

Cytotaxonomic Study of Gila Trout, *Salmo gilae*

R. J. BEAMISH AND R. R. MILLER¹

Department of Fisheries and the Environment, Fisheries and Marine Service, Pacific Biological Station,
Nanaimo, B.C. V9R 5K6

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Gila trout, *Salmo gilae*, has a diploid chromosome complement of $2n = 56$ consisting of 49 metacentric and submetacentric chromosomes and 7 acrocentric or telocentric chromosomes. Karyotypes of this and other species of western North American trouts of the genus *Salmo* suggest that chromosomal modifications are correlated to speciation in this genus.

Key words: salmonidae, *Salmo gilae*, karyotype, chromosome, speciation

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Salmo gilae a un complément de chromosomes diploïdes de $2n = 56$, comprenant 49 chromosomes métacentriques et submetacentriques, et 7 chromosomes acrocentriques ou télacentriques. Les caryotypes de cette espèce et d'autres espèces de truites de l'Amérique du Nord occidentale suggèrent que des modifications chromosomiques sont associées à la formation d'espèces au sein de ce genre

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CHROMOSOME structure and behavior comprise an important tool for deciphering evolution, as clearly shown by the increasing use of karyotypes in evolutionary studies. Since the karyotype of a species is the physical basis of its genetic system, we are necessarily interested in the evolution of karyotypes. In the case of gila trout, *Salmo gilae*, a species limited in distribution to a few streams of New Mexico and Arizona (Miller 1972), the question of its specific distinction and of its derivation is materially aided by this first report of its karyotype.

Materials and methods — Chromosome squashes were prepared from gill tissue according to the

method of McPhail and Jones (1966) as modified slightly by Beamish et al. (1971). Photographs were taken from freshly squashed preparations using phase contrast microscopy. Squash preparations were then made permanent by the method described in Beamish et al. (1971) and karyotypes were established by comparing enlarged prints with the permanently mounted preparations. All permanent preparations have been saved and are available for examination upon request.

Juvenile *Salmo gilae* were obtained from Sterling Springs Hatchery in Arizona, of which the stock came from Main Diamond Creek, New Mexico (type locality of the species).

Results and discussion — *Salmo gilae* has a diploid chromosome complement of $2n = 56$ (Fig. 1, 2d). There was some variation in chromosome number (Table 1); however 39 of the 60 squashes that were counted had a $2n = 56$ complement. The greatest number of interpretable

¹Present address (R.R.M.): University of Michigan, Ann Arbor, Mich. 48109, USA.



FIG. 1. Somatic chromosomes at metaphase from a 43 mm *Salmo gilae*. Karyotype prepared from chromosome squash preparation. Bar represents 5 μ m.

squashes was obtained from fish 3 (Table 1) and as might be expected, the largest variation in chromosome counts came from squashes from this fish. The amount of variation in counts obtained in this study should not be interpreted as being indicative of the degree that chromosome numbers vary in this species. Variation in chromosome numbers can, and probably does, result from interpretative errors caused by chromosome overlaps, loss, breaks, or chromosome additions from other squashes. Addition of chromosomes to a squash causes fewer interpretative errors, as it is often possible to differentiate such additions by the differences in the appearance of chromosomes, caused in part by differences in the amount of contraction. There were several well-spread and apparently intact squashes (Fig. 2a) that had counts higher than $2n = 56$, indicating that some variation in chromosome number probably occurs, at least in cells from gill tissue.

TABLE 1. Frequency of chromosome counts.

$2n$	Fish no.				Total
	1	2	3	4	
53					
54					
55			6	1	7
56	1	7	26	5	39
57	1	5	5		11
58			2		2
No. of karyotypes	2	3	14	1	20

When $2n = 56$ most squashes had 49 metacentric and submetacentric chromosomes and 7 acrocentric or telocentric chromosomes (Table 2). One good squash (Fig. 2a) had 58 chromosomes with 9 acrocentric chromosomes. Other preparations had 57 chromosomes and 8 acrocentrics, and two squashes with 56 chromosomes also had

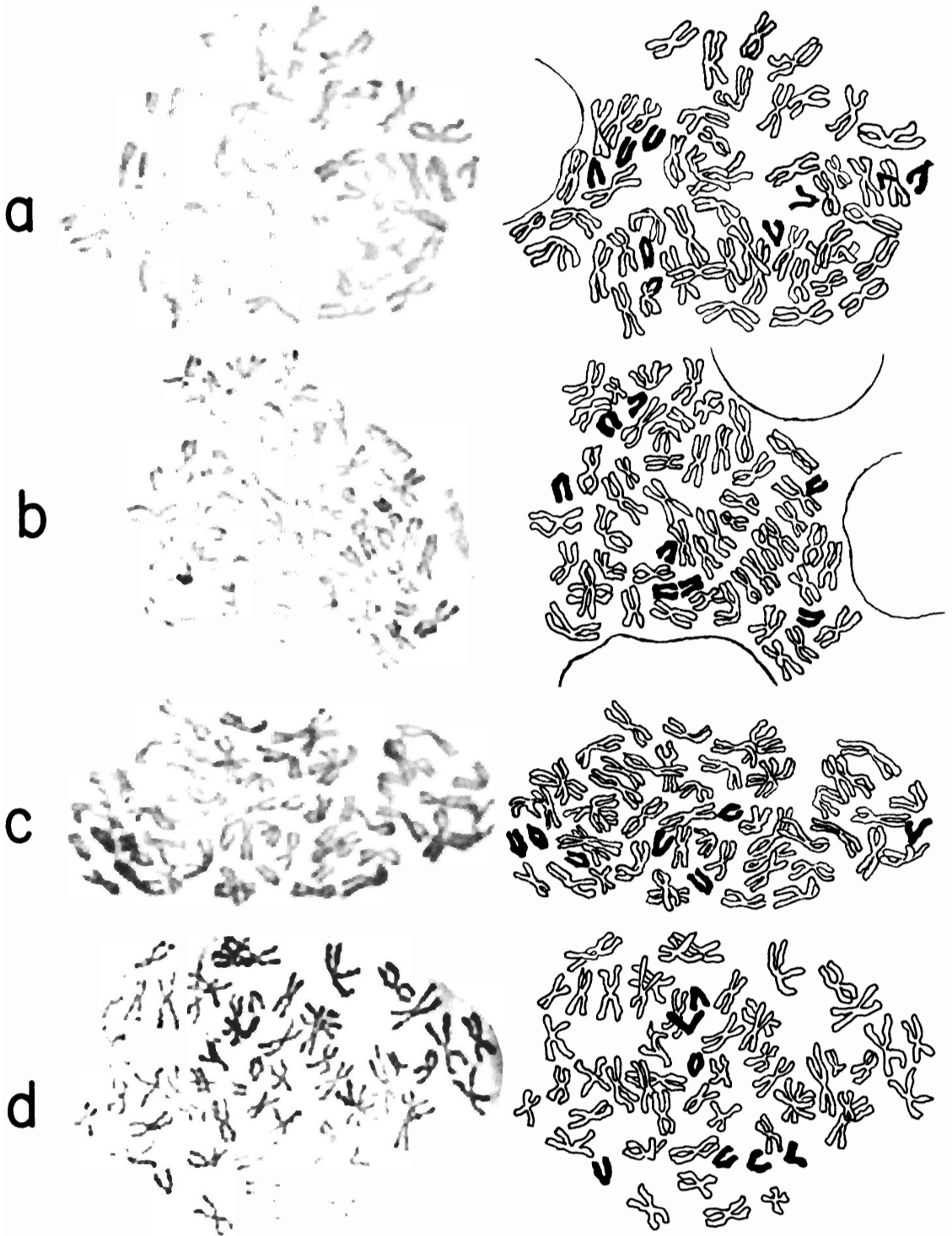


FIG. 2. Squash preparations and interpretation to show the number of acrocentric chromosomes. a, Fish 3, $2n = 58$; b, fish 1, $2n = 57$ and 8 acrocentrics; c, fish 2, $2n = 56$ and 7 acrocentrics; d, fish 3, $2n = 56$ and 7 acrocentrics. All interpretations and photographs are made from original permanent squash preparations since some detail is not apparent in the photograph.

TABLE 2. Karyotype analysis.

2n	No. of karyotypes	No. of acrocentrics	Fish no.
55	2	7	3
56	14	7	1, 2, 3, 4
56	2	8	3
57	1	8	1
58	1	9	3

8 acrocentric chromosomes (Table 2). While interpretive problems caused by chromosome breaks may account for some of the variation in chromosome morphology, some of the variation was apparently real. Despite this variation in number and morphology, at least one preparation from each fish examined had the modal count of $2n = 56$ with 7 acrocentric chromosomes (Table 2). The number of squashes counted or karyotyped was not constant for each fish sampled because of the varying number of divisions obtained and the varying quality of squashes.

The karyotype of *Salmo gilae* is very similar to that of *Salmo apache* (Miller 1972), but *Salmo gilae* possesses one more acrocentric and one fewer metacentric or submetacentric chromosome. This may have resulted from a pericentric inversion in only one metacentric or submetacentric chromosome. Unfortunately the four fish sampled were too small to determine if the chromosome heterozygosity was related to sex. While the presence of an apparently unpaired chromosome is unusual, there are reports of the occurrence of univalent chromosomes in fishes (Makino 1941) and chromosome polymorphism in the genus *Salmo* has been clearly demonstrated (Ohno et al. 1965; Roberts 1968). In the study of Atlantic salmon, *Salmo salar*, Roberts (1968) found a single large unpaired submetacentric in one of the stocks examined and was unable to explain its significance. Thus, the finding of an apparently unpaired chromosome for the stock of *S. gilae* used in this study is not unique.

A sufficient number of species in the genus *Salmo* from western North America have been karyotyped to allow some speculation concerning the pattern of evolution as indicated by their karyotypes. Unlike some genera, such as *Lepomis* (Roberts 1964) and *Esox* (Beamish et al. 1971), speciation in the *Salmo* may be correlated to chromosomal rearrangements. In an examination of the karyotypes of five North American species of Pacific salmon, Simon (1963), concluded that Robertsonian translocations were an important mechanism in explaining speciation in Salmonidae. There also is a definite pattern of chromosomal

TABLE 3. Hypothetical and present-day karyotypes for western North American *Salmo* species formed by Robertsonian and pericentric inversions. First number refers to chromosome number, second number to acrocentric or submetacentric chromosomes, and third number refers to the number of chromosome arms. Boxes around karyotypes indicate present-day karyotypes of species listed below boxes. Karyotypes other than *S. gilae* were obtained from Miller 1972.

		Pericentric inversion →		
Robertsonian fusion	64-22-106		64-24-104	
	<i>S. clarki</i>			
	62-18-106		62-20-104	
	60-14-106		60-16-104	
			<i>S. gairdneri</i>	
	58-10-106		58-12-104	
			<i>S. aguabonita</i>	
	56- 6-106	56-7-105		56- 8-104
	<i>S. apache</i>	<i>S. gilae</i>		
	54- 2-106			54- 4-104

number and morphology in the species of western North American *Salmo* that may be explained by a series of Robertsonian fusions and associated pericentric inversions (Table 3). If the karyotype of Cutthroat trout, *Salmo clarki*, of $2n = 64$ (Simon and Dollar 1963; Miller 1972) with 106 chromosome arms and 22 submetacentric or acrocentric chromosomes can be considered to be primitive, it is possible to construct a table of possible *Salmo* karyotypes that would be derived from Robertsonian fusions and pericentric inversions or both (Table 3). The table was constructed by considering that Robertsonian fusions reduce the chromosome number by 2 and the number of acrocentrics or submetacentrics by 4, but maintain a constant number of arms. Pericentric inversions can reduce the number of metacentric or submetacentric chromosomes by placing the centromere close to the end of the chromosome but will also decrease the number of chromosome arms. The close relationship of karyotypes among western North American *Salmo* to predicted chromosomal rearrangements is strongly suggestive that speciation is related to chromosomal rearrangements. If speciation is related to chromosomal changes, then species may have evolved in isolation rather than by crosses of two established species. If the preceding is true, species with karyotypes at the bottom and right of the table may have evolved later than species with karyotypes at the top and left of the table.

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