length from 7.2 to 9.2 inches. In weight the fish averaged 0.74 pound and ranged from 0.50 to 0.86 pound.

GB F₁ Hybrids. — On August 26, 1957, 322 (800 per surface acre) hybrids, which averaged 1.5 inches total length, were placed in a 0.4-acre pond. Wire traps were set in this pond on October 18, 1959, and 44 of

cies. In the first experiment, a pond which contained no other fish was stocked with equal numbers of BG F₁ hybrids and green sunfish of approximately similar ages and sizes. In the second experiment, another pond was stocked in a similar manner with GR F₁ hybrids, green sunfish, and red-ear sunfish. In both of these experiments the population densities of fish were con-

siderably below the carrying capacities of the ponds. Intraspecific and interspecific competitions were low, and the fish grew at rates approaching the optimum for their respective environments.

Green Sunfish vs. BG F₁ Hybrids. — On June 3, 1958, a very small (0.02 surface acre) rearing pond was stocked with 1-day-old, free-swimming, laboratory-produced green sunfish fry. Another rearing pond of the same shape and size was stocked on four different dates (May 26, June 3, June 10, and June 18, 1958) with 1-day-old, free-swimming, laboratory-produced BG F₁ hybrid fry. On July 10, 1958, both ponds were seined, and 171 green sunfish, which averaged in total length approximately 0.75 inch, and 171 BG F₁ hybrids, which averaged in total length approximately 1.0 inch, were removed from the rearing ponds and placed in an 0.8-acre gravel pit pond, fig. 6, that contained no other fish.

Ten months later (but only 4 months during which waters were warm enough for fish growth) the fish population in the gravel pit pond was censused by trapping and rotenone poisoning. All of the fish recovered were individually measured, weighed, and sexed. Statistics on this population are shown in tables 5 and 6.

The BG F₁ hybrid and green sunfish males had in-

creased in length at approximately the same rates. The hybrid males averaged 0.4 inch longer than the green sunfish males. However, as the hybrid sunfish averaged approximately one-fourth of an inch longer than the green sunfish when the pond was stocked, the length advantage of the hybrid males may not be significant. The BG hybrid sunfish males between the lengths of 4.8 and 5.7 inches weighed considerably more than the green sunfish males of similar sizes. The BG hybrids were much deeper bodied than the green sunfish.

Population density of the fish in this pond was low. At the time the experiment was terminated, the pond contained only 28 pounds of fish per acre. In the past this pond had supported considerably more than 100 pounds of sunfish per acre. Intraspecific and interspecific competitions between the fish in this pond were very low, and the fish had grown at extremely rapid rates.

The results of this experiment indicate that under low population densities there is not much difference between the growth potentials of BG F₁ hybrids and green sunfish during the first year. Under high population densities, the hybrid sunfish might grow faster than the parent types because of a superior ability to compete for food and space.

Fig. 6. — An 0.8-acre gravel pit pond that was stocked with equal numbers of green sunfish and BG F₁ hybrids of similar sizes.

Table 5. — The numbers of green sunfish and BG F₁ hybrid sunfish placed in an 0.8-acre pond and the numbers and the percentages of the original stock recovered 10 months later.

Category	Green Sunfish		BG F ₁ Hybrids	
	Number	Per Cent	Number	Per Cent
Fish used in stocking (July 10, 1958)	171	100	171	100
Fish trapped and removed (April 29-May 8, 1959)	123	72	116	68
Fish recovered during rotenone census (May 8-11, 1959)	4	2	26	15
Total fish recovered (trapping and rotenone census)	127	74	142	83
Fish unaccounted for	44	26	29	17

Table 6. — Number and weights of green sunfish and BG F₁ hybrids of various length classes recovered 10 months after release in an 0.8-acre pond.

Total Length, Inches	Green Sunfish				BG F ₁ Hybrids			
	Number of Fish	Weight			Number of Fish	Weight		
		Range, Grams	Average			Range, Grams	Average	
			Grams	Pounds			Grams	Pounds
M A L E S								
2.8-3.2	---	---	---	---	3	7-8	7	0.02
3.3-3.7	2	7-14	10	0.02	6	8-13	10	0.02
3.8-4.2	5	16-24	20	0.04	5	14-26	20	0.04
4.3-4.7	13	24-38	31	0.07	6	23-36	28	0.06
4.8-5.2	33	37-49	43	0.09	10	37-65	58	0.13
5.3-5.7	12	47-56	51	0.11	79	52-81	63	0.14
5.8-6.2	---	---	---	---	29	79-92	80	0.18
Total	65	---	---	---	138	---	---	---
Average*	---	---	39	0.08	---	---	59	0.13
F E M A L E S								
2.8-3.2	---	---	---	---	---	---	---	---
3.3-3.7	6	11-22	16	0.04	---	---	---	---
3.8-4.2	13	14-23	19	0.04	---	---	---	---
4.3-4.7	26	21-38	30	0.07	2	28-30	34	0.08
4.8-5.2	17	31-50	38	0.08	1	30	30	0.09
5.3-5.7	---	---	---	---	1	50	50	0.13
5.8-6.2	---	---	---	---	---	---	---	---
Total	62	---	---	---	4	---	---	---
Average*	---	---	28	0.06	---	---	41	0.09

*The average total length of green sunfish males was 4.9 inches, females 4.4 inches; BG F₁ hybrid males 5.3 inches, females 5.0 inches.

GR F₁ Hybrids vs. Green Sunfish and Red-Ear Sunfish. —During the period August 1-15, 1958, 200 GR F₁ hybrids averaging 4.6 inches total length, 200 green sunfish averaging 4.2 inches, and 200 red-ear sunfish averaging 3.5 inches were placed in a 1.1-acre pond that contained no other fish. During the spring of 1959 (April and May) some of these fish were taken by hook and line, some were trapped, and the rest removed by rotenone treatment of the pond. One hundred forty-nine GR F₁ hybrids, 136 green sunfish, and 127 red-ear sunfish were recovered. The numbers of fish recovered by each of the three methods of collection, the length-frequency distributions, and the range of weights are shown in tables 7, 8, and 9.

The population densities in this pond were low. When the experiment was terminated, there were 26, 21, and 14 pounds per acre of GR F₁ hybrids, green sunfish, and red-ear sunfish, respectively, in the pond; the fish ranged in length between 4.5 and 7.0 inches. This pond had in the past supported more than 150 pounds of sunfish per surface acre. Fish in this experiment grew at an exceedingly rapid rate; they increased in length about 2 inches (average) and nearly doubled their weight between August of 1958 and late May of 1959. The results of this experiment give additional support to the concept that in uncrowded ponds there is little difference in the growth rates of hybrid sunfish and their parent species.

Table 7.— Numbers of red-ear sunfish, green sunfish, and hybrids of these species placed in a 1.1-acre pond and numbers and percentages of original-stock recovered approximately 9 months later.

Category	Red-Ear Sunfish		Green Sunfish		GR F ₁ Hybrids	
	Number	Per Cent	Number	Per Cent	Number	Per Cent
Fish at stocking (August 1-15, 1958)	200	100.0	200	100.0	200	100.0
Fish removed by hook and line fishing (April 20-22, 1959)	3	1.5	24	12.0	23	11.5
Fish trapped and removed (May 19-22, 1959)	80	40.0	99	49.5	122	61.0
Fish recovered during rotenone census (May 22-25, 1959)	44	22.0	13	6.5	4	2.0
Total fish recovered	127	63.5	136	68.0	149	74.5
Fish unaccounted for	73	36.5	64	32.0	51	25.5

Table 8.— Length-frequency distribution of red-ear sunfish, green sunfish, and F₁ hybrids of these species when placed in a 1.1-acre pond, August 1-15, 1958, and when recovered May 19-23, 1959. Numbers in parentheses show average lengths.*

Total Length, Inches	Red-Ear Sunfish		Green Sunfish		GR F ₁ Hybrids	
	Number at Stocking, Aug., 1958 (3.5 Inches)	Number Recovered, May, 1959 (5.4 Inches)	Number at Stocking, Aug., 1958 (4.2 Inches)	Number Recovered, May, 1959 (6.2 Inches)	Number at Stocking, Aug., 1958 (4.6 Inches)	Number Recovered, May, 1959 (6.4 Inches)
1.8-2.2	3					
2.3-2.7	73					
2.8-3.2	23					
3.3-3.7	14		16			
3.8-4.2	31		81		4	
4.3-4.7	49	7	92		154	
4.8-5.2	7	39	11		41	
5.3-5.7		38		16	1	2
5.8-6.2		27		39		28
6.3-6.7		6		46		75
6.8-7.2				9		19

*The average length increment in the period was 1.9 inches for the red-ear sunfish, 2.0 inches for the green sunfish, and 1.8 inches for the GR F₁ hybrids.

Table 9.— Length-frequency distribution, range of body weights, and average body weights of red-ear sunfish, green sunfish, and GR F₁ hybrid sunfish that were recaptured May 19-23, 1959, from a 1.1-acre pond that had been stocked August 1-15, 1958, with 200 individuals of each kind.

Total Length, Inches	Red-Ear Sunfish				Green Sunfish				GR F ₁ Hybrids			
	Number of Fish	Weight			Number of Fish	Weight			Number of Fish	Weight		
		Range, Grams	Average			Range, Grams	Average			Range, Grams	Average	
			Grams	Pounds			Grams	Pounds			Grams	Pounds

MALES

4.3-4.7	2	25-30	28	0.06								
4.8-5.2	9	40-48	43	0.09								
5.3-5.7	25	49-74	57	0.13	6	49-64	57	0.13				
5.8-6.2	12	65-84	71	0.16	33	62-90	79	0.17	5	83-95	87	0.19
6.3-6.7	5	88-100	93	0.20	44	72-110	97	0.21	46	80-110	98	0.22
6.8-7.2					9	111-150	126	0.28	16	110-132	119	0.26

FEMALES

4.3-4.7	5	28-36	33	0.07								
4.8-5.2	30	34-48	44	0.10								
5.3-5.7	13	42-60	53	0.12	11	47-67	55	0.12	2	72-86	79	0.17
5.8-6.2	15	60-83	73	0.16	5	65-99	75	0.16	23	61-86	78	0.17
6.3-6.7	1	94	94	0.21	2	91-95	93	0.20	28	78-113	101	0.22
6.8-7.2									3	109-110	110	0.24

SUMMARY

1. Ripe eggs of bluegills, red-ear sunfish, and green sunfish were fertilized with sperm of these species in the laboratory to make each of the six possible P_1 crosses. From each of these crosses large numbers of F_1 hybrid embryos developed normally, hatched, and became active free-swimming fry. The fry were released in outdoor ponds, where they grew to sexual maturity. There appears to be no direct genetic isolation between these three species.

2. Attempts to produce hybrids naturally were made by isolating males of one species (that is, bluegill, red-ear sunfish, or green sunfish) with females of another (six possible combinations) in ponds that contained no other fish. Each of the P_1 crosses was attempted two or more times. Only the $G \times B$ and $R \times G$ crosses produced significantly large numbers of F_1 hybrids. Results of the other four P_1 crosses were negative or inconclusive.

3. Of the six F_1 hybrids produced, only the GR hybrids exhibited an approximate 50:50 sex ratio. Of the RG and GB hybrids, 70 per cent were males; of the BG and BR hybrids, 97 per cent were males; and of the RB hybrids, 100 per cent were males.

4. When placed in ponds containing no other fish, the RG F_1 and the GB F_1 hybrids produced large numbers of F_2 hybrids; the GR F_1 hybrids produced only a few F_2 hybrids. The BG, BR, and RB hybrids failed to produce an F_2 generation.

5. Varying numbers (200-10,000 per surface acre) of F_1 hybrid fry were placed in ponds containing no other fish. These ponds differed in size, depth, shape of basin, and fertility; consequently, no valid comparisons between the growth rates of the different F_1 hybrids in these ponds could be made.

6. BG F_1 hybrids and green sunfish of similar sizes and ages in equal numbers in a pond that contained no other fish increased in length at approximately the same rates. The BG hybrid sunfish males between the lengths of 4.8 and 5.7 inches weighed considerably more than the green sunfish males of similar sizes. The BG hybrids were much deeper bodied than the green sunfish. The population density of the fish in this pond was low (28 pounds per acre when the experiment was terminated), and the fish grew rapidly. The results of

the experiment in this pond indicate that under low population densities there is not much difference between the growth potentials of BG F_1 hybrids and green sunfish during the first year. Under high population densities, the hybrid sunfish might grow faster than the parent types because of a superior ability to compete for food and space.

7. GR F_1 hybrids, green sunfish, and red-ear sunfish in equal numbers in a pond that contained no other fish increased in length and weight at approximately the same rates. The population density of the fish in this pond was low (61 pounds per acre when the experiment terminated), and the fish grew rapidly. The results of the experiment in this pond give additional support to the concept that in uncrowded ponds there is little difference in the growth rates of hybrid sunfish and their parent species.

ACKNOWLEDGMENTS

The authors acknowledge the field assistance of Messrs. John C. Cralley, William Leicht, Richard Ward, Ronald G. Altig, Ronnie D. Havelka, and John Schilling in the study reported here.

Pond owners and fishing clubs who gave the use of their ponds for this study are Messrs. Henry Green, Everett E. Glasgow, David Malcolmson, H. C. Paterson, A.J. Plankenhorn, William S. Pusey, Ross Rudisill, John Taylor, William Utterback, Gene Ziegler, and G. G. Maxfield of the Fairmount Quarry Fish-Game Club, the St. Joseph Sportsmen's Club, and the Farmer City Country Club. Professor F.H. Root and other staff members of Robert Allerton Park allowed the use of certain ponds at the 4-H Memorial Camp. The authors are grateful for the co-operation of these persons and clubs.

The manuscript was read by Dr. R.W. Larimore and edited by Mr. James S. Ayars, both of the Illinois Natural History Survey.

The cover illustration, depicting a nesting male green sunfish challenging a female bluegill, was drawn by Mr. Edward C. Visnow, assistant technical editor of the Illinois Natural History Survey.

The photographs are the work of Dr. Bennett, Mr. William E. Clark, and Mr. Wilmer D. Zehr; Mr. Clark was formerly photographer and Mr. Zehr is presently photographer for the Natural History Survey.

LITERATURE CITED

- Hubbs, Carl L.
 1920. Notes on hybrid sunfishes. *Aquatic Life* 5(9):101-3.
 1955. Hybridization between fish species in nature. *System. Zool.* 4(1):1-20.
- Hubbs, Carl L., and Laura C. Hubbs
 1931. Increased growth in hybrid sunfishes. *Mich. Acad. Sci., Arts, and Letters Papers* 13:291-301.
 1932. Experimental verification of natural hybridization between distinct genera of sunfishes. *Mich. Acad. Sci., Arts, and Letters Papers* 15:427-37.
 1933. The increased growth, predominant maleness, and apparent infertility of hybrid sunfishes. *Mich. Acad. Sci., Arts, and Letters Papers* 17:613-41.
- Krumholz, Louis A.
 1950. Further observations on the use of hybrid sunfish in stocking small ponds. *Am. Fish. Soc. Trans.* for 1949, 79:112-24.
- Lagler, Karl F., and Charles Steinmetz, Jr.
 1957. Characteristics and fertility of experimentally produced sunfish hybrids, *Lepomis gibbosus* and *L. macrochirus*. *Copeia* 1957(4):290-2.
- Luce, Wilbur M.
 1937. Hybrid crosses in sunfishes. *Ill. Acad. Sci. Trans.* 30(2):309-10.
- Moenkhaus, W. J.
 1911. Cross fertilization among fishes. *Ind. Acad. Sci. Proc.* for 1910:353-93.
- Ricker, William
 1948. Hybrid sunfish for stocking small ponds. *Am. Fish. Soc. Trans.* for 1945, 75:84-96.
- Thompson, David H.
 1935. Hybridization and racial differentiation among Illinois fishes. *Ill. Nat. Hist. Surv. Bul.* 20(5):492-4.
(Appendix to Annotated list of the fishes of Illinois, by D. John O'Donnell.)