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Some Genetic Characteristics of Three Walleye Stocks in Iowa and the Impact of Fry Stockings in the Cedar River

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Fry stockings of walleye *Stizostedion v. vitreum* into the Cedar River, Iowa began in the early 1950's, and two studies of the success of these plants gave conflicting results. The objectives of my study were to identify some genetic characteristics in three stocks of walleye and to utilize these characteristics to assess the genetic impact of fry stockings in the Cedar River. Electrophoretic analysis of malate dehydrogenase (MDH) and isocitrate dehydrogenase (IDH) was completed for walleye stocks from the Mississippi and Cedar Rivers, and Big Spirit Lake, Iowa. Phenotypic frequencies of Mdh $-$ B^2 and Mdh $-$ B^3 were as follows: Mississippi River walleye, 83 and 17%, respectively; Cedar River walleye, 71 and 29%, respectively; and Spirit Lake walleye, 19 and 81%, respectively. Chi-Square testing of these frequencies demonstrated that Spirit Lake fish differed significantly from Mississippi ($P<0.001$) and Cedar River fish ($P<0.05$), but there was no difference in frequencies between the two river walleye populations. Phenotypic frequencies of Idh $-$ B^1 and B^2 were 54 and 46% for Mississippi River fish; 65 and 35%, respectively, for Spirit Lake fish; and, 65 and 35% for Cedar River walleye, respectively. Chi-square testing indicated that these frequencies were similar ($P>0.05$). My study indicated that there are some genetic differences between walleye stocks in the two Iowa rivers and Big Spirit Lake. It is probable that the native Cedar River and Mississippi River stocks are genetically similar, particularly since the former is a tributary to the latter. Also, fry stockings of Big Spirit Lake walleye, and their progeny, have contributed only about 19% to the Cedar River population. Fry stockings are less successful than was once thought, and a more refined stocking program is needed.
INDEX DESCRIPTORS: Walleye, genetics, electrophoretic analysis, Cedar River, Mississippi River, and Spirit Lake

The walleye (*Stizostedion v vitreum*) is the most important coolwater hatchery product in the United States and Canada. Conover (1986) reported one billion coolwater fish were stocked in 47 states and 9 provinces (reports of 1983 or 1984). Of these fish, 95% were walleye, of which 98% were fry. The demand by fisheries managers for walleye is expected to increase, but more information is needed to evaluate size and survival (Conover 1986), as well as performance of genetic strains (Murphy and Kelso 1986).

Stockings of hatchery-produced walleye in interior Iowa rivers have become a traditional part of the management program of the Fisheries Bureau of the Iowa Department of Natural Resources. These plants of walleye occurred as early as 1893 (Anonymous 1893) when 300 adult walleye from the Ohio River were stocked into the Cedar River near Waterloo. Plants of adult walleye continued sporadically, but advances in fish culture made it possible to stock large numbers of walleye fry. In 1951, the walleye fry stocking program in Iowa was expanded to include some major rivers (Cleary and Mayhew 1961). Since 1951, river plants of hatchery walleye fry have varied from none to 26,000 fry/kilometer. The original intent of the fry stocking program was to supplement natural reproduction (Mayhew 1956).

Cleary and Mayhew (1961) reported that walleye fry stocked during alternate years did not influence year-class abundance in the Cedar River. They felt year-class strength was attributed to the intensity of spring flows. Nearly 20 years later, an unpublished management report (Northeast District Files, Manchester, Iowa) found natural reproduction was often nearly non-existent and year-class abundance appeared to be dependent on fry stockings. In addition, a statewide investigation of stream and river fish indicated that walleye comprised a small component of these communities, even in stocked rivers (Paragamian 1986). The impact of fry stockings and natural reproduction on the quality and quantity of Iowa's various stream walleye fisheries remained unsolved.

The estimated contribution of walleye fry stockings and natural reproduction to year-class strength is dependent upon the discrimination of fry from each source. Marking larval fish before release for long-term studies is a difficult task. The use of electrophoretic analysis of tissue enzymes has become a standard procedure for identifying various strains of walleye (Uthe and Ryder 1970, Vely 1970, Wingo 1982, and Murphy and Terre 1984). One of the key enzymes used as a marker is malate dehydrogenase (MDH) (Murphy and Terre 1984). The objectives of my study were: 1) to determine allelic frequencies of MDH and isocitrate dehydrogenase (IDH) in walleye from Big Spirit Lake, Mississippi River and Cedar River, 2) to identify any evidence of Big Spirit Lake walleye in the Cedar River and, 3) to provide direction for further research and management of walleye in interior Iowa rivers. Walleye were collected from the Mississippi River because virtually all interior river systems (in Iowa) have been stocked with Big Spirit Lake walleye fry and thus it was felt rhis stock would genetically be most representative of the native Cedar River walleye.

DESCRIPTION OF STUDY AREA

The Cedar River is within the Upper Mississippi River drainage basin while Big Spirit Lake (herein Spirit Lake) is within the Missouri River drainage (Figure 1). The Cedar River traverses through 11 counties in eastern Iowa and has a total drainage basin of 20,252km^2. The drainage basin of the Cedar River study area, at Waverly, is about 4,025km^2 and the river has mean annual discharge of 24.4 m^3/sec. The source of the Cedar River is in southern Minnesota while its confluence is with the Iowa River (Figure 1).

GENETIC SOURCES

The source of walleye fry for the Cedar River study area since 1979 has been the Rathbun State Fish Hatchery. Before 1979 fry came from the Spirit Lake State Fish Hatchery while fish from this same source are still released into the Cedar River upstream of the study area. Brood fish for the Rathbun Hatchery are taken from Rathbun Reservoir. Rathbun Reservoir was impounded in 1969 and received

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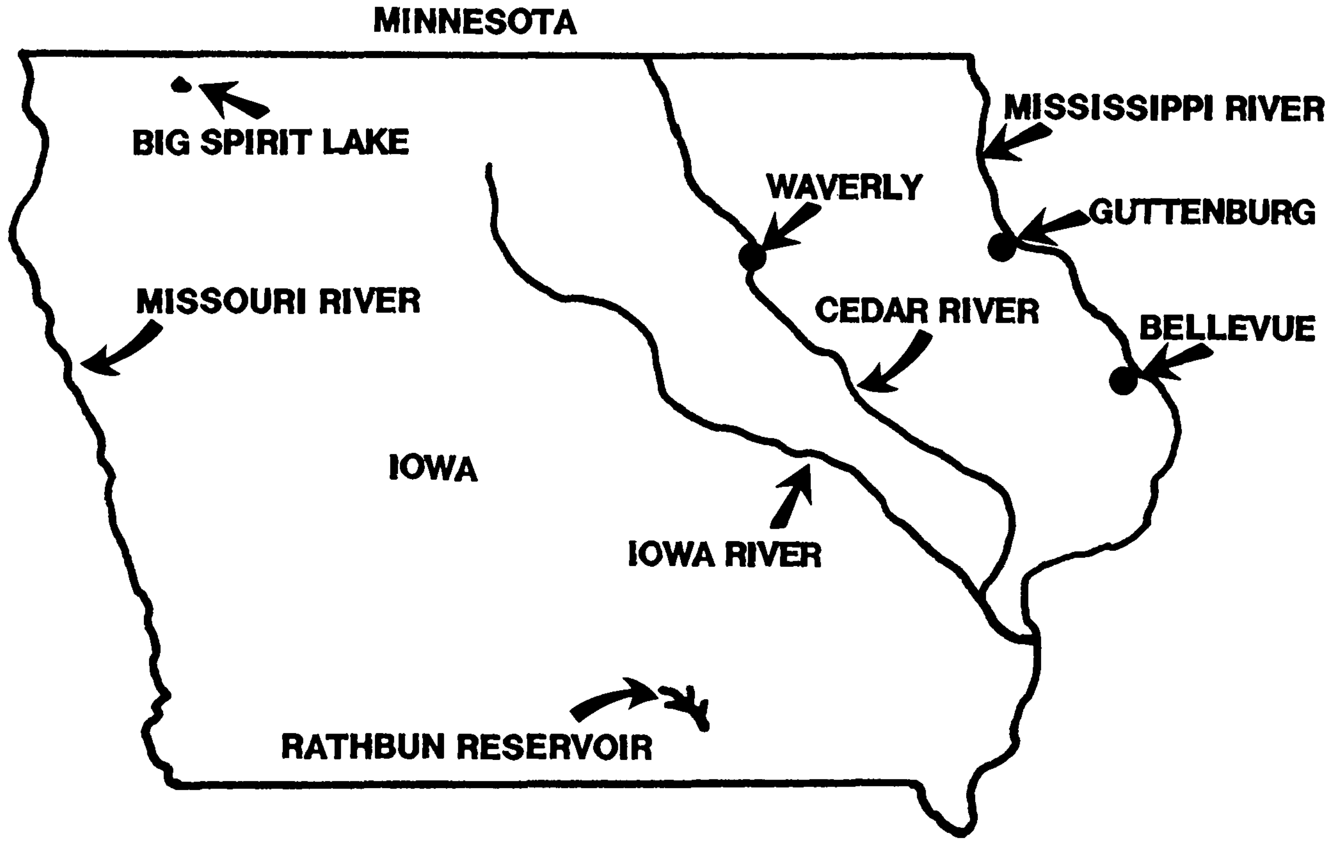

Fig. 1. Location of study area and walleye sample sites in Iowa.

an initial stocking of 11 million walleye fry (Paragamian 1977) from the Spirit Lake Fish Hatchery. Walleye stockings of the Spirit Lake strain continued each subsequent year. Thus, the Rathbun fish are of the same genetic make-up as those in Spirit Lake.

METHODS AND PROCEDURES

Walleye for this study were collected from the Cedar River (n = 29) at Waverly, the Mississippi River (N = 33) at Guttenberg and Bellevue, Iowa, and Spirit Lake (N = 13), Iowa (Figure 1). Upon capture with electrofishing gear all walleye were packed in ice and transported to a freezer. Walleye from Spirit Lake were mortalities of brood fish during spawning operations at the Spirit Lake Hatchery. The Spirit Lake fish were caught by gill-net.

Walleye from the Mississippi River ranged from 188 to 409 mm in total length (TL). Fifty percent were age-0, 29% were age-1, 14% were age-2, and the remainder age-3. Walleye from the Cedar River ranged from 211 to 554 mm TL. Ages of these walleye were: 20% age-0, 50% age-1, 23% age-2, and 7% age-4. The size and age range of walleye from Spirit Lake are not known other than all fish were adults.

White muscle tissue (about 2 g) was excised from frozen fish for MDH and liver tissue for IDH. Tissue samples were homogenized with grinding buffer and centrifuged for 20 minutes at 3,500 RPMs (31,500 g) while refrigerated (0.01M Tris. Citrate at pH 6.7).

Vertical starch gels were prepared using the same technique of Selander et al. (1971); Clayton et al. (1971) and, Harris and Hopkinson (1976). Scoring of phenotypic frequencies followed Clayton et al. (1971) except that Mdh − B is substituted for Mdh − C (Murphy et al. 1983), whereas electrophoretic analysis and scoring of Spirit Lake walleye was by Murphy and Lee (1986). Electrophoretic analysis of MDH and IDH was completed for all walleye in the samples, with one exception, 20 fish were analyzed for IDH in the Cedar River sample.

I calculated the contributed proportion of stocked Spirit Lake walleye fry and their possible progeny using Murphy et al.'s (1983) modification of the formula of Widrig and Taft (1957), as

$$C = |(a-u)\ (100)/(s-u)|;$$

Where: C = the percentage of the supplemented population sample comprised of Spirit Lake walleye, and a, u, and s are the observed Mdh-B^3 allele frequencies of the Cedar River walleye stock, the proposed frequency of native stock (Mississippi River walleye), and the introduced stock (Spirit Lake walleye). Confidence limits (C.I.) at 95% for C were calculated as

$$C \pm t[C(100-C)/n]^{1/2}$$

(Hogg and Tanis 1977), with t corresponding to P = 0.05 for (n-1)df.

FINDINGS AND DISCUSSION

Chi-Square testing of phenotypic frequencies of MDH (Table 1) indicated a significant difference existed between Spirit Lake fish when compared to the Mississippi River fish ($P<0.001$) and Cedar River fish ($P<0.05$); however, IDH frequencies did not differ significantly ($P>0.05$) (Table 1).

Phenotypic similarities of walleye MDH and IDH in the Cedar and Mississippi Rivers are not surprising. These results are explained by the fact the Cedar River is nearly a direct tributary of the Mississippi River. Although dams impassable to fish at most times of the year are present, the populations have been separated for only a very brief period in evolutionary time. But differences between MDH in the Cedar River walleye population and that of fish from Spirit Lake is of significant interest.

Table 1. Estimated allele frequency of Mdh-B^2, Mdh-B^3, Idh-B^1, and Idh-B^2 of walleye in three Iowa waters.

	MDH		IDH	
	B^2	B^3	B^1	B^2
Cedar River	71	29	65	35
Mississippi River	83	17	54	46
Spirit Lake	19	81	65	35

If stockings of Spirit Lake strain walleye would have made a measurable contribution to the Cedar River population, it should have been apparent in my study. Ninety-three percent of the Cedar River walleye used were of the 1985, 1984, and 1983 year classes. Walleye fry stockings in the Cedar River within the study reach in those years were 500,000; 1,000,000; and 1,000,000, respectively. Stockings in the Cedar River within the county immediately downstream during the same years totaled 4,000,000, while 750,000 were stocked in the county upstream (Northeast District Headquarters files).

Electrophoretic analysis indicated that the stockings of Spirit Lake strain walleye in the Cedar River had little genetic impact. The estimated contribution of Spirit Lake walleye to the Cedar River walleye sample, by fry stockings and possible progeny, was 19% (±9% C.I.). Spirit Lake walleye had a significantly different MDH phenotype frequency compared to the Cedar and Mississippi River walleye. Spirit Lake walleye stocked in the Cedar River have created little genetic drift and have only slightly driven Mdh − B^3 frequencies up and B^2 down. Thus, while walleye fry stockings were recently thought to be the mainstay of the fishery, the total management program is now open to question since the MDH gene frequencies of Cedar River fish are still very similar to those of the Mississippi River, despite 35 years of stocking Spirit Lake fish. Wingo (1982) presented comparative results using MDH and electrophoresis and indicated that walleye in the Tombigbee River, Mississippi, drainage were of the southern strain despite stockings of fish from Spirit Lake, Iowa. However, Murphy et al. (1983) found supplemental stockings of walleye fry from a Kansas reservoir accounted for an average of 67% of year-class strength in Claytor Lake, Virginia.

Stockings of walleye fry in interior rivers of Iowa are questionable and a more successful program should be designed. Further research will need to identify limiting factors to stocking success and that of natural reproduction. Some factors could be genetic strain performance, size of fish released, timing of releases or environmental variables, and habitat at release sights. These same questions may need to be answered by managers of other river systems in which walleye fry stockings are made.

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