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Linkage Relationships Reflecting Ancestral Tetraploidy in Salmonid Fish

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ABSTRACT

Fifteen classical linkage groups were identified in two salmonid species (*Salmo trutta* and *Salmo gairdneri*) and three fertile, interspecific hybrids (*S. gairdneri* × *Salmo clarki*, *Salvelinus fontinalis* × *Salvelinus namaycush* and *S. fontinalis* × *Salvelinus alpinus*) by backcrossing multiply heterozygous individuals. These linkage relationships of electrophoretically detected, protein coding loci were highly conserved among species. The loci encoding the enzymes appeared to be randomly distributed among the salmonid chromosomes. Recombination frequencies were generally greater in females than in males. In males, certain linkage groups were pseudolinked with other linkage groups, presumably because of facultative multivalent pairing and directed disjunction of chromosomes. Five such pseudolinkage groups were identified and they also appeared to be common among species and hybrids. Duplicate loci were never classically linked with each other, although some exhibited pseudolinkage and some showed evidence of exchanging alleles. Gene-centromere recombination frequencies estimated from genotypic distributions of gynogenetic offspring were consistent with map locations inferred from female intergenic recombination frequencies. These linkage relationships support the contention that all extant salmonids arose from a common tetraploid progenitor and that this progenitor may have been a segmental allotetraploid.

POLYPLOIDIZATION may be a far more important evolutionary force than previously believed for sexually reproducing animal species (OHNO 1970; SCHULTZ 1980; BOGART 1980; BUTH 1983). A unique feature of tetraploid salmonids is that they appear still to be undergoing diploidization. Unusual progeny types, which probably result from double reduction since they are impossible to account for by strictly disomic segregation, have been observed for some duplicated loci that still share electrophoretically identical alleles (isoloci). In hybridized genomes (strain or species hybrids), loci belonging to different linkage groups often show pseudolinkage in males, but not in females; that is, nonparental (recombinant) male gamete types are formed in excess of parental ones.

Both nondisomic inheritance of isoloci and pseudolinkage were interpreted by WRIGHT *et al.* (1983) as forms of residual tetrasomy and to be due to homoeologous multivalent pairing. Linkage mapping in salmonids has been complicated by these phenomena. The many chromosomes ($1n$ = 26–42) of salmonid genomes likewise make gene mapping difficult. Nevertheless, linkage mapping in salmonids is a fruitful area of study because of the important evolutionary implications of genomic organization in a successful tetraploid derivative vertebrate group.

Increased heterozygosity in fertile species hybrids greatly increases the number of loci that can be tested for joint segregation. In this study, we examined brook trout (*Salvelinus fontinalis*) × lake trout (*Salvelinus namaycush*) hybrids (splakes), brook trout × arctic charr (*Salvelinus alpinus*) hybrids (sparctics) and rainbow trout (*Salmo gairdneri*) × cutthroat trout (*Salmo clarki*) hybrids (cutbows). We also report new linkage associations in rainbow trout and brown trout (*Salmo trutta*).

Induced gynogenesis has been used to estimate gene-centromere distances in rainbow trout (THORGAARD, ALLENDORF and KNUDSEN 1983; GUYOMARD 1984; ALLENDORF *et al.* 1986). We have used this technique to estimate gene-centromere distances of additional loci in rainbow trout, in brown trout, in cutbow hybrids, and in splake hybrids.

In this paper, we present our current results and address the following questions.

1. To what extent are classical linkage relationships conserved among salmonid species and genera? Do gene arrangements determined from intergenic recombination agree with those obtained from gene-centromere recombination?
2. What is the relationship between male and female recombination frequencies? Are differences consistent among species and loci?
3. What is the distribution of loci among the sal-

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monid chromosomes? Are protein-coding loci clustered or randomly distributed?

4. How many and what types of pseudolinkage arrangements are there? Are all pseudolinkages restricted to males and are they conserved among species and genera?

5. Why does allelic exchange occur between some duplicate loci (isoloci) but not others?

MATERIALS AND METHODS

Species and sources of fish: Brown trout were obtained from hatcheries in Pennsylvania, New Jersey, West Virginia, Connecticut, and New York, and from inbred lines maintained by J. E. WRIGHT.

Rainbow trout were from stocks maintained in hatcheries in Pennsylvania, West Virginia, Montana, Washington ("Donaldson" strain), and Lake Erie.

Cutthroat trout were obtained as crosses of Goshute Creek, Nevada, males, and Star Lake Utah, females, and probably represent the Bonneville subspecies (*Salmo clarki utah*) of inland cutthroat trout as described by LOUDENSLAGER and GALL (1980). All of the cutbow hybrids were derived from males of this source of cutthroat trout; however, females from several diverse strains of rainbow trout contributed to these hybrids.

Brook trout used to produce splake hybrids were from wild populations, inbred lines maintained by J. E. WRIGHT, and hatchery stocks from Pennsylvania and New York.

Lake trout used to produce the splake hybrids were from Lake Cayuga, New York, and Lake Manitou, Ontario.

Arctic charr were obtained from Flood Pond, Maine, and were crossed with inbred brook trout maintained by J. E. WRIGHT to produce the sparctic hybrids.

Blind pair matings of Atlantic salmon were made by personnel at the Huntsman Marine Laboratory, St. Andrews, New Brunswick.

Detailed information concerning the genetic backgrounds of parental fish used in these crosses will be furnished upon request.

Electrophoresis: A detailed description of the horizontal starch-gel electrophoretic methods and nomenclature employed in this study is given in MAY, WRIGHT and STONEKING (1979) and MAY (1980). The enzymes were visualized using the staining recipes given by HARRIS and HOPKINSON (1976). Increased resolution of the banding patterns for the enzymes ADA, AK, CK, FDP, GPI, MPI, PGD, and PEP was achieved by mixing the staining ingredients in 10 ml of buffer, combining this mixture with 10 ml of 2% agar maintained at 56°, and pouring over the gel slice lying on a glass plate. The albumin and hemoglobin proteins were visualized using a general protein stain consisting of 1% amido black solution in a 1:5:4 acetic acid:ethanol:water mixture. This mixture without amido black was used as a fixative to destain and preserve gel slices.

Diagrams, descriptions, and genetic interpretations of the electrophoretic banding patterns that were observed for each species and protein-tissue-buffer combination are given in JOHNSON (1984) with the exception of hemoglobin. Hemoglobin differences between rainbow and cutthroat trout are described and illustrated in BRAMAN *et al.* (1977).

Single-pair matings and production of diploid gynogens: In 1980, 1981, 1982, and 1983 single-pair matings of sexually ripe fish were individually spawned as described by MAY, WRIGHT and STONEKING (1979) and MAY, STONEKING and WRIGHT (1980).

In the fall of 1983, gynogenetic diploid trout were produced by fertilizing eggs with UV-inactivated sperm and heat-shocking to induce retention of the second polar body (CHOURROUT 1980). Rainbow trout sperm carrying the dominant color mutant "golden" or brook trout sperm were used to activate rainbow trout, cutbow hybrid, and brown trout eggs; brown or rainbow trout sperm were used to activate splake hybrid eggs. In this way, paternal leakage was easily prevented or monitored by the use of the color marker or by several marker genes having species-specific alleles.

Statistics: The observed frequencies of phenotypes in each progeny lot were compared with expected frequencies based on known or proposed parental genotypes, assuming Mendelian segregation ratios and independent assortment of loci. Chi-square analysis as described by MATHER (1951) was used to apportion deviations into component sources. A computer program (written by R. WOJCIK) was used for calculations because of the large numbers of locus pairs that were analyzed for linkage.

Each family was considered a separate sample and chi-square tests for heterogeneity among samples (MATHER 1951) were made before pooling results; results for males and females were kept separate.

Individual parental genotypes, progeny genotypic distributions, and orthogonal chi-square segregation analyses for each test family and chi-square heterogeneity tests among families are given in JOHNSON (1984) or can be obtained from the authors upon request.

Maximum likelihood estimates of recombination fractions and their standard errors were calculated according to MATHER (1951) for each sample and for the pooled data from homogeneous samples. Recombination fractions were based on known parental and nonparental types when the linkage phase was known. The least frequent class was sometimes assumed to be nonparental if there were substantiating data from other families or species. Knowledge of correct linkage phase is important in salmonid linkage studies because of the possibility of pseudolinkage.

Each gene-centromere recombination frequency was calculated as the proportion of gynogenetic diploid progeny that were heterozygous for that locus. Map distances in centimorgans were calculated as 50 × the gene-centromere recombination fractions, or as 100 × the intergenic recombination fractions. Mapping functions to correct for multiple crossovers were not used because of the nearly complete interference exhibited in salmonid meiosis (THORGAARD, ALLENDORF and KNUDSEN 1983).

RESULTS

The results of tests for linkage are presented separately below for each species or hybrid. Selected, exemplary data are presented in Table 1 while a complete summary of all classical linkages is presented in Table 2 and a complete summary of all pseudolinkages is presented in Table 3. Gene-centromere recombination values are given in Table 4.

Brown trout: Electrophoretic variation was detected at 16 loci—*Aat-(1,2),4*; *Ck-1*; *Dia*; *G3p-1*; *Gpi-3*; *Idh-3*; *Ldh-2*; *Mdh-1,2,(3,4)*; *Me-2*; *Mpi*; *Pgk-1*; *Sdh-1*; and *Sod*. Segregation of progeny types was consist-

TABLE 1

Exemplary patterns of nonrandom associations

Informative parents					Progeny genotypic frequencies						
			Loci; genotypes								
Family	Sex	Species	*Ck-1*	*Dia*	*AABB*	*AA'BB*	*AABB'*	*AA'BB'*	Chi-square joint	r^a	2 SE(r)
D-92	M	Brown	*AA'*	*BB'*	36	0	0	39	75.0*	0.00	0.04
D-97	F	Brown	*AA'*	*BB'*	15	3	2	17	19.7*	0.14	0.11
			Mdh-3,4		*AAAA*	*AAAA'*		*AAA'A'*			
B-46	M	Rainbow	AA/A'A'		14	128		18	57.6*	0.80	0.06
C-72	M	Rainbow	AA/A'A'		20	38		17	0.25	0.51	0.12
			Palb-1,2		*AAAA*	*AAAA'*		*AAA'A'*			
B-26	M	Rainbow	*AAA'A'*		2**	154		4**			
C-29	M	Cutbow BC	*AAA'A'*		20	62		14	8.9*	0.65	0.10

Informative parents					Progeny genotypic frequencies								
			Loci; Genotypes										
Family	Sex	Species	*Mdh-(3,4)*	*Palb-(1,2)*	*AAAA BBBB*	*AAAA BBBB'*	*AAAA BBB'B'*	*AAAA' BBBB*	*AAAA' BBBB'*	*AAAA' BBB'B'*	*AAA'A' BBBB*	*AAA'A' BBBB'*	*AAA'A' BBB'B'*
C-72	F	Rainbow	*AAAA*	*BBBB*									
	M	Rainbow	*AAA'A'*	*BBB'B'*	17	3	0	0	38	1	0	0	16

Selected, exemplary patterns of nonrandom associations and residual tetrasomic inheritance in salmonid species and hybrids. Except where noted, noninformative parents were homozygous for common alleles.

[a] Recombination fraction estimate.

* Significant at $P = 0.05$ level with d.f. = 1.

** Unexpected progeny; probably the result of double reduction gametes.

ent with simple Mendelian disomic ratios at all loci except the duplicate *Aat-1,2* isoloci. Excluding *Aat-(1, 2)*, only three statistically significant departures occurred out of 103 tests performed for single-locus segregation (JOHNSON 1984).

Independent assortment was tested for 59 different locus pairs in males and 37 locus pairs in females (JOHNSON 1984). Only three of these pair-wise tests revealed nonrandom segregation (Table 2). The loci *Ck-1* and *Dia* showed complete linkage in the two males tested, but the one female tested exhibited a recombination fraction (r value) of 0.14 (Table 1). The loci *Idh-3* and *Me-2* showed nonrandom segregation in two females examined. The loci *Gpi-3* and *Mpi* exhibited nonrandom assortment in the one male tested.

Allelic exchange between the duplicate *Aat-1,2* isoloci was tested by crossing three full-sib males that had *AAA'A'* phenotypes (*AA/A'A'* or *AA'/AA'* genotypes) with three homozygous (*AA/AA*) females. In two of the resulting families (C-186 and D-95), almost all progeny (181 of 187) had *AAAA'* phenotypes implying that the male parents were both homozygous for *Aat-1* and *Aat-2* (*AA/A'A'*). Six unexpected progeny types (two *AAAA* and four *AAA'A'*) probably resulted from double reduction male gametes (as a result of tetrasomy and interlocus recombination). In the third family (D-92), progeny segregated as would be expected if the male parent had been heterozygous at both *Aat-1* and *Aat-2* (*AA'/AA'*). Exchanges of alleles between *Aat-1* and *Aat-2* must have occurred in one of the parents (both had *AAAA'* phenotypes) of the full-sib males in order for these males to have *AA/A'A'* or *AA'/AA'* genotypes.

Five loci were mapped with respect to their centromeres in brown trout—*Aat-1*, *Ck-1*, *Mdh-3*, *Mpi*, and *Pgk-1* (Table 4). All five appeared to be distal to the centromere based on the high proportion of heterozygous gynogenetic progeny observed (0.85–1.00). The large degree of recombination observed between *Aat-1* and its centromere (0.97) is consistent with the observation that the duplicate *Aat-1,2* loci are able to exchange alleles (see DISCUSSION).

Rainbow trout: Only new linkage associations are presented in this paper; a compilation of all locus pairs (125 in males, 81 in females) examined for joint segregation in rainbow trout is given in MAY, WRIGHT and JOHNSON (1982) and JOHNSON (1984).

MAY, WRIGHT and JOHNSON (1982) proposed pseudolinkage to account for inheritance of the duplicated isoloci *Mdh-3,4* in two families of rainbow trout. One of these families, Z-58, resulted from crossing a female homozygous for the common allele with a male double heterozygote obtained from the Arlee, Montana hatchery strain, known to have been derived from intercrosses of diverse rainbow trout stocks. Results

TABLE 2

Classical linkages detected between locus pairs tested in this study

Loci tested	Species[a]	Informative parent	No. of families[b]	No. of informative progeny[c]	r^d	2 SE (r)
Aat-1 Mdh-1	Splake	F	4	290	0.34	0.06
Aat-2 G3p-1	Splake	F	1	64	0.52	0.12
	Cutbow	M	4	268	0.20	0.05
Aat-2 Hgb-1	Cutbow	F	2	84	0.48	0.08
	Cutbow	M	3	145	0.17	0.06
Ada-1 G3p-3	Sparctic	F	2	33	0.12	0.11
	Sparctic	M	3	106	0.03	0.03
Ak-2 Sod	Sparctic	F	3	108	0.08	0.05
	Sparctic	M	3	135	0.32	0.08
Ck-1 Dia	Brown	F	1	37	0.14	0.11
	Brown	M	2	109	0.00	0.02
Gpi-2 Sdh-2	Sparctic	F	1	36	0.11	0.10
Gpi-3 Ldh-1	Sparctic	F	1	38	0.42	0.16
	Sparctic	M	4	309	0.06	0.03
Gpi-3 Mpi	Brown	M	1	31	0.03	0.06
	Sparctic	M	1	80	0.10	0.07
Idh-1 Ldh-1	Cutbow	F	1	42	0.31	0.14
	Cutbow	M	7	443	0.01	0.01
Idh-1 Ldh-5	Cutbow	M	1	152	0.01	0.01
Idh-1 Mdh-3	Cutbow	M	1	150	0.09	0.05
Idh-1 Palb-1	Cutbow	M	4	231	0.08	0.04
Idh-3 Me-2	Brown	F	2	149	0.09	0.05
	Cutbow	F	1	40	0.32	0.14
	Cutbow	M	2	171	0.06	0.04
Idh-(3, 4) Me-(3, 4)	Rainbow	M	3	222	0.005	0.01
Ldh-1 Ldh-5	Cutbow	M	1	156	0.01	0.02
Ldh-1 Mdh-3	Cutbow	M	1	153	0.10	0.05
Ldh-1 Mpi	Sparctic	M	1	80	0.01	0.02
Ldh-1 Palb-1	Cutbow	M	2	109	0.07	0.05
Ldh-3 PepB-1	Cutbow	F	1	80	0.07	0.06
	Cutbow	M	1	57	0.04	0.05
	Splake	F	1	39	0.08	0.09
	Splake	M	1	78	0.00	0.03
Ldh-4 PepB-2	Splake	F	1	39	0.00	0.05
	Splake	M	2	90	0.06	0.05
	Sparctic	F	4	139	0.02	0.03
	Sparctic	M	2	149	0.04	0.03
Ldh-5 Mdh-3	Rainbow	M	2	229	0.05	0.03
	Cutbow	M	1	157	0.10	0.05
Ldh-5 Palb-1	Rainbow	M	2	90	0.02	0.03
	Cutbow	M	1	72	0.11	0.07
Mdh-(3, 4) Palb-(1, 2)	Rainbow	F	2	79	0.03	0.04
	Rainbow	M	5	222	0.06	0.03
	Cutbow	M	1	71	0.04	0.05
PepD-1 Sdh-1	Cutbow	M	7	299	0.02	0.02

[a] Splake hybrids include backcrosses to brook trout, as well as F_1's.
[b] Only statistically homogeneous families were combined.
[c] Only progeny with known parental contributions.
[d] Recombination fraction: nonparental/total.

shown for families B-46 and C-72 in Table 1, in which parental genotypes were known, reveal the pseudolinkage form of residual tetrasomy more definitively. A male progeny of Z-58, of genotype *AA/A'A'*, was used for family B-46; 80% of the progeny are nonparental types (79% were nonparentals in Z-58). A precocious male progeny of B-46, of genotype *AA/A'A'*, was used for family C-72; random assortment resulted from a breakdown of pseudolinkage in the second backcross male parent. Largely preferential multivalent pairing followed by directed disjunction would account for the results from the male in B-46 (WRIGHT *et al.* 1983). Random multivalent pairing and/or bivalent pairing would account for the results from the male parent of C-72.

A similar interpretation would account for the very

TABLE 3

Pseudolinkage between locus pairs tested

Loci tested	Hybrid	Informative parent	No. of families[a]	No. of informative progeny[b]	r^c	2 SE (r)
Aat-1 Aat-2	F_1 Sparctic	M	1	40	0.92	0.06
Aat-1 G3p-1	F_1 Sparctic	F	1	40	0.43	0.16
	F_1 Sparctic	M	1	76	0.68	0.11
	F_1 Cutbow	M	4	267	0.63	0.06
Aat-1 Hgb-1	F_1 Cutbow	F	2	84	0.59	0.10
	F_1 Cutbow	M	3	145	0.88	0.05
G3p-1 Mdh-1	F_1 Sparctic	F	3	118	0.48	0.09
	F_1 Sparctic	M	3	200	0.61	0.07
Idh-2 Idh-3	F_1 Cutbow	M	3	168	0.85	0.06
	BC_1 Cutbow	M	1	33	0.48	0.17
Idh-2 Me-2	F_1 Cutbow	M	5	244	0.83	0.05
Mdh-1 PepC	F_1 Sparctic	F	1	26	0.58	0.19
	F_1 Sparctic	M	3	83	0.98	0.03
Palb-1 Palb-2	BC_1 Cutbow	M	1	96	0.65	0.10
	BC_1 Cutbow	M	1	80	0.51	0.11
PepD-1 Sdh-2	F_1 Sparctic	F	3	115	0.50	0.09
	F_1 Sparctic	M	3	229	0.96	0.02
	F_1 Cutbow	M	3	164	0.81	0.07
Sdh-1 Sdh-2	F_1 Cutbow	M	3	107	0.81	0.07

Locus pairs that exhibited pseudolinkage in F_1 hybrids tested in this study. Note that in all females and some backcross males tested these same locus pairs segregated independently.

[a] Only statistically homogeneous families were combined.

[b] Only progeny with known parental contributions.

[c] Recombination fraction: nonparentals/total.

similar results reported for *Mdh-3,4* joint segregation by ALLENDORF and THORGAARD (1984). They used the Arlee, Montana, strain of rainbow trout to make a series of blind crosses. Three of the crosses of doubly heterozygous males to homozygous females gave *AAA'A'* progeny far in excess (86–92%) of that expected for classical tetrasomic ratios.

One such cross gave this phenotype in approximately 50% frequency (46%), a result similar to that found for doubly heterozygous females. Their two crosses of doubly heterozygous males giving the *AAA'A'* progeny phenotype in 57% and 59% frequencies probably represent a partial breakdown in pseudolinkage, similar to that occurring for *Palb-1,2* in family C-29, Table 1.

Results summarized in Table 2 show that one of the *Mdh-3,4* isoloci assorts nonrandomly with one of the *Palb-1,2* isoloci; linkage phases of the informative parents were not known. The results from one male rainbow trout (C-72, Table 1) suggested that it was heterozygous for *Palb-1*, *Palb-2*, *Mdh-3*, and *Mdh-4* and supported a model in which *Palb-1* and *Mdh-3* are classically linked to each other, *Palb-2* and *Mdh-4* are classically linked to each other, but *Mdh-3* and *Palb-1* are not classically linked with *Palb-2* and *Mdh-4*.

Examination of feral rainbow trout from Lake Erie revealed variation in the more anodal, cytosolic form of ME, designated *Me-3,4*. Genetic variation attributed to *Me-3,4* has not been reported previously in rainbow trout. Phenotypes in muscle tissue were often difficult to interpret, but resolution in eye tissue was good. Joint segregation analysis showed a tight linkage of one of the duplicate *Me-3,4* loci with one of the duplicate *Idh-3,4* loci (Table 2).

Two other previously untested locus pairs showed linkage in rainbow trout—*Ldh-5* with either *Palb-1* or *Palb-2* and *Ldh-5* with either *Mdh-3* or *Mdh-4*. Linkage with *Ldh-5* was assigned to *Mdh-3* and *Palb-1*, because data from cutbow hybrids later confirmed that *Ldh-5* was linked to *Mdh-3* and *Palb-1* and not to *Mdh-4* and *Palb-2* (Table 2).

Progeny genotypic distributions from family B-26 of rainbow trout provided evidence that the duplicate isoloci *Palb-1,2* exchange alleles (Table 1). The unexpected progeny genotypes (marked by asterisks) probably resulted from double reduction gametes as a result of tetrasomy and interlocus recombination, in a similar fashion to *Aat-1,2* in brown trout.

Eleven loci were mapped with respect to their centromeres in rainbow trout (Table 4). The large degree of recombination observed between *Palb-1* and its centromere is consistent with the observation that the duplicate *Palb-1,2* loci are able to exchange alleles (see DISCUSSION). Rainbow trout gene-centromere recombination frequencies estimated in this study were not statistically different from those obtained by THORGAARD, ALLENDORF and KNUDSEN (1983), GUYOMARD (1984), or ALLENDORF *et al.* (1986) for those loci that

TABLE 4

Gene-centromere recombination frequencies

Locus	Species	No. of females[a]	Progeny genotypes			Proportion heterozygotes Y	2 SE (Y)
			AA	*AA'*	*A'A'*		
Aat-(1, 2)	Brown	2	2	151	2	0.97	0.01
Aat-(1, 2)	Splake	2	6	36	0	0.86	0.11
Aat-1	Cutbow	5	5	88	6	0.89	0.06
Aat-2	Cutbow	5	4	91	4	0.92	0.05
Ada-1	Splake	2	3	33	2	0.86	0.12
Ada-2	Cutbow	7	3	108	2	0.96	0.04
Ck-1	Brown	1	7	66	5	0.85	0.08
Ck-1	Splake	2	3	39	0	0.93	0.08
Dia	Splake	2	3	31	4	0.82	0.12
Fdp	Cutbow	5	1	108	0	0.99	0.02
G3p-1	Splake	1	1	28	0	0.96	0.08
G3p-3	Rainbow	2	13	135	9	0.86	0.06
G3p-3	Cutbow	6	8	107	16	0.82	0.07
Gpi-3	Splake	1	1	28	0	0.96	0.08
Hgb-1	Cutbow	2	9	69	9	0.79	0.09
Idh-1	Cutbow	8	35	90	24	0.60	0.08
Idh-2	Rainbow	2	16	113	20	0.76	0.08
Idh-2	Cutbow	3	7	42	3	0.81	0.11
Idh-3	Rainbow	4	91	217	87	0.55	0.06
Idh-3	Splake	2	3	32	7	0.76	0.14
Idh-4	Cutbow	8	14	109	30	0.71	0.07
Ldh-1	Cutbow	1	32	0	33	0.00	0.09
Ldh-3	Rainbow	4	190	38	147	0.10	0.04
Ldh-4	Rainbow	2	56	2	82	0.01	0.02
Ldh-4	Splake	2	16	6	18	0.15	0.12
Ldh-5	Rainbow	3	137	2	149	0.01	0.01
Mdh-1	Splake	2	14	9	19	0.21	0.12
Mdh-(3, 4)	Rainbow	2	2	235	2	0.98	0.02
Mdh-(3, 4)	Brown	1	0	73	0	1.00	0.02
			AAAA'	*AAA'A*	*AA'A'A'*		
Mdh-3, 4[b]	Splake	2	2	38	2	0.96	0.06
			AA	*AA'*	*A'A'*		
Me-2	Splake	2	0	41	1	0.98	0.04
Mpi	Brown	2	3	145	3	0.96	0.04
Mup-2	Splake	2	2	37	2	0.90	0.10
Palb-(1, 2)	Rainbow	2	1	235	2	0.99	0.02
Palb-(1, 2)	Cutbow	1	0	16	0	1.00	0.34
PepB-1	Cutbow	8	68	18	66	0.12	0.05
PepB-2	Rainbow	1	31	6	35	0.08	0.06
PepB-2	Splake	2	16	2	13	0.06	0.05
PepC	Splake	2	12	24	6	0.57	0.16
PepD-(1, 2)	Cutbow	1	0	12	0	1.00	0.44
			AAAA'	*AAA'A'*	*AA'A'A'*		
PepD-1, 2[b]	Splake	2	1	38	3	0.96	0.06
			AA	*AA'*	*A'A'*		
Pgk-1	Brown	2	11	136	10	0.87	0.06
Pgk-2	Cutbow	3	5	34	4	0.79	0.12
Pgm-2	Rainbow	2	118	22	97	0.09	0.04
Pgm-2	Cutbow	6	47	14	50	0.13	0.06
Sdh-1	Cutbow	5	25	60	27	0.54	0.19
Sdh-2	Cutbow	2	9	15	4	0.54	0.19
Sdh-2	Splake	2	4	26	6	0.72	0.14
Sod	Rainbow	1	0	77	1	0.99	0.02
Sod	Cutbow	3	0	103	0	1.00	0.02
Sod	Splake	2	1	41	0	0.98	0.04

Estimates are given for female salmonids determined from genotypic distributions of their gynogenetic progeny.

[a] Only females with statistically homogeneous progeny distributions were combined.

[b] Both loci heterozygous; assumed to have equal gene-centromere recombination.

could be compared—*Idh-2, Idh-3, Ldh-4, Mdh-3, Palb-1, Pgm-2,* and *Sod.*

Cutbow hybrids: The rainbow trout × cutthroat trout hybridized genome permitted inheritance analyses of 26 loci—*Aat-1,2; Ada-1,2; Ck-1; Fdp; G3p-1; Hgb-1; Idh-1,2,3,4; Ldh-1,5; Mdh-(3,4); Me-2; Palb-(1, 2); PepA-1; PepB-1,2; PepD-1; Pgk-2; Pgm-2; Sdh-1,2;* and *Sod.* Cutbow F_1 hybrids were highly fertile and produced normal diploid backcross progeny that segregated in expected Mendelian ratios for all loci tested (JOHNSON 1984). Twenty of 179 tests made for single-locus segregation in 27 families showed significant departures from the expected 1:1 backcross ratio. However, if linked genes are taken into account, the 20 loci that exhibited aberrant segregation actually represented 17 chromosomes. Detecting only 17 aberrant chromosome assortments out of so many tests implies that gamete formation in the cutbow F_1 hybrids proceeded normally and that inheritance results can be straightforwardly interpreted.

A list of 204 locus pairs tested for joint segregation in male cutbow hybrids is given in JOHNSON (1984). A summary of the results from those locus pairs that exhibited nonrandom segregation is presented in Tables 1–3.

The duplicate *Aat-1,2* loci were pseudolinked with each other in 11 cutbow males. The locus *G3p-1* was classically linked to *Aat-2* and pseudolinked to *Aat-1* in four cutbow males. *Aat-1* and *Hgb-1* were pseudolinked and *Aat-2* and *Hgb-1* were classically linked in three male cutbow hybrids, whereas *Aat-1, Aat-2* and *Hgb-1* assorted randomly with one another in two female cutbow hybrids.

Variation of PEPD was arbitrarily attributed to *PepD-1* in male cutbow hybrids. (It is unclear whether one or two loci code for PEPD in these hybrids.) *PepD-1* was shown to be classically linked to *Sdh-1* in cutbow males and pseudolinked to *Sdh-2*; *Sdh-1* and *Sdh-2* were pseudolinked. *Ldh-3* and *PepB-1* were classically linked.

Two cutbow backcross hybrids were tested for joint segregation of *Palb-1* with *Palb-2*. These two loci assorted randomly in one male, but were weakly pseudolinked ($r = 0.65$) in the other male tested.

The loci *Idh-1, Ldh-1, Ldh-5, Mdh-3* and *Palb-1* were all shown to be classically linked in male cutbow hybrids. The one female cutbow hybrid tested for linkage of *Idh-1* with *Ldh-1* showed a recombination value of 0.31 as contrasted to the average value of 0.01 obtained from seven male hybrids; this discrepancy illustrates the difficulty of determining locus arrangements based on male recombination values (see DISCUSSION).

Idh-3 was classically linked with *Me-2* in two male and one female cutbow hybrids. *Me-2* and *Idh-3* were pseudolinked to *Idh-2* in male F_1 hybrids. In rainbow trout, the duplicate *Idh-3,4* loci usually cannot be distinguished; however, *Idh-3* and *Idh-4* each had unique alleles in the cutbow hybrid that allowed their distinction. The *Idh-3,4* locus whose unique allele was linked with *Me-2* was designated *Idh-3* to conform with results reported by MAY, WRIGHT and JOHNSON (1982). *Idh-4* showed no linkage associations with any other loci.

Eighteen loci were mapped with respect to their centromeres in rainbow × cutthroat trout F_1 hybrids (Table 4). The duplicate loci *Aat-1* and *Aat-2* had distinctive alleles in the cutbow hybrids (JOHNSON 1984); gene-centromere recombination frequencies for both loci could, therefore, be obtained from females heterozygous at both loci. The recombination frequency between *Aat-1* and its centromere (0.89) was not statistically different from the recombination frequency between *Aat-2* and its centromere (0.92). Likewise, the duplicate *Sdh-1,2* loci showed identical gene-centromere recombination frequencies. On the other hand, the duplicate loci *Idh-1* and *Idh-2* had statistically distinct recombination frequencies. Gene-centromere recombination frequencies in rainbow trout and cutbow hybrids, where comparable, were not statistically different.

Splake hybrids: Over 300 locus pairs have been examined for joint segregation in splake hybrids (MAY, STONEKING and WRIGHT 1980; HOLLISTER, JOHNSON and WRIGHT 1984; JOHNSON 1984); additional locus pairs that segregated nonrandomly are included in this paper.

Female F_1 hybrids had a much higher estimated recombination value (0.34) for *Aat-1* and *Mdh-1* (Table 2) than the value (0.04) reported for males (MAY, STONEKING and WRIGHT 1980). The one female splake examined for joint segregation of *Aat-2* and *G3p-1* showed random assortment of these two loci (Table 2); male splakes had an estimated recombination value of 0.09 (MAY, STONEKING and WRIGHT 1980).

Two new linkage associations were detected in splake hybrids (Table 2). *Ldh-3* was classically linked to *PepB-1* in one male and two female splakes, and *Ldh-4* was linked to *PepB-2* in one female and two male splakes. Gene-centromere recombination frequencies were estimated for 17 loci in splake hybrids (Table 4).

Sparctic hybrids: The highly heterozygous brook trout × arctic charr hybridized genome permitted inheritance analyses of 25 variable loci—*Aat-(1,2),4; Ada-1; Ak-2; Ck-1,3; Dia; Fdp; G3p-1,3; Gpi-2,3; Idh-3; Ldh-1,3,4; Mdh-1; Mpi; PepB-2; PepC; PepD-1; Pgm-1,2; Sdh;* and *Sod.* Eleven of 146 tests for single-locus segregation in 10 families of sparctic hybrids showed

significant departures from the expected 1:1 backcross ratio (JOHNSON 1984). Some of the 11 loci that exhibited aberrant segregation were linked and thus actually represented only eight abnormal chromosome assortments. About seven departures out of 146 tests would be expected by chance alone at a significance level of 0.05. Thus, meiosis appeared to be normal in these fertile F_1 sparctic hybrids, producing normally segregating diploid progeny types in backcrosses to brook trout.

All locus pairs (211 in males, 233 in females) tested for joint segregation in sparctic hybrids are given in JOHNSON (1984); only nonrandom associations are reported in this paper (Tables 2 and 3).

Aat-1 was pseudolinked with *Aat-2* in one sparctic male; other sparctic males showed weak pseudolinkage of *G3p-1* with *Aat-1* and with *Mdh-1*. *Mdh-1* showed strong pseudolinkage with *PepC* in three sparctic males, but these two loci assorted randomly in one sparctic female. *Gpi-3*, *Ldh-1* and *Mpi* were found to be classically linked. *Ada-1* was classically linked with *G3p-3* and *Ldh-4* was classically linked to *PepB-2*. *PepD-1* was pseudolinked to *Sdh-2* in sparctic males. A single female sparctic hybrid showed classical linkage of *Gpi-2* with *Sdh-2*. *Ak-2* was classically linked to *Sod* in sparctic hybrids; surprisingly, the recombination estimate for males ($r = 0.32$) was much higher than for females ($r = 0.08$).

Atlantic salmon: Electrophoretic variation was detected for seven enzyme coding loci—*Ck-1*, *Idh-4*, *Ldh-4*, *Mdh-1*, *Mdh-3*, *Me-2* and *Sdh-1*—and one regulator locus—*PgmR-1* (ALLENDORF, KNUDSEN and PHELPS 1982). Among 33 Atlantic salmon families, 16 of 28 possible locus-pair combinations in males and 12 of 28 in females were tested for random assortment. No cases of nonrandom assortment were detected. However, none of the locus-pairs tested have been found to be linked in other salmonids.

DISCUSSION

Conservation of linkage associations in Salmoninae: Loci found to be classically linked in one species or hybrid were found to be classically linked in other species or hybrids where they could be tested. The one apparent exception to this general observation was the linkage of *Ck-1* and *Dia* (the locus coding for the darkest zone of *DIA* expression) in brown trout. The locus coding for this same zone of *DIA* activity in *Salvelinus* was linked with neither *Ck-1* nor *Ck-2*. Considering the variability of karyotypes among salmonid species, a few linkage rearrangements might be expected, especially for linkages across centromeres. Indeed, MAY, JOHNSON and WRIGHT (1987) have shown sex linkage of *Ldh-1* and *Gpi-3* in sparctics while these loci are not sex linked in any other species.

Pseudolinkages also tend to be conserved among salmonid species and hybrids. To date, five pseudolinkage groups have been identified. These pseudolinkage groups involve certain chromosomes that still retain homologies with other chromosomes from an ancestral tetraploidization and, in hybrid males, can form multivalent pairs that, through preferential pairing and directed disjunction, result in pseudolinkage (WRIGHT *et al.* 1983).

Since both classical and pseudolinkages tend to be conserved among species, a composite linkage map was constructed for the whole Salmoninae subfamily including data from other studies summarized in JOHNSON and WRIGHT (1984) (Figure 1). Loci that have been shown to be linked in any species or hybrid are placed together on a single line representing a chromosome. These classical linkage groups are given arabic numerals; fifteen have been established so far. Species and hybrids examined to establish linked loci are indicated in the regions between loci.

The Roman numerals next to the dots connecting some classical linkage groups (chromosomes) represent pseudolinkage groups (Figure 1). For instance, loci on chromosome *1* have shown pseudolinkage with loci on chromosome *2* (pseudolinkage group I), loci on chromosome *3* with those on chromosome *4* (pseudolinkage group II), and so forth. Species or hybrids that have exhibited pseudolinkage among any of the loci of a particular pseudolinkage group and the maximum degree of pseudolinkage that was observed are listed next to the dots connecting the presumed homoeologous chromosomes of that pseudolinkage group. Species hybrids exhibited stronger degrees of pseudolinkage than did pure species; sparctic hybrids showed the strongest degree of pseudolinkage.

We felt that recombination estimates obtained from females gave more realistic estimates of actual physical map distances than did estimates obtained from males based on the distribution shown in Figure 2. Therefore, the gene order and map distances shown in Figure 1 were based only on intergenic recombination estimates obtained from females and/or female gene-centromere recombination estimates. However, joint segregation analysis in males was useful for detecting linkage; several loci that randomly assorted in females were shown to be linked in males (Table 2).

Most of the locus arrangements (with the possible exception of *Ck-1* and *Dia*) appeared to be similar among the species and hybrids examined (Figure 1). Estimates of map distances also were similar among species with the following exceptions. The map distance between *Me-2* and *Idh-3* was greater in cutbow hybrids than in splake hybrids or brown trout (Table 2) and the gene-centromere map distance for *Sdh-2* was greater in splake hybrids than in cutbow hybrids

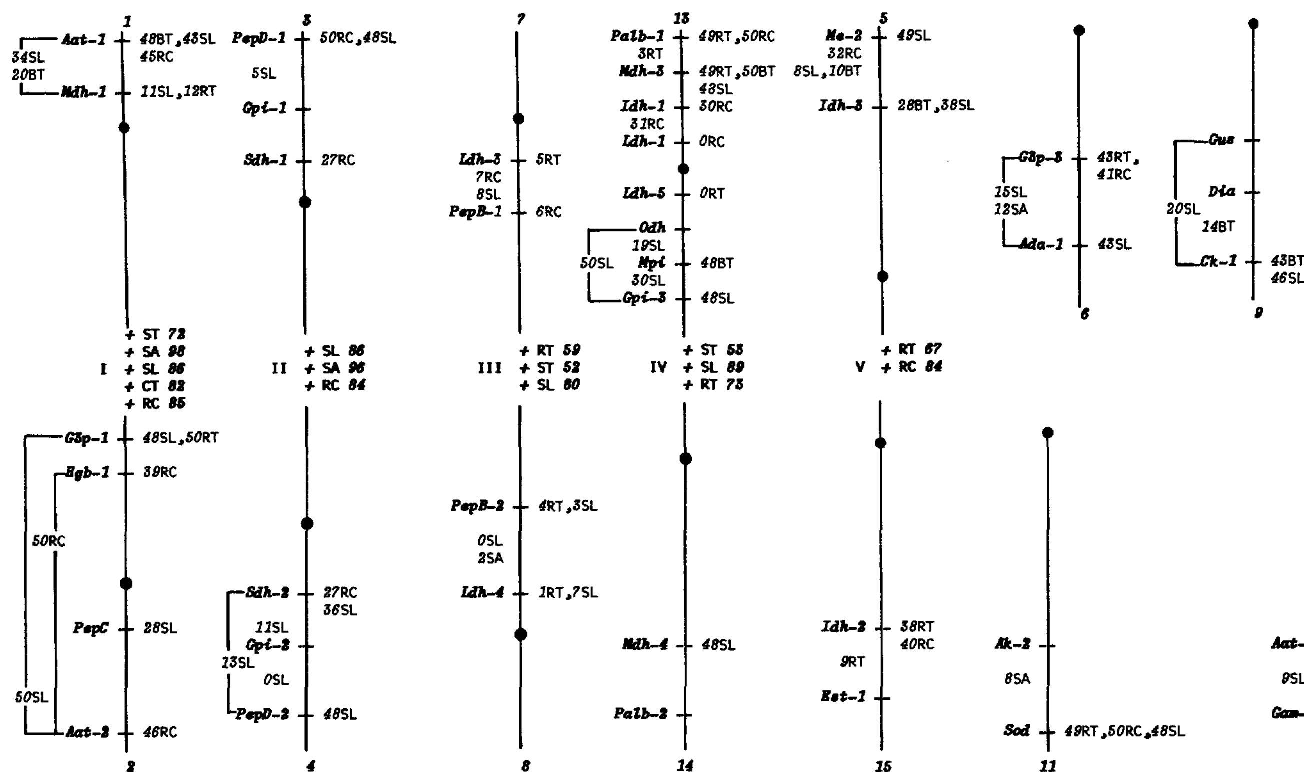

FIGURE 1.—Composite linkage map for the subfamily Salmoninae. Classical linkage groups (chromosomes) are given Arabic numerals and are represented by *vertical lines* with *dots* for centromeres. Pseudolinkage groups are given Roman numerals and are represented by plus (+) signs connecting certain classical linkage groups. Estimated map distances in centimorgans are given for each species based on female intergenic recombination frequencies (values on the left side of the chromosomes) and based on female gene-centromere recombination frequencies (values on the right side of the chromosomes). Maximum pseudolinkage values are given next to the plus sign for each species. Species abbreviations are as follows: RT = rainbow trout, CT = cutthroat trout, RC = rainbow × cutthroat trout hybrid, BT = brown trout, ST = brook (speckled) trout, SL = brook × lake trout hybrid, SA = brook × arctic charr hybrid.

(Table 4). Our estimates of the *Ck-1*-centromere distance in brown trout (43 cM) and in splake hybrids (46 cM) differed from that estimated for rainbow trout (23 cM) by ALLENDORF *et al.* (1986). Likewise our estimate for the *Mpi*-centromere distance in brown trout (48 cM) differed from that estimated for rainbow trout (4 cM) by ALLENDORF *et al.* (1986).

Map distances inferred from female intergenic recombination frequencies (shown on the left side of the chromosomes illustrated in Figure 1) agreed, in general, with the map distances inferred from female gene-centromere recombination frequencies (shown on the right side of the chromosomes illustrated in Figure 1). A possible exception to this general observation was the locus arrangement of linkage group 13 (Figure 1). The map distance between *Mpi* and *Gpi-3* was estimated to be 30 cM and the map distance between *Gpi-3* and its centromere to be 48 cM in splake hybrids. However, the map distance between *Mpi* and its centromere estimated from brown trout (48 cM) was inconsistent with the arrangement of the loci inferred from the splake hybrids. This discrepancy could be due to interspecific differences in the location of *Mpi*.

Repressed recombination in males: The complete absence of recombination in one sex has been observed only in a few insect families; in these cases it is the heterogametic sex in which crossing over does not occur. When recombination occurs in both sexes and when significant differences are found between sexes, recombination is generally less frequent in the heterogametic sex (reviewed in DUNN and BENNETT 1967). However, based on their analysis of extensive data for the mouse, DUNN and BENNETT (1967) concluded that differences in recombination probability are affected by local factors and that sex differences in recombination can occur in opposite directions at different locations in the genome of the same species.

Recombination in male salmonids is generally repressed (Table 2, Figure 2), presumably because of structural constraints imposed on crossing over by multivalent pairing involving metacentric (centrically fused) chromosomes (WRIGHT *et al.* 1983). An exception to this general rule was that male sparctic hybrids exhibited a greater recombination between *Ak-2* and *Sod* than did females. This exception could be explained as follows. If *Ak-2* and *Sod* were located near each other but distal to the centromere, and if high interference usually permitted only a single gene-centromere crossover (THORGAARD, ALLENDORF and

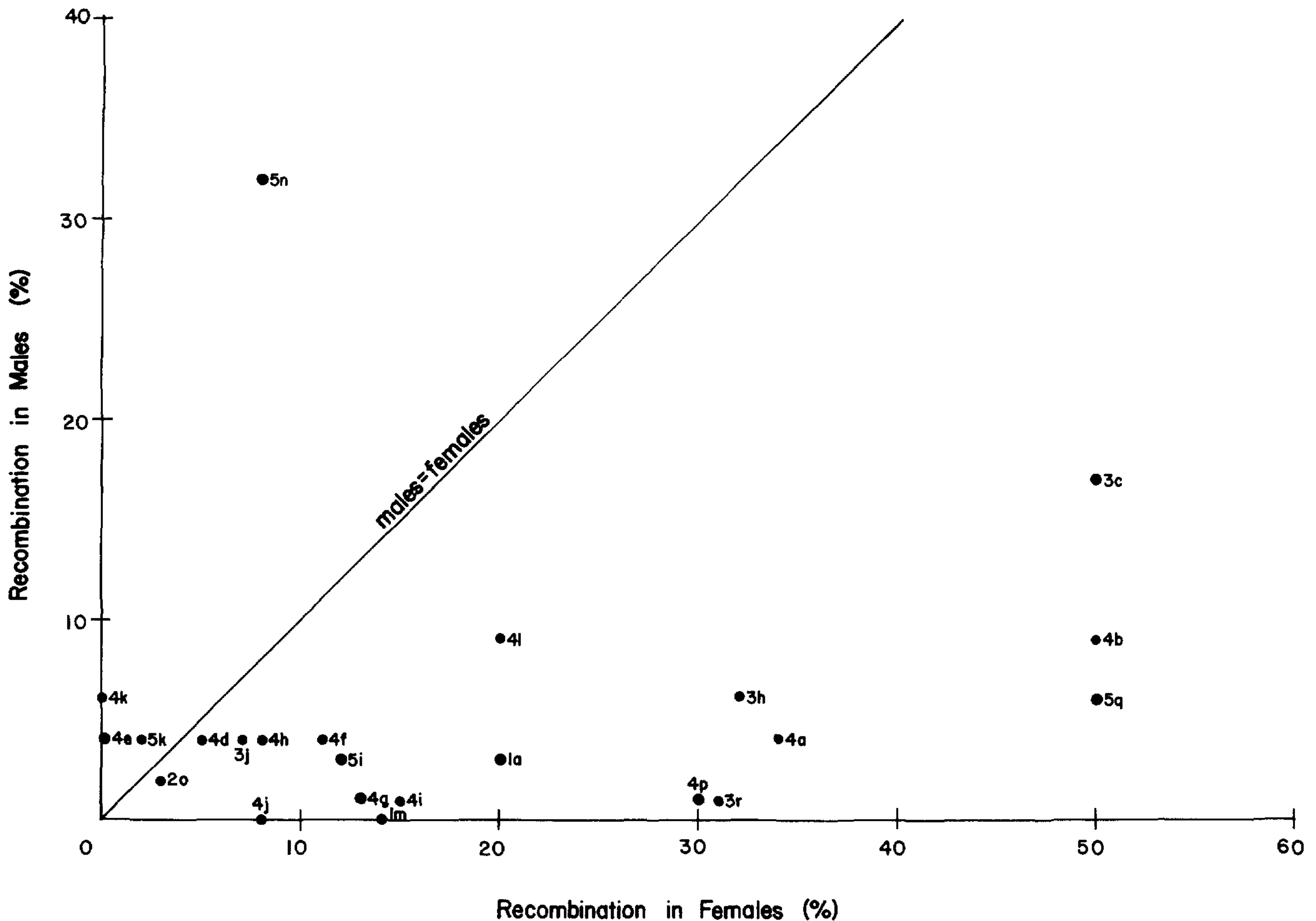

FIGURE 2.—Recombination frequencies in female versus male salmonids. Numbers represent species or hybrids that were tested: 1 = brown trout, 2 = rainbow trout, 3 = cutbow hybrid, 4 = splake hybrid, and 5 = sparctic hybrid. Letters represent locus pairs that were tested: a = *Aat-1* with *Mdh-2*, b = *Aat-2* with *G3p-1*, c = *Aat-2* with *Hgb-1*, d = *Gpi-1* with *PepD-1*, e = *Gpi-2* with *PepD-2*, f = *Gpi-2* with *Sdh-2*, g = *PepD-2* with *Sdh-2*, h = *Idh-3* with *Me-2*, i = *Ada-1* with *G3p-3*, j = *Ldh-3* with *PepB-1*, k = *Ldh-4* with *PepB-2*, l = *Gus* with *Ck-1*, m = *Dia* with *Ck-1*, n = *Ak-2* with *Sod*, o = *Mdh-3* with *Palb-1*, p = *Mpi* with *Gpi-3*, q = *Gpi-3* with *Ldh-1*, and r = *Idh-1* with *Ldh-1*.

KNUDSEN 1983), then *Ak-2* and *Sod* would tend to be inherited as a unit in females. In males, structural constraints imposed by multivalent pairing might limit crossovers to distal locations; recombination between distally located, linked loci such as *Ak-2* and *Sod* might therefore occur at a higher frequency than in females.

Chiasma localizations that would produce sex differences in recombination similar to what we have observed in salmonid fish have been described in the newt *Triturus helveticus* (CALLAN and PERRY 1977). Meiosis in the male newt is characterized by proterminal chiasma localization; essentially all chiasmata are restricted to the ends of the chromosomes. In oocytes, although the chiasma frequency is similar to males, all chiasmata occupy intercalary positions and, therefore, females generally exhibit higher frequencies of recombination than males.

Distribution of loci among chromosomes: Studies of electrophoretic variation usually include only general metabolic enzymes. Are the loci that code for these enzymes randomly distributed among all chromosomes or are they clustered on just a few? Salmonids are ideally suited to test the distribution of loci among chromosomes. Many isozyme loci have already been examined for linkage relationships. Salmonids have many chromosomes which enables testing distributions of loci among many small, similarly sized chromosomes rather than the more difficult task of testing distributions of loci among and within a few large chromosomes.

The many locus pairs examined for joint segregation in western *Salmo* (rainbow, cutthroat and cutbow) and in *Salvelinus* (brook, splake and sparctic) permitted a comparison to be made between the observed number of chromosomes that had a given number of marked loci and the number of chromosomes that would be expected to have the same number of marked loci, if loci were randomly distributed. The expected locus distributions were calculated based on the number of loci tested for joint segregation and

TABLE 5

Chromosomal distribution of marked loci

No. of marked loci	No. of chromosomes			
	Western *Salmo*		*Salvelinus*	
	Observed[a]	Expected[b]	Observed[c]	Expected[d]
0	8	9.15	16	14.52
1	14	11.05	12	14.92
2	5	6.48	9	7.44
3	2	2.46	2	2.40
4	0	0.69	1	0.56
5	1	0.15	0	0.12
Goodness-of-fit test[e]	$\chi^2 = 1.20$, d.f. = 3		$\chi^2 = 1.05$, d.f. = 3	

Distribution of marked loci among the chromosomes of western *Salmo* (rainbow trout, cutthroat trout, and cutbow hybrids) and *Salvelinus* (brook trout, splake hybrids, splake × brook trout backcrosses, and sparctic hybrids).

[a] Observed number of chromosomes having 0, 1, 2, 3, 4 or 5 marked loci, assuming that all linkages were detected among the 35 marked loci tested for joint segregation.

[b] Expected number of chromosomes having 0, 1, 2, 3, 4 or 5 marked loci based on a random distribution of 35 marked loci among 30 chromosomes as calculated from the binomial expansion.

[c] Observed number of chromosomes having 0, 1, 2, 3, 4 or 5 marked loci, assuming that all linkages were detected among the 40 marked loci tested for joint segregation.

[d] Expected number of chromosomes having 0, 1, 2, 3, 4 or 5 marked loci based on a random distribution of 40 marked loci among 40 chromosomes as calculated from the binomial expansion.

[e] Categories of 3, 4 and 5 marked loci were combined for χ^2 analysis.

the approximate haploid number of chromosomes. The observed and expected distributions are shown in Table 5. There were no statistically significant departures of the observed distributions from the expected in either group.

Isozyme loci appear to be randomly distributed among the chromosomes of the salmonid genome. Therefore, isozyme data probably are a reasonable reflection of the entire genome in studies of population structure. Isozyme loci in humans have been found on every chromosome except the Y, and in general, genes for enzymes having similar functions, subunit structures, cellular locations, or metabolic pathways have not as yet been found to be clustered on specific chromosomes (SHOWS 1983).

Evolutionary implications: The following duplicate loci have been tested for joint segregation in at least one salmonid species or hybrid: *Aat-1,2*; *Gpi-1,2*; *Idh-1,2*; *Idh-3,4*; *Ldh-3,4*; *Mdh-1,2*; *Mdh-3,4*; *Me-1,2*; *Palb-1,2*; *PepD-1,2*; *Pgm-1,2*; and *Sdh-1,2*. None of these loci was found to be classically linked with its duplicate in this study or in previous studies (see review by MAY, WRIGHT and JOHNSON 1982). Some linkage groups also appear to be duplicated but not classically linked (Figure 1). Nonlinkage of duplicate loci and duplicate linkage groups is compelling evidence that salmonid genomes arose from tetraploidy and not from tandem duplications.

The fact that some duplicate loci and duplicate linkage groups are pseudolinked but none are classically linked implies that post-tetraploid Robertsonian fusions involved nonhomologous chromosomes. (Fusions of homologs would have resulted in classical linkage of duplicate loci.) The conservation of most pseudolinkage groups among the salmonid species suggests that most of the Robertsonian fusions occurred prior to speciation.

The ancestor of the extant salmonids may have been a segmental allotetraploid (having some auto- and some allotetrasomes). Segmental allotetraploidy could explain why different duplicate loci exhibit different degrees of differentiation, but particular duplicate loci are similarly expressed in all species. Initial hybridization differences, as well as subsequent evolutionary divergence, could have contributed to the differentiation of duplicate loci.

Segmental allotetraploidy could also explain why the degree of duplicate locus divergence may not always correspond with gene-centromere distances, as proposed by ALLENDORF and THORGAARD (1984). For instance, the loss of duplicate gene expression requires structural and functional divergence, yet many of the loci that have lost duplicate gene expression (*e.g.*, *Gpi-3*; *Mpi*; *Sod*; Table 4) are located distal to their centromeres (where interlocus recombination would presumably retard duplicate gene divergence). The retention of allelic commonality between duplicate loci (lack of divergence) requires tetrasomic pairing as well as gene-centromere recombination. Therefore, duplicate genes, even if located distal to the centromere, would not be able to exchange alleles if they were located on chromosomes that did not pair (because of initial hybridization differences). Only in the case of duplicate loci located distal to the centromere on chromosomes known to associate (Figure 1) have we found evidence of interlocus allelic exchange (*Aat-1,2*; *Mdh-3,4*; *Palb-1,2*). In this regard, a distinction should again be made between pseudolinkage and allelic exchange between duplicate loci, both the result of residual tetrasomy. Pseudolinkage, which occurs in hybrid genomes and involves entire chromosomes, is thought to be the result of preferential tetrasomic pairing and directed disjunction. Interlocus allelic exchange, which can occur in pure strains and involves only duplicate loci that are distal to their centromeres, is thought to be the result of tetrasomic pairing and interlocus recombination. However, in many instances the two phenomena are confounded.

Gene-centromere recombination frequency comparisons suggest that some duplicate loci may be located on structurally distinct chromosomes. In cutbow

hybrids, the duplicate loci *Aat-1* and *Aat-2* had similar gene-centromere distance estimates, as did the duplicate loci *Sdh-1* and *Sdh-2* (Table 4). On the other hand, the duplicate loci *Idh-1* and *Idh-2* had statistically different gene-centromere distance estimates (Table 4). *Aat-1,2* and *Sdh-1,2* have shown residual tetrasomic inheritance (Table 3), whereas *Idh-1,2* have not (JOHNSON 1984). Duplicate loci located on structurally distinct chromosomes would not be expected to exhibit tetrasomic inheritance. Gene-centromere recombination frequency comparisons of the duplicate loci *Idh-3* and *Idh-4* in rainbow trout (ALLENDORF *et al.* 1986) and cutbow hybrids (Table 4) suggest that these loci are also located on structurally different chromosomes; residual tetrasomic inheritance of *Idh-3* and *Idh-4* has not been observed (JOHNSON 1984).

Hybridization is an integral first step in models of polyploid evolution in other vertebrate groups (SCHULTZ 1980, BOGART 1980). A disruption of normal meiotic pairing in diploid interspecific hybrids is thought to favor alternative types of gamete formation that could lead to polyploidy. Indeed, female brown trout × Atlantic salmon hybrids have been shown to produce diploid, digenomic eggs (JOHNSON and WRIGHT 1986), suggesting that hybridization may have played a role in the evolution of salmonid tetraploidy. Segmental allotetraploidy is consistent with an initial hybridization event, but also allows for some degree of tetrasomy.

In conclusion, we have presented additional linkage information which supports a high concordance among the genomes of salmonid genera, both classical and pseudolinkage. This conservation is surprising in light of the tremendous differences observed in chromosomal number (GOLD, KAREL and STRAND 1980). These data support the view that most of the structural differences relate to the degree of Robertsonian fusions of acrocentric arms and that limited intra-arm rearrangements have occurred.

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