CHROMOSOME CYTOLOGY AND POLYMORPHISM IN THE CALIFORNIA HIGH SIERRA GOLDEN TROUT (*SALMO AGUABONITA*)[1]

J. R. GOLD AND G. A. E. GALL

Fisheries Biology Research Facility, Department of Animal Science, University of California, Davis, California 95616

The population variability in chromosome karyotype was examined in six samples of California High Sierra golden trout, *Salmo aguabonita*. From the analysis of 1,318 cells from anterior kidney tissue of 92 specimens, the modal diploid chromosome number was $2n = 58$ with 104 chromosome arms. Of cells with $2n = 58$ chromosomes, the typical karyotype was one containing 44 chromosomes with median centromeres, 2 chromosomes with submedian centromeres, 2 with subterminal centromeres and 10 with terminal centromeres. In addition, many cells contained a chromosome with a prominent satellite. Variability in chromosome number within the six populations followed a Robertsonian pattern and permitted the identification of two distinct population distributions. One population, made up of two samples, was more variable than the other and supported the hypothesis that this population was of golden trout × rainbow trout hybrid origin. Examination of metaphase I cells from testes showed numerous ring and rod multivalents consistent with the random nature of Robertsonian variability.

Introduction

Numerous chromosome studies of species of the family *Salmonidae* have revealed the existence of extensive chromosomal polymorphisms in several genera, notably the genus *Salmo*. In the most widely studied species, the Atlantic salmon, *Salmo salar*, diploid chromosome numbers ranging from $2n = 54$ to $2n = 60$ have been reported from several North American and European populations (Prokofieva, 1934; Svärdson, 1945; Boothroyd, 1959; Nygren *et al.*, 1968, 1972; Roberts, 1968, 1970). Similar polymorphisms at the intrapopulation and intraindividual levels have been observed in the rainbow trout, *Salmo gairdneri*, by Ohno *et al.* (1965) where from two to seven distinct chromosome complements from $2n = 58$ to $2n = 65$ were found.

The extact nature of the chromosome polymorphisms in the *Salmonidae* is as yet unclear. Roberts (1968, 1970) and Ohno *et al.* (1965) have proposed that the variation in chromosome number is due to Robertsonian translocations (Robertson, 1916) where centric fusions of nonhomologous acrocentric chromosomes produce metacentric chromosomes thereby reducing chromosome number without altering the amount of chromosomal material. Their data show that while chromosome number varies in both *S. salar* and *S. gairdneri*, the chromosome arm number, or *nombre fondamental* (Matthey, 1945), remains invariant.

Boothroyd (1959) and Svärdson (1945) both noted chromosome number variability in *S. salar*, but could not demonstrate a Robertsonian pattern, and thus suggested that perhaps genuine aneuploidy, without phenotypic alteration, was indicated. However, Boothroyd did note that difference between the European and North American *S. salar* karyotype might be explained by two Robertsonian fusions. In support of the suggestion that true aneuploidy exists, Davisson *et al.* (1972) found cytological and biochemical-genetic evidence of a trisomic but phenotypically normal Eastern brook

[1]The project was partially supported by the Department of Fish and Game, The Resources Agency of California, Contract No. 813.

Manuscript received September 23, 1974.

Can. J. Genet. Cytol. 17: 41–53, 1975.

trout, *Salvelinus fontinalis*. Rees (1967) on the other hand felt, as did Boothroyd (1959), that considering the large diploid chromosome numbers in most *Salmonidae* (between $2n = 60$ and $2n = 100$, see Roberts, 1967) much of the described variability was due to counting error and to chromosome loss during cell preparation.

Unfortunately, the study of gonadal preparations has done little to elucidate the nature of the polymorphisms. Instead of revealing a definitive pattern of prophase I irregularities indicative of specific chromosome rearrangements, the examination of meiotic cells has shown the presence of numerous ring and rod multivalents (Nygren *et al.*, 1968; Ohno *et al.*, 1965). Interestingly, the high number of multivalents observed has been cited as evidence for ancestral polyploidy rather than translocation or structural heterozygosity (Nygren *et al.*, 1968). Later, Nygren *et al.* (1972) concluded that reciprocal translocations caused the occurrence of multivalents at meiosis in salmon, based on a cross with the sea trout, *S. trutta*.

In the present study, six populations of the High Sierra California golden trout, *Salmo aguabonita*, were examined first to determine the karyotype of *S. aguabonita* and whether the system of chromosomal polymorphism found in other *Salmonidae* occurred in this species and; second, if polymorphisms did exist, to examine their nature and to determine whether populations could be separated on the basis of differences in their karyotypes.

Materials and Methods

Specimens of golden trout were collected from six different sites in the High Sierra, California during the summer of 1973. The six samples were as follows: 1) LKR — Little Kern River; 2) LSSC — lower Soda Springs Creek; 3) USSC — upper Soda Springs Creek; 4) GTC — Golden Trout Creek; 5) CWC — Cottonwood Creek; and 6) SFK — South Fork of Kern River. In an earlier taxonomic study, Gold and Gall (1975) identified the first two populations, LKR and LSSC, as possible remnants from previous hybridizations between endemic golden trout and introduced rainbow trout; the third, USSC, as *S. aguabonita whitei* Evermann; and the remaining three, GTC, CWC, and SFK, as *S. aguabonita aguabonita*. In that paper, a detailed map of the collection sites and a history of the six populations was presented.

All trout were transferred live to the Fisheries Biology Research Facility at Davis, and held until sacrifice. The method of chromosome preparation is described fully in Gold (1974). Briefly, the method involved intramuscular injection of colchicine 3-4 h prior to sacrifice, removal of desired tissue, treatment in hypotonic solution, and fixation in 3:1 methanol : acetic acid. Slides were prepared by dropping a suspension of fixed cells onto a clean microscope slide, air-drying, and staining with undiluted Giemsa. Cells with well spread chromosomes were selected for study, and were photographed using high contrast copy film.

Mitotic chromosome analyses were carried out on 1,318 cells prepared from anterior kidney tissue of 92 specimens. The number of cells (number of fish) examined from each population was LKR:211(16); LSSC:204(14); USSC:252(21); GTC:219(15): CWC:209(12); and SFK:223(15). From 10-15 cells per specimen were examined except for the Cottonwood Creek (CWC) trout where from 16-20 cells per specimen were counted due to the smaller number of collected fish. Cells with less than a chromosome number of $2n = 52$ were excluded.

Results

Karyology of S. aguabonita

A histogram illustrating the distribution of chromosome numbers observed in all cells from each sample is presented in Fig. 1. A clear and consistent mode of $2n = 58$

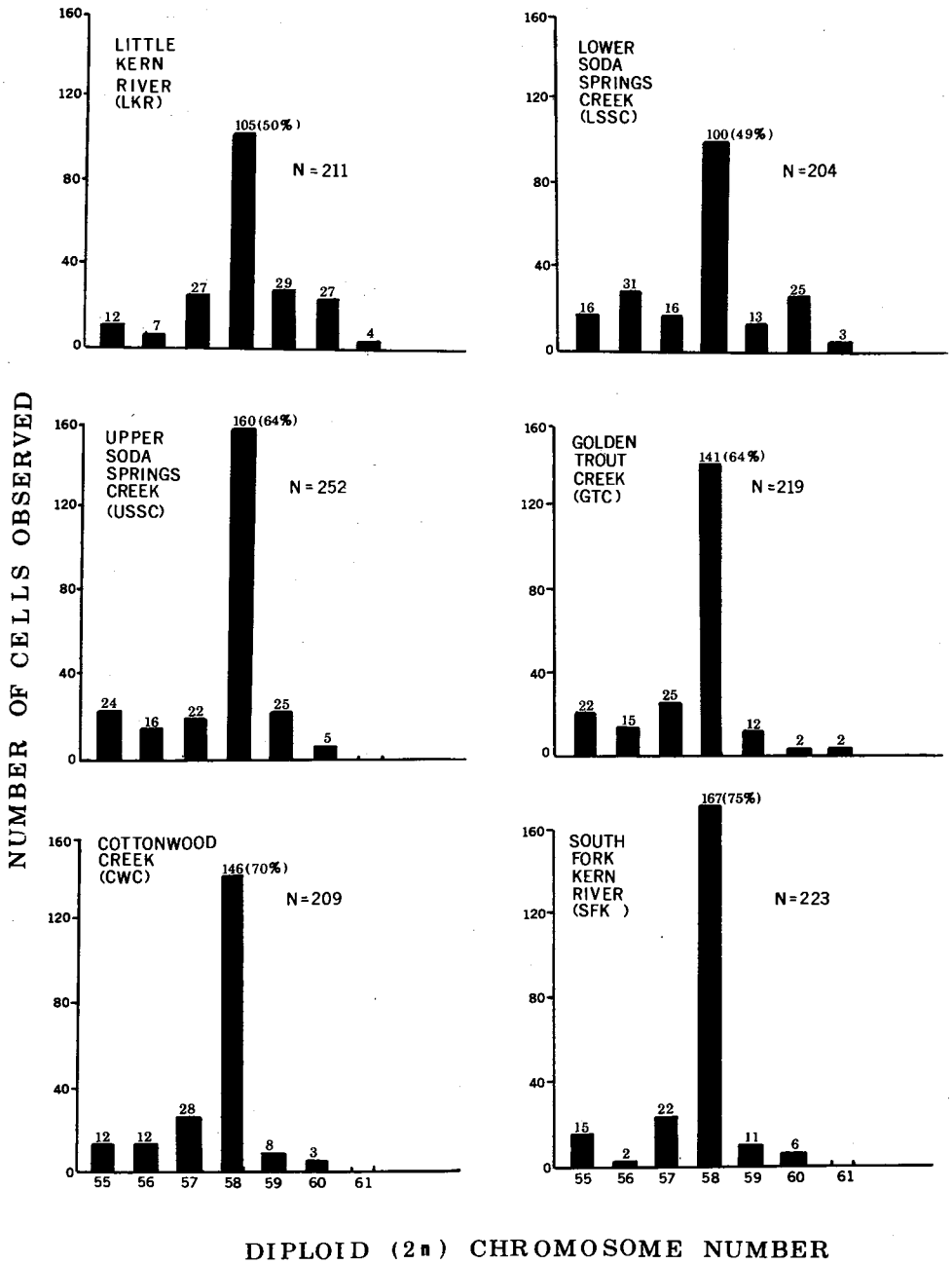

Fig. 1. Frequency distribution of somatic chromosome counts taken from anterior kidney tissue of six populations of golden trout, *Salmo aguatonita*.

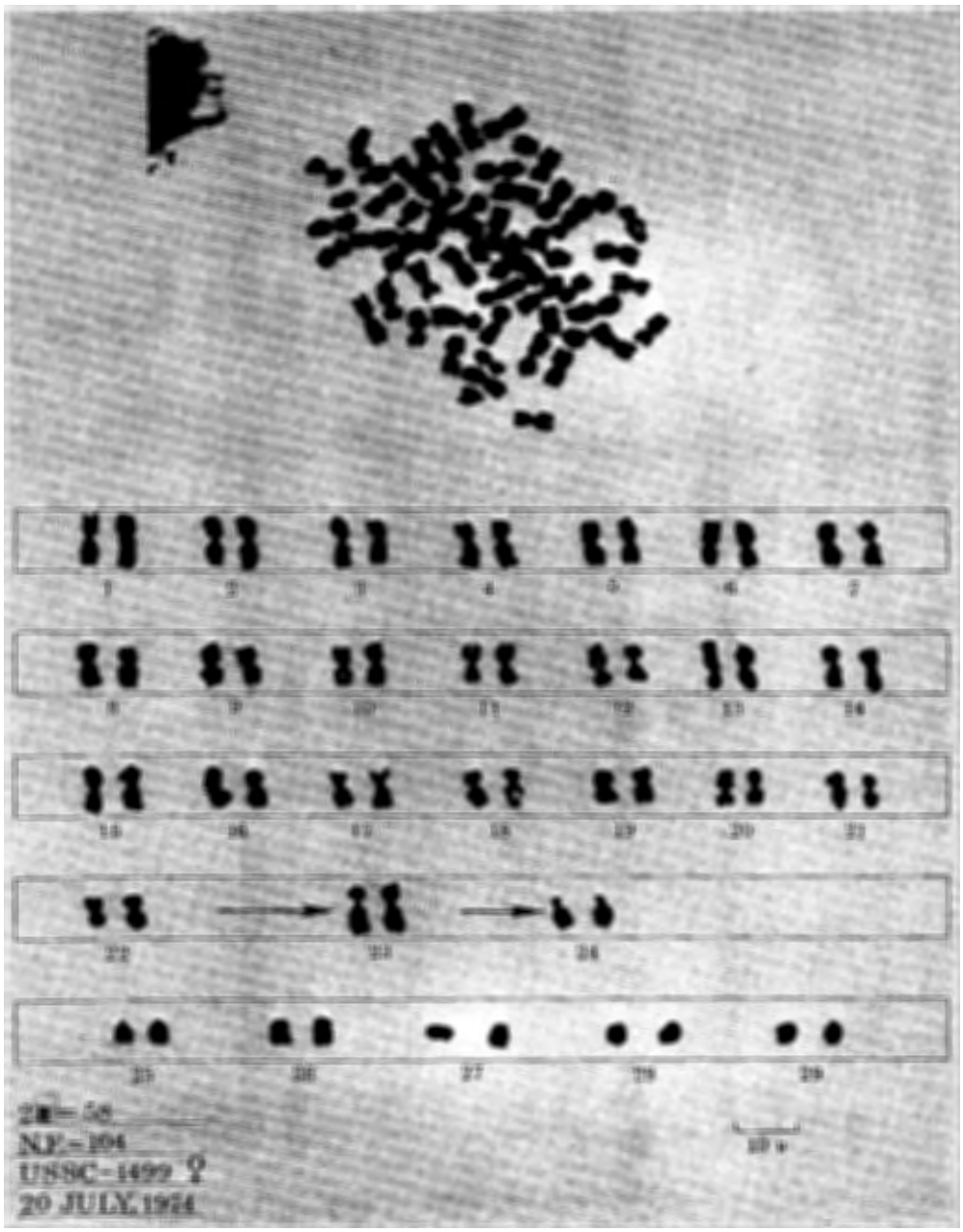

Fig. 2. Metaphase chromosomes from kidney and the karyotype of *Salmo aguabonita*. The arrows in the karyotype indicate chromosomes with submedian (#23) and subterminal (#24) centromeres.

chromosomes was found for all six samples, which is in agreement with previous estimates of diploid chromosome number in *S. aguabonita* (Simon, R. C. in Schreck and Behnke, 1971; Miller, 1972; Wilmot, 1974). However, variability of chromosome number in *S. aguabonita* is apparent. Cells with chromosome numbers ranging from $2n = 55$ to $2n = 60$ were observed in all six samples and a few cells were found with $2n = 61$ chromosomes in three of the six.

Of the modal class of $2n = 58$, the most frequently encountered karyotype (Fig. 2) was one containing 44 chromosomes with median centromeres, 2 chromosomes with

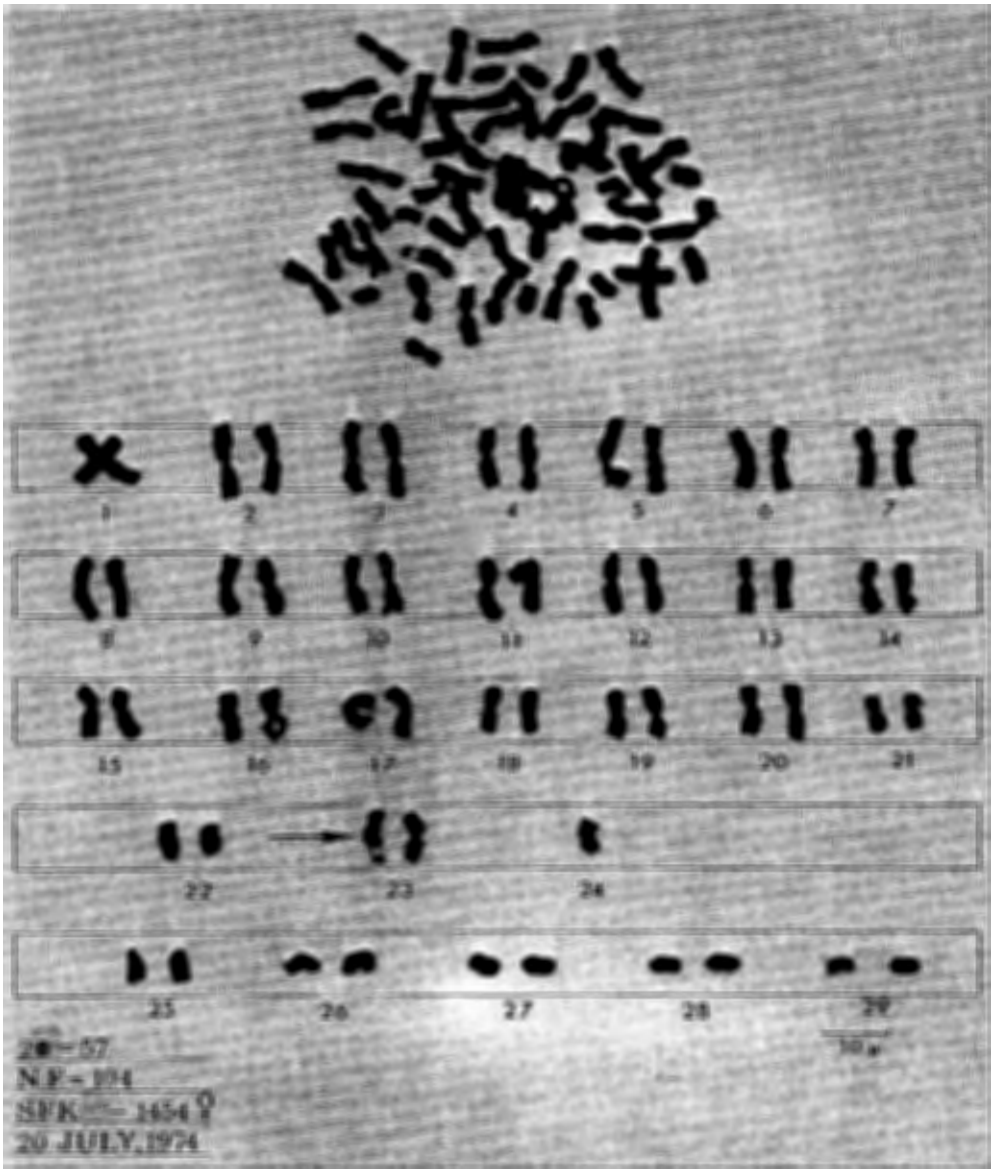

Fig. 3. Metaphase chromosomes from kidney and the karyotype of *Salmo aguabonita* showing the chromosome with a prominent satellite.

submedian centromeres, 2 chromosomes with subterminal centromeres and 10 chromosomes with terminal centromeres (terminology after Levan *et al.*, 1964). By scoring the chromosomes with median and submedian centromeres as two-armed chromosomes and the chromosomes with subterminal and terminal centromeres as one-armed chromosomes, the arm number of *S. aguabonita* is estimated as 104. This estimate is identical to that reported by Miller (1972) and in a careful study by Wilmot (1974), but is in disagreement with that of Simon (in Schreck and Behnke, 1971). In the latter, a total of 106 chromosome arms was estimated, a difference that was likely due to the scoring of the subterminal centrometric chromosome as a two-armed chromosome. This distinction is important since it establishes a possible evolutionary relationship of *S. aguabonita* with *S. gairdneri* which also has 104 chromosome arms (Ohno *et al.*, 1965) rather than with *S. clarki*, the inland cutthroat trout, which has 106 chromosome arms (Simon and Dollar, 1963).

Several cells were found which contained a chromosome with a prominent satellite (Fig. 3), similar to that described by Wilmot (1974). The chromosome with the satellite was identified as one with an approximately median centromere, and along with the chromosomes with the subterminal and submedian centromeres served as the only chromosomes easily identified in the *S. aguabonita* karyotype. No evidence of sexual dimorphism was observed among all cells scored.

Population Characteristics

The finding of variability in chromosome number among the samples of *S. aguabonita* permitted the examination of whether population separation through differences in chromosome karyotype was possible. Interpopulation differences were observed in the distributions of chromosome counts among the samples shown in the histogram in Fig. 1, although obvious differences such as different modal values as found by Roberts (1968, 1970) in *S. salar* were not found. In the LKR and LSSC samples, the modal number of $2n = 58$ was found in only 49 to 50% of counts as compared to the sharp modes ranging from 64 to 75% of counts in the USSC, GTC, CWC, and SFK samples. The lower frequency of the modal class in LKR and LSSC trout was apparently due to a comparatively higher frequency of hypermodal counts, i.e., 24% *vs.* only 8% in USSC, GTC, CWC, and SFK trout. It is important to note that chromosome numbers above $2n = 58$ might be expected in the LKR and LSSC trout since they were identified previously (Gold and Gall, 1975) as possible remnants from hybridization between *S. aguabonita* ($2n = 58$) and *S. gairdneri* ($2n = 60$).

In order to examine more closely the variability patterns among the six samples, arm number determinations were carried out on 749 cells. In Table I, the variation of chromosome arm number around the expected number of 104 is shown in relation to the variation in chromosome number. The picture which emerges from the table is remarkably similar to that observed by Roberts (1968) in *S. salar*. In all six samples examined, the highest proportion of cells (between 65% in LSSC and 85% in SFK) had 104 chromosome arms despite variability in the somatic chromosome number. This strongly indicates that the nature of chromosomal polymorphisms in *S. aguabonita* is Robertsonian. Moreover, of those cells with other than 104 chromosome arms, the large majority (from 83% in LSSC to 100% in GTC and SFK) were cells with *less* than 104 chromosome arms. Such a pattern suggests that most hypodiploid cells (<104 arms) resulted from chromosome counting errors or chromosome loss during preparation as suggested by Rees (1967). Hyperdiploid cells (>104 arms) on the other hand constituted only from 0 to 6% of the cells counted and may also stem from counting error.

TABLE I

Chromosome arm numbers for cells with different chromosome numbers from the six populations of golden trout

Population	Number cells	Chromosome arm number	Diploid ($2n$) chromosome number							Total	%
			55	56	57	58	59	60	61		
LKR	127	< 104	8	3	12	6	3	–	–	32	25
		104	–	1	3	58	13	15	–	90	71
		> 104	–	–	–	1	1	1	2	5	4
LSSC	116	< 104	9	16	7	2	–	–	–	34	29
		104	–	2	2	55	2	14	–	75	65
		> 104	–	–	–	–	5	–	2	7	6
USSC	147	< 104	14	9	9	4	4	–	–	40	28
		104	–	–	4	90	8	3	–	105	71
		> 104	–	–	–	–	2	–	–	2	1
GTC	138	< 104	14	9	9	2	–	–	–	34	25
		104	–	–	7	87	6	1	–	101	73
		> 104	–	–	–	–	2	–	1	3	2
CWC	107	< 104	8	6	8	1	–	–	–	23	22
		104	–	–	6	72	4	2	–	84	78
		> 104	–	–	–	–	–	–	–	0	—
SFK	114	< 104	8	3	4	2	–	–	–	17	15
		104	–	–	5	83	6	3	–	97	85
		> 104	–	–	–	–	–	–	–	0	—

Since the determinations of the number of chromosome arms indicated that the majority of the chromosomal polymorphisms in the six samples was Robertsonian, interpopulation comparisons were carried out using only data from cells with 104 chromosome arms. The distributions of chromosome numbers in cells with 104 chromosome arms and used in chi-square tests of independence (Goulden, 1960) are presented in Table II. Individual fish did not differ significantly in the frequency of nonmodal numbers, therefore, significant differences among the distributions can be attributed to interpopulation variability. Comparisons between sexes within samples were carried out since Davisson *et al.* (1973) found evidence of greater chromosome variability in males than in females of the hybrid splake trout. No significant difference ($P>0.05$) in the distribution of chromosome counts was found between the sexes in any of the six samples. The data for the two sexes were then pooled and comparisons made among the samples.

Highly significant differences ($P<0.01$) in the variability of chromosome numbers existed only for the comparison of LKR and LSSC with USSC, GTC, CWC and SFK ($\chi^2 = 53.4$; 4 df). Comparisons of the LKR with LSSC ($\chi^2 = 7.41$; 4 df) and among USSC, GTC, CWC and SFK ($\chi^2 = 3.07$; 9 df) were both nonsignificant. From the results, it can be inferred that the LKR and LSSC trout and the USSC, GTC, CWC, and SFK trout were samples from populations with different distributions of chromosome numbers. Considering the percentage of cells with other than $2n = 58$ chromosomes (Table II), it seems evident that the difference between the two groups was due to a 2 to 3-fold higher level of hypermodal counts in LKR and LSSC trout than in the other samples. Frequencies of hypomodal counts were similar in all six samples. A marked deficiency of cells with $2n = 59$ was observed in the LSSC sample; however, the comparison of LSSC with LKR did not show this difference to be significant.

TABLE II

Robertsonian variation in metacentric chromosome number in cells with 104 chromosome arms

Population	No. fish	Sex	No. chromosomes / No. metacentrics	56 / 48	57 / 47	58 / 46	59 / 45	60 / 44	% cells with $2n < 58$	% cells with $2n > 58$
LKR	6	♀			2	36	7	6		
	4	♂		1	1	22	6	9		
	10			1	3	58	13	15	4.4	31.1
LSSC	4	♀		1		25	2	5		
	5	♂		1	2	30		9		
	9			2	2	55	2	14	5.3	20.1
USSC	6	♀			1	43	5			
	6	♂			3	47	3	3		
	12				4	90	8	3	3.8	10.4
GTC	5	♀			5	48	1			
	5	♂			2	39	5	1		
	10				7	87	6	1	6.9	6.9
CWC	4	♀			2	47	3	1		
	2	♂			4	25	1	1		
	6				6	72	4	2	7.1	7.1
SFK	5	♀			3	49	2			
	4	♂			2	34	4	3		
	9				5	83	6	3	5.1	9.1

Meiosis in S. aguabonita

The variation of diploid chromosome number observed for cells with a constant chromosome arm number indicated that in *S. aguabonita* the chromosomal polymorphisms were Robertsonian in nature and suggested that in each population more than one structural alteration had occurred. In most organisms such alterations when structurally heterozygous produce various irregular metaphase I configurations from which the type or amount of alteration can be deduced. The data from 176 metaphase I cells from testis tissue with $2n = 58$ are presented in Table III, and a sample cell is shown in Fig. 4. Thirteen different meiotic configurations were observed ranging from the most frequent type (19.9%) with 13 ring bivalents, 3 ring tetravalents, 1 ring hexavalent, 5 rod bivalents and 1 rod tetravalent to the least frequent type (0.5%) with 18 ring bivalents, 1 ring tetravalent and 9 rod bivalents. Attempts to correlate the frequency of any one of the meiotic configurations with a specific population proved unsuccessful. Apparently, a large degree of structural heterozygosity, or at least of highly irregular meiotic configurations, is present in *S. aguabonita*. While such a meiotic behavior raises the interesting question of how proper segregation is insured, the difficulty of identifying specific alterations renders it of little help in determining possible population differences or in elucidating the system of chromosomal polymorphisms.

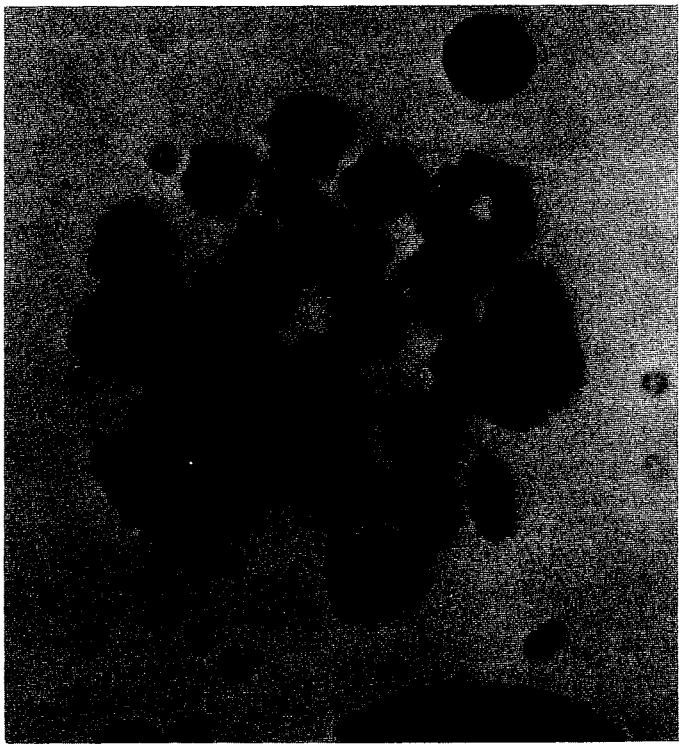

Fig. 4. Paired chromosomes of first meiotic metaphase from testis of *Salmo aguabonita* showing numerous multivalent configurations.

TABLE III

Meiotic configurations of cells with $2n = 58$ observed among 15 specimens from six populations of golden trout

No. cells observed	Frequency (%)	No. members	Rings			Rods	
			2	4	6	2	4
35	19.9		13	3	1	5	1
26	14.8		14	2	1	6	1
23	13.1		13	2	1	5	2
19	10.8		15	3		6	1
17	9.7		14	3		5	2
16	9.1		12	5		5	1
12	6.8		13	2		10	1
11	6.3		11	3	1	9	
10	5.7		15	3		8	
3	0.7		18	2		5	1
2	0.7		11			10	4
1	0.5		9	2		8	4
1	0.5		18	1		9	
176	100	*$\bar{X}$ =	13.4	2.8	0.5	6.1	7.2

*Weighted average number of configurations observed per cell.

Discussion

Any discussion of karyology in species of the family *Salmonidae* must contend first with the unique and highly unusual variability in chromosomal number which apparently characterizes most species of the genus *Salmo*. In the foregoing data, it was shown that diploid chromosome number varies in *S. aguabonita* from $2n = 56$ to $2n = 60$ in cells with 104 chromosome arms. This pattern indicates that a large majority of the variability in chromosome number can be accounted for as Robertsonian rearrangements involving centric fusions of nonhomologous acrocentric chromosomes. Furthermore, the occurrence of a small proportion of hypermodal counts in cells containing 104 chromosome arms suggests that centric fission also occurs in *S. aguabonita*. Similar conclusions were reached by Roberts (1968), Ohno *et al.* (1965) and Davisson *et al.* (1973) to account for chromosome variability in *S. salar*, *S. gairdneri* and *S. fontinalis*.

Other suggestions from the literature to account for the chromosome number variability in *Salmonidae* have included genuine aneuploidy (Svärdson, 1945; Boothroyd, 1959) and technique error (Rees, 1967). Neither seems a tenable explanation of most of the chromosome variability observed in *S. aguabonita*. First, 70 to 80% of the cells counted in all six samples contained 104 chromosome arms despite variation in $2n$ number. Were aneuploidy a viable alternative, one would expect a higher proportion of cells with other than 104 chromosome arms. Secondly, between 80 and 100% of those cells with other than 104 chromosome arms, were hypodiploid. The high proportion of hypodiploid cells strongly implicates chromosome loss during cell preparation rather than aneuploidy as the source of most counts with less than 104 chromosome arms. The very small proportion of hyperdiploid cells, however, cannot be totally excluded as possible aneuploids. Davisson *et al.* (1972, 1973), through a biochemical polymorphism and cytogenetic evaluation, presented strong evidence of a phenotypically normal but trisomic Eastern brook trout, *S. fontinalis*. Thus, true aneuploidy remains a possibility.

A further explanation of the chromosome variability is the so called "somatic segregation" mechanism proposed by Beçak *et al.* (1966). From their study of chromosome variability in the green sunfish, *Lepomis cyanellus*, they suggested that in a cell heterozygous for a Robertsonian translocation and undergoing mitotic division, preferential segregation of homozygous structural combinations could account for much of the variability in 2n number. The data from the *S. aguabonita* populations neither support nor contradict this theory. It should be pointed out, however, that if somatic segregation does occur at an appreciable frequency and efficiency, a deficiency of odd-numbered, nonmodal karyotypes, i.e., $2n = 57$ or 59, relative to the even-numbered, nonmodal karyotypes, i.e., $2n = 56$ or 60, should be observed. That is, whether the Robertsonian event is fusion or fission, somatic segregation should produce equal numbers of $2n = 58$ and $2n = 56$ daughter cells from a $2n = 57$ fusion heterozygote, and equal numbers of $2n = 58$ and $2n = 60$ daughter cells from a $2n = 59$ fission heterozygote. With the exception of the LSSC trout where a deficiency of $2n = 59$ cells was found (Table II), the proportion of odd-numbered, nonmodal karyotypes observed was nearly the same as or greater than the proportion of even-numbered, nonmodal karyotypes. Somatic segregation, then, does not appear to be a significant factor in causing chromosome number variation in *S. aguabonita*. Considering the fact that the proportions of cells with $2n = 59$ and $2n = 60$ are almost identical in LKR trout, and that the distribution of counts in the LKR and LSSC trout do not differ significantly, it seems likely that the paucity of $2n = 59$ cells in the LSSC trout was due to sampling error.

The examination of metaphase I figures of *S. aguabonita* did little to elucidate the nature of the chromosomal polymorphisms. Although the observed configurations were indicative of numerous structural rearrangements within the karyotype, quantitative identification of specific rearrangements was impossible. As was found in *S. salar* (Nygren *et al.*, 1968) and in *S. gairdneri* (Ohno *et al.*, 1965), meiotic figures of *S. aguabonita* presented a complex picture of numerous ring and rod multivalents. Nygren *et al.* (1968) had suggested that the presence of the numerous multivalents in *S. salar* was evidence for ancestral polyploidy in the *Salmonidae*, a hypothesis generally supported by others. This explanation is tenable since polyploidization and subsequent rearrangement could result in numerous chromosomes with heterobrachial homologies (Gropp and Winking, 1972). However, Nygren *et al.* (1972) examined a hybrid of the Atlantic salmon ($2n = 58$) and the sea trout, *S. trutta* ($2n = 80$) and observed only rod multivalents plus many univalents. They argued that if the salmonids represent a polyploid series, as Svärdson (1945) originally suggested, the salmon should be an autopolyploid. Then at least the haploid set of salmon chromosomes should contain chromosomes with homologous ends able to form ring multivalents in the hybrid. In their examination of several hundred metaphase preparations, no ring configurations were found although bivalent, trivalent and some quadrivalent rods were observed. Consequently, they concluded that reciprocal translocations rather than autopolyploidy cause the multivalents at meiosis in salmon.

Allendorf and Utter (1973) have shown that, while duplicate loci control several biochemical-genetic polymorphisms in the *Salmonidae*, inheritance is disomic. This raises the perplexing question of how correct segregation at meiosis, presumably leading to the normal diploid state, is accomplished among those *Salmonidae* with numerous meiotic irregularities. While we have no immediate answers to this question, it is interesting that our own studies at the Davis trout hatchery and an evaluation of survival records at California Department of Fish and Game trout hatcheries have indicated that progeny survival of rainbow trout, a species with numerous chromosomal polymorphisms (Ohno *et al.*, 1965; Gold and Gall, unpbl.),

may be as high as 85-90%. Apparently, the existence of numerous chromosomal polymorphisms and meiotic multivalents has little detectable effect on progeny survival.

Chi-square tests of independence of the distributions of the chromosomal polymorphisms among the six sampled populations revealed that the LKR and LSSC trout were drawn from one population distribution, and the USSC, GTC, CWC and SFK from a second. The difference between the two groups was highly significant, and stemmed from a higher frequency of hypermodal cells ($2n > 58$) in the LKR and LSSC populations.

The high frequency of hypermodal cells in the LKR and LSSC trout may be the result of previous hybridizations of endemic *S. aguabonita* with *S. gairdneri* introduced into these waters during 1931-41 (Dill, 1941; 1945). Since *S. gairdneri* has $2n = 60$ with 104 chromosome arms (44 metacentrics and 16 acrocentrics — Ohno *et al.*, 1965), introgression of *S. gairdneri* chromosomes into a *S. aguabonita* population could produce a higher proportion of hypermodal counts while still maintaining 104 chromosome arms. In this connection, Gold and Gall (1975) have found that the LKR and LSSC trout are somewhat intermediate between *S. aguabonita* and *S. gairdneri* in several meristic characters and in the intensity of body spotting patterns. However, while the present data support the conclusion that the LKR and LSSC are remnants of previous *S. aguabonita* × *S. gairdneri* hybridizations, it cannot be ruled out that certain of the polymorphisms are of adaptive significance, and hence that differences among populations stem from differential fitness of individual polymorphisms.

It is apparent that the nature of the chromosome polymorphisms in *S. aguabonita* is complex. As noted by Roberts (1968), the high chromosome number and relatively high proportion of chromosomes with median and submedian centromeres in salmonids distinguishes them from other fishes which have been studied. Certainly, the high degree of chromosomal polymorphism sets the salmonids apart from other vertebrates. It would seem, therefore, that future study of salmonid cytology should address itself directly to the questions regarding the nature and distributions of chromosomal polymorphisms in various salmonid species.

References

Allendorf, F. W. and Utter, F. M. 1973. Gene duplication within the family Salmonidae: Disomic inheritance of two loci reported to be tetrasomic in rainbow trout. Genetics **74**: 647–654.

Beçak, W., Beçak, M. L. and Ohno, S. 1966. Intraindividual chromosomal polymorphism in green sunfish (*Lepomis cyanellus*) as evidence of somatic segregation. Cytogenetics **5**: 313–320.

Boothroyd, E. R. 1959. Chromosome studies on three Canadian populations of Atlantic salmon, *Salmo salar* L. Can. J. Genet. Cytol. **1**: 161–172.

Davisson, M. T., Wright, J. E. and Atherton, L. M. 1972. Centric fusion and trisomy for the LDH-B locus in brook trout, *Salvelinus fontinalis*. Science **178**: 992–994.

Davisson, M. T., Wright, J. E. and Atherton, L. M. 1973. Cytogenetic analysis of pseudolinkage of LDH loci in the teleost genus *Salvelinus*. Genetics **73**: 645-658.

Dill, W. A. 1941. The Little Kern River Drainage, Tulare County — Progress report No. 1, California Department of Fish and Game, Administrative rep. no. 41–21.

Dill, W. A. 1945. The Little Kern River Drainage, Tulare County — Progress report No. 2, California Department of Fish and Game, Administrative rep. no. 45–29.

Gold, J. R. 1974. A fast and easy method for chromosome karyotyping in adult teleosts. Prog. Fish-Cult. **36**: 169-171.

Gold, J. R. and Gall, G. A. E. 1975. The taxonomic structure of six golden trout *(Salmo aguabonita)* populations from the Sierra Nevada, California. Proc. Calif. Acad. Sci. In press.

Goulden, C. H. 1960. Methods of statistical analysis. John Wiley & Sons, Inc., New York, London.

Gropp, A. and Winking, H. 1972. Robertsonian chromosomal variation and identification of metacentric chromosomes in feral mice. Chromosoma **39**: 265–288.

Levan, A., Fredga, K. and Sandberg, A. A. 1964. Nomenclature for centromeric position on chromosomes. Hereditas **52**: 201–220.

Matthey, R. 1945. L'évolution de la formule chromosomiale chez les vertébrés. Experientia **1**: 50–56.

Miller, R. R. 1972. Classification of the native trouts of Arizona with the description of a new species, *Salmo apache*. Copeia **3**: 401–422.

Nygren, A., Nilsson, B. and Jahnke, M. 1968. Cytological studies in the Atlantic salmon (*Salmo salar*). Ann. Acad. Regiae Sci. Upsal. pp. 21–52.

Nygren, A., Nilsson, B. and Jahnke, M. 1972. Cytological studies in Atlantic salmon from Canada, in hybrids between Atlantic salmon from Canada and Sweden and in hybrids between Atlantic salmon and sea trout. Hereditas **70**: 295–306.

Ohno, S., Stenius, C., Gaisst, E. and Zenzes, M. T. 1965. Postzygotic chromosomal rearrangements in rainbow trout (*Salmo irideus* Gibbons). Cytogenetics **4**: 117–129.

Prokofieva, A. 1934. On the chromosome morphology of certain pisces. Cytologia **5**: 498–506.

Rees, H. 1967. The chromosomes of *Salmo salar*. Chromosoma **21**: 472–474.

Roberts, F. L. 1967. Chromosome cytology of the Osteichthyes. Prog. Fish-Cult. **29**: 75–83.

Roberts, F. L. 1968. Chromosomal polymorphism in North American landlocked *Salmo salar*. Can. J. Genet. Cytol. **10**: 865–875.

Roberts, F. L. 1970. Atlantic salmon (*Salmo salar*) chromosomes and speciation. Trans. Am. Fish. Soc. **99**: 105–111.

Robertson, W. R. B. 1916. Chromosome studies. 1. Taxonomic relationships shown in the chromosomes of *Tettigidae* and *Acrididae*. V-shaped chromosomes and their significance in *Acrididae*, *Locustidae*, and *Gryllidae*. Chromosomes and variation. J. Morphol. **27**: 179–332.

Schreck, C. B. and Behnke, R. J. 1971. Trouts of the Upper Kern River Basin, California, with reference to systematics and evolution of Western North American *Salmo*. J. Fish. Res. Bd. Can. **28**: 987–998.

Simon, R. C. and Dollar, A. M. 1963. Cytological aspects of speciation in two North American teleosts, *Salmo gairdneri* and *Salmo clarki lewisi*. Can. J. Genet. Cytol. **5**: 43–49.

Svärdson, G. 1945. Chromosome studies on Salmonidae. Rept. Swedish State Inst. Freshwater Fishery Res. Drottingholm No. **23**: 1–151.

Wilmot, R. L. 1974. A genetic study of the red-banded trout (*Salmo* sp.). Ph.D. Thesis, Oregon State University, Corvalis, Oregon.