A Biochemical and Cytological Study of the Longnose Sucker (*Catostomus catostomus*) and Large and Dwarf Forms of the White Sucker (*Catostomus commersoni*)

R. J. BEAMISH[1]

Department of Zoology
University of Toronto, Toronto, Ont.

AND H. TSUYUKI

Fisheries Research Board of Canada
Vancouver Laboratory, Vancouver, B.C.

BEAMISH, R. J., AND H. TSUYUKI. 1971. A biochemical and cytological study of the longnose sucker (*Catostomus catostomus*) and large and dwarf forms of the white sucker (*Catostomus commersoni*). J. Fish. Res. Bd. Canada 28: 1745–1748.

White (*Catostomus commersoni*) and longnose (*C. catostomus*) suckers possess diploid complements of 98 chromosomes, including metacentrics, submetacentrics, and acrocentrics. White sucker karyotypes differ consistently from longnose karyotypes by the presence of an additional four metacentrics. The karyotypes of the size and age at maturity of variants of white suckers were indistinguishable. Biochemically, longnose and white suckers are distinctive with respect to muscle myogens, hemoglobins, serum esterases, serum and muscle lactate dehydrogenases, and serum transferrins. The last group of proteins provides a clear genetic separation of the large-sized, late-maturing, and the dwarf, early-maturing white suckers. The former is polymorphic for serum transferrins.

BEAMISH, R. J., AND H. TSUYUKI. 1971. A biochemical and cytological study of the longnose sucker (*Catostomus catostomus*) and large and dwarf forms of the white sucker (*Catostomus commersoni*). J. Fish. Res. Bd. Canada 28: 1745–1748.

Le meunier noir (*Catostomus commersoni*) et le meunier rouge (*C. catostomus*) possèdent chacun un stock diploide de 98 chromosomes médiocentriques, submédiocentriques et acrocentriques. Le caryotype des meuniers noirs se distingue essentiellement de celui des meuniers rouges par la présence de quatre chromosomes médiocentriques supplémentaires. Il est impossible d'identifier le caryotype des meuniers noirs qui ont subi une mutation affectant la taille et l'âge à maturité. Biochimiquement, les deux espèces se distinguent par leurs myogènes musculaires, hémoglobines, estérases sériques, lacticodéshydrogénases sériques et musculaires et transferrines sériques. Ce dernier groupe de protéines permet de différencier génétiquement le grand meunier noir à maturité tardive de la forme naine à maturité hâtive. Le grand meunier noir est polymorphe quant aux transferrines sériques.

Received June 21, 1971

DURING a study of factors affecting the age and size of the white sucker *Catostomus commersoni* at maturity it was desirable to determine if genetic differences existed between two allopatric populations. Two populations from lakes located in Killarney Provincial Park, Ontario, were studied. A rapid-growing large form existed in George Lake, whereas in Lumsden Lake, an extremely slow-growing and early-maturing form was found. For a complete description of the lakes and their sucker populations, see Beamish (MS 1970).

Dence (1948) was unable to find morphometric distinctions between sympatric rapid- and slow-growing forms of white suckers. Because in some investigations cytological studies have proven successful in the differentiation of polymorphic forms of a species (Setzer 1970; Thorneycroft 1966), this approach was used to investigate possible genetic differences between the two forms. Karyotypes were studied from the two forms of the white sucker as

[1]Present address: Woods Hole Oceanographic Institution, Woods Hole, Massachusetts 02543, USA.

Printed in Canada (J2211)

well as from the closely related longnose sucker, *Castostomus catostomus*.

A biochemical approach, used successfully in distinguishing, for example, sibling species of sea cucumbers *Thyonella gemmata* (Manwell and Ann Baker 1963) and several species of morphologically indistinguishable rockfishes (Tsuyuki et al. 1968), was also used. Genetically linked polymorphism is known to exist in the muscle myogens of *C. catostomus* (Tsuyuki et al. 1967). Other variations in the electrophoretic patterns of muscle myogens and plasma proteins in several species of catostomids have also been reported (Tsuyuki et al. 1967). Variation in the serum esterases of *Catostomus clarki* is probably controlled by a two-allele system at a single locus (Koehn and Rasmussen 1967). Polymorphisms in the hemoglobins of a number of species of catostomids have been described previously (Tsuyuki et al. 1967; Koehn 1969). The general topic of assessing genetically distinct populations of fishes by serological and biochemical techniques has been reviewed by de Ligny (1969).

Methods

White suckers were obtained from Lumsden Lake (slow-growing, early-maturing), George Lake (fast-growing), and the Don River in Toronto, Ont. (intermediate size). Longnose suckers were the offspring of fish taken from Lake Huron and reared from eggs at the Laboratory for Experimental Limnology, Maple, Ont. In general, small young fish were preferred for chromosome squashes.

Chromosome squashes were prepared using a modification of the technique of McPhail and Jones (1966). For a detailed description of the method see Beamish (MS 1970) or Beamish et al. (1971). For catostomids, an intramuscular injection of 0.1 cc of 0.1% colcemid for 4 hr produced suitable preparations. Only good quality squashes were scored, and counts were obtained by photographing squashes and counting chromosomes from the photograph with reference to the original squash.

The electrophoretic mobilities of various groups of proteins from longnose suckers, as well as the rapid- and slow-growing forms of white suckers, were compared. No proteins from the intermediate-size Don River white suckers were analyzed. The muscle extracts and blood proteins were prepared for starch-gel zone electrophoresis as described by Tsuyuki et al. (1962, 1965, and 1967). The method of preparing plasma transferrins for autoradiography by the method of Giblett et al. (1959) was described by Tsuyuki et al. (1969). Comparisons of the two white sucker variants and the longnose sucker were made using hemoglobins, serum esterases, muscle myogens, serum transferrins, and lactate dehydrogenase in muscle, heart, and liver. Extracts were prepared from fresh material and analyzed immediately, thus avoiding problems associated with storage.

Results

Karyotype Analysis

Counts made of 182 squashes obtained from 9 longnose suckers and 12 white suckers were divided into the following two categories: squashes in which there was no significant overlap of chromosomes were classified as "very good," and squashes in which chromosomes were well spread but chromosome overlap could cause errors in interpretation were termed "good." Only preparations that showed no obvious signs of chromosome loss were counted.

All white and longnose suckers had a diploid complement of 98 chromosomes. The frequency of chromosome numbers obtained for longnose suckers and Lumsden and George Lake suckers is presented in Fig. 1. Squashes from Lumsden Lake white suckers were obtained from older fish and, in general, were of poorer quality than those of George Lake origin. Squashes prepared from longnose suckers yielded preparations of better quality than either George or Lumsden Lake fish. The deviation from the value $2n = 98$ according to the quality of the preparation is evident from Fig. 1. Values greater than 98 appeared to be the result of chromatid separation or breaks during squashing, or both. Similarly, it was felt that counts lower than 98 resulted from chromosome loss during squashing. Counts of 97 were found for both male and female suckers. The inability to interpret chromosomes in some preparations was considered to be a prime reason of variation in chromosome number. In general, when squashes of the quality shown in Fig. 2 were counted, the resulting diploid number was consistently 98.

The complements of both species include metacentrics, submetacentrics, and acrocentrics. Fourteen karyotypes prepared from seven different George Lake white suckers were identical. Similarly, no differences were observed among the 13 karyotypes of Lumsden Lake white suckers or the karyotypes of 13 longnose suckers.

A comparison of white and longnose karyotypes (Fig. 3–6) shows that the longnose karyotype (Fig. 4) was readily distinguishable from that of the white sucker (Fig. 3, 5, and 6). The diploid complement of white suckers contains 12 metacentrics, whereas, the longnose diploid complement possesses eight metacentrics. No diagnostic chromosome differences could be found among the size variants of white suckers from George Lake, Lumsden Lake, or the Don River (Fig. 3, 5, and 6).

Biochemical

No diagnostic differences between Lumsden and George Lake white suckers were found for hemo-

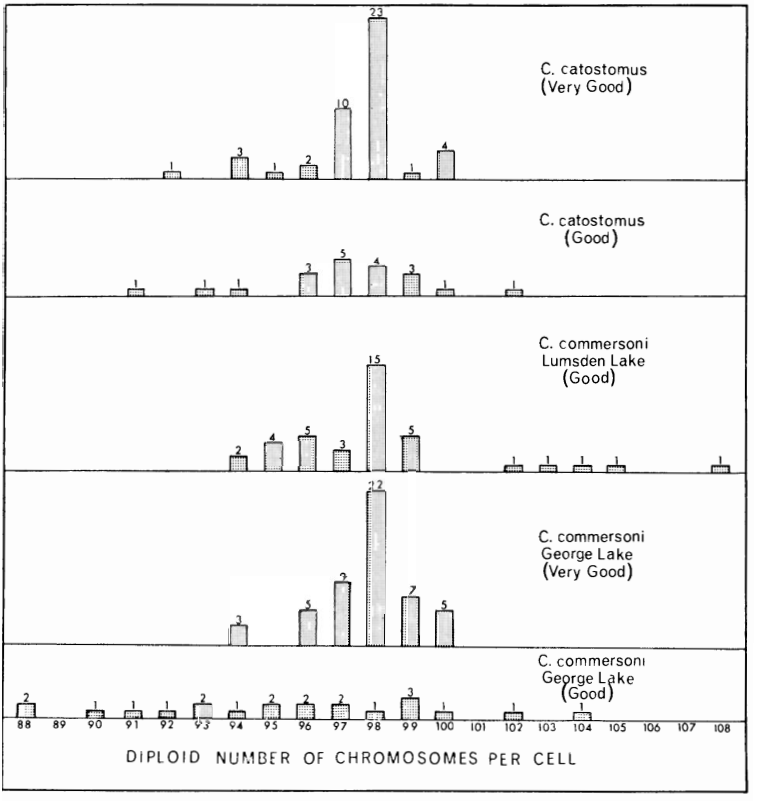

FIG. 1. Frequency distribution of chromosome counts of longnose and white suckers.

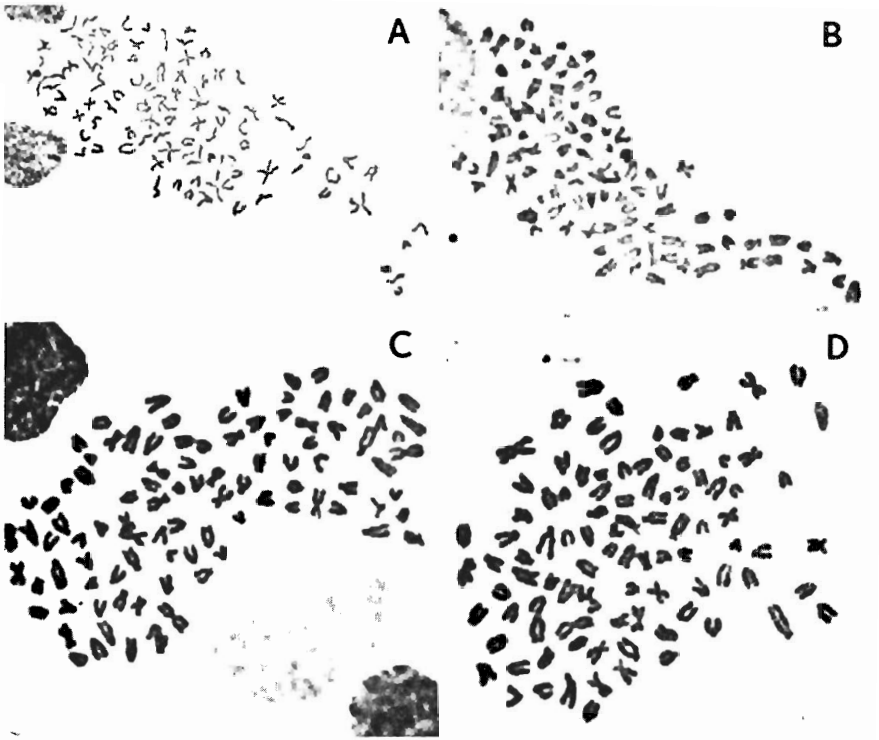

FIG. 2. Representative squashes from white and longnose suckers. A, George Lake white sucker; B, Lumsden Lake white sucker; C, Don River white sucker; D, longnose sucker.

FIG. 3. Karyotype of Lumsden Lake white sucker.

FIG. 4. Karyotype of longnose sucker.

FIG. 5. Karyotype of Don River white sucker.

FIG. 6. Karyotype of George Lake white sucker.

globins, esterases, and muscle myogens. Although some differences between the George and Lumsden suckers were found in the lactate dehydrogenase systems in muscle, heart, and liver, these differences were not sufficiently distinct to allow a separation of the two forms on this basis alone. In the plasma transferrins, however, a diagnostically useful character was found. A diagram of the transferrin bands of the zymograms identified by autoradiography is shown in Fig. 7. The five Lumsden specimens (Fig. 7, no. 12–16) differed consistently from the George Lake suckers (Fig. 7, no. 7–11). The transferrins of the George Lake variant appear in polymorphic form, designated AA, AB, and BB. The George and the Lumsden forms do not appear to have any transferrins in common, and the electrophoretic mobilities of the transferrin molecules of the two forms differ widely.

The longnose sucker possesses transferrins of mobility somewhat similar to the Lumsden form (Fig. 7, no. 2–6), but the white and the longnose suckers are clearly distinguishable by their hemoglobins, muscle, and other protein systems (Tsuyuki et al. 1967).

Discussion

Cytologically, white suckers could readily be distinguished from longnose suckers by the presence of four additional metacentrics in the total complement of $2n = 98$ chromosomes. Change in the karyotype has resulted without a change in the chromosome number; hence, the Robertsonian phenomenon of centric fusion does not account for the observed karyotype dissimilarities between these two species. Pericentric inversions may be involved. No variations in chromosome number of morphology were observed for white suckers from different habitats.

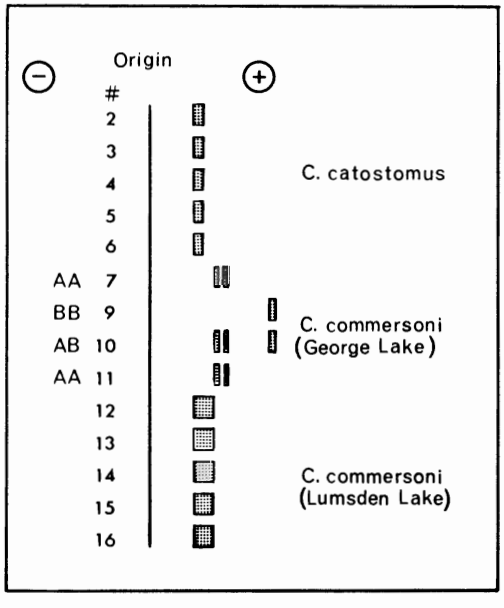

FIG. 7. Diagram of transferrin bands of the zymograms identified by autoradiography.

No diagnostic chromosome differences were observed in the karyotypes of white suckers representing different rates of growth and age at maturity. This does not necessarily mean that the karyotypes are similar, since the large number of acrocentric chromosomes of continuous gradation could obscure changes caused by translocations. The large number of chromosomes possessed by these species may be indicative that their ancestors may have at one time undergone a doubling of their chromosome complement (see Ohno et al. 1967 and 1968).

Cytological studies were inconclusive with respect to genetic differences between the various forms of white suckers. Although neither the effects of sialic acid and other factors influencing the electrophoretic mobility of serum transferrins nor the effect of oxidation levels of iron (Gaber and Aisen 1970) as possible causes of formation of double zoning in phenotype AA have been investigated at this time, a consistent separation of the George and Lumsden suckers was possible using this protein. In spite of the small sample, the Lumsden and George Lake suckers have to be regarded as genetically distinct forms with respect to this serum protein. The similarity of the other protein systems suggested that a close association did exist some time in the past.

Acknowledgments

We thank Prof K. Rothfels, Department of Botany, University of Toronto, for his advice, and critical review of the manuscript. This work was supported in part by funds received from the Fisheries Research Board of Canada and the National Research Council.

BEAMISH, R. J. MS 1970. Factors affecting the age and size of the white sucker *Catostomus commersoni* at maturity. Ph.D. Thesis. Univ. of Toronto, Toronto, Ont. 170 p.

BEAMISH, R. J., M. MERRILEES, AND E. J. CROSSMAN. 1971. Karyotypes and DNA values for members of the suborder Esocoidei (Osteichthyes: Salmoniformes). (Submitted for publication)

DE LIGNY, W. 1969. Serological and biochemical studies on fish populations. Oceanogr. Mar. Biol. Annu. Rev. 7: 411–513.

DENCE, W. A. 1948. Life history, ecology, and habits of the dwarf sucker, *Catostomus commersonii utawana* Mather, at the Huntington Wildlife Station. Roosevelt Wild Life Bull. 8: 81–150.

GABER, B. P., AND P. AISEN. 1970. Is divalent iron bound to transferrin? Biochim. Biophys. Acta 221: 228–233.

GIBLETT, E. R., C. G. HICKMAN, AND O. SMITHIES. 1959. Serum transferrins. Nature 183: 1589–1590.

KOEHN, R. K. 1969. Hemoglobins of fishes of the genus *Catostomus* in western North America. Copeia 1969: 21–30.

KOEHN, R. K., AND D. I. RASMUSSEN. 1967. Polymorphic and monomorphic serum esterase heterogeneity in catostomid fish populations. Biochem. Genet. 1: 131–144.

MANWELL, C., AND C. M. ANN BAKER. 1963. A sibling species of sea cucumber discovered by starch gel electrophoresis. Comp. Biochem. Physiol. 10: 39–53.

MCPHAIL, J. D., AND R. L. JONES. 1966. A simple technique for obtaining chromosomes from teleost fishes. J. Fish. Res. Bd. Canada 23: 767–768.

OHNO, S., J. MURAMOTO, AND L. CHRISTIAN. 1967. Diploid-tetraploid relationship among old-world members of the fish family Cyprinidae. Chromosoma 23: 1–9.

OHNO. S., U. WOLF, AND N. B. ATKIN. 1968. Evolution from fish to mammals by gene duplication. Hereditas 59: 169–187.

SETZER, P. Y. 1970. An analysis of a natural hybrid swarm by means of chromosome morphology. Trans. Amer. Fish. Soc. 99: 139–146.

THORNEYCROFT, H. B. 1966. Chromosomal polymorphism in the white throated sparrow, *Zonotrichia albicollis* (Gmelin). Science 154: 1571–1572.

TSUYUKI, H. E., E. ROBERTS, AND E. A. BEST. 1969. Serum transferrin systems and the hemoglobin of the Pacific halibut (*Hippoglossus stenolepis*). J. Fish. Res. Bd. Canada 26: 2351–2362.

TSUYUKI, H. E., E. ROBERTS, AND R. E. A. GADD. 1962. Muscle proteins of Pacific salmon (*Oncorhynchus*). III. The separation of muscle proteins soluble in low ionic strength salt solutions by starch gel electrophoresis. Can. J. Biochem. Physiol. 40: 929–936.

TSUYUKI, H. E., E. ROBERTS, R. H. KERR, J. F. UTHE, AND L. W. CLARKE. 1967. Comparative electropherograms of the family Catostomidae. J. Fish. Res. Bd. Canada 24: 299–303.

TSUYUKI, H. E., E. ROBERTS, R. H. LOWES, W. HADAWAY, AND S. J. WESTRHEIM. 1968. Contribution of protein electrophoresis to rock fish (Scorpaenidae) systematics. J. Fish. Res. Bd. Canada 25: 2477–2501.

TSUYUKI, E. H., E. ROBERTS, AND W. E. VANSTONE. 1965. Comparative zone electropherograms of muscle myogens and blood hemoglobins of marine and freshwater vertebrates and their application to biochemical systematics. J. Fish. Res. Bd. Canada 22: 203–213.