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September 29, 2000

Joe Huston
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Joe:

We have completed the electrophoretic analysis of the trout collected from Okaga Lake (N=29) in the East Fork Yaak River drainage January 19, 2000. Horizontal starch gel electrophoresis was used to determine each fish's genetic characteristics (genotype) at 43 loci (genes) coding for proteins present in muscle, liver, or eye tissue (TABLE 1). At some of these loci, the rainbow trout, *Oncorhynchus mykiss,* and westslope cutthroat trout, *O. clarki lewisi,* rarely share alleles (form of a gene) in common (TABLE 2). This situation also pertains to a comparison of rainbow and Yellowstone cutthroat trout, *O.c. bouvieri* (TABLE 2). Loci at which such fixed genetic differences exist between taxa are commonly termed diagnostic loci because the alleles detected at them can be used to determine if a sample came from a genetically pure population of one of these taxa or a population in which hybridization between two or all three of these fishes has or is occurring.

The allele frequencies at *LDH-B2** and *sSOD-1** also differentiate redband trout, *O.m. gairdneri,* and coastal rainbow trout, *O.m. irideus.* Redband trout, or those native to waters east of the Cascade Crest, usually possess the *LDH-B2*76* allele at a frequency greater than 0.250 and the *sSOD-1*152* allele at a frequency less than 0.100 (Leary 2000). The only exception to this of which we are aware is some populations in the Deschutes River drainage, Oregon, that have an unusually low frequency of *LDH-B2*76* (Currens et al. 1990; Williams et al. 1997). In contrast, coastal rainbow trout, or those native to waters west of the Cascade Crest, usually possess *LDH-B2*76* at a frequency less than 0.150 and a frequency of *sSOD-1*152* at a frequency greater than 0.100 (Leary 2000).

Alleles characteristic of only rainbow trout were detected at all the loci analyzed in the sample (TABLE 3). With the 29 fish collected and number of diagnostic loci analyzed, we have better than a 95 percent chance of detecting as little as a one percent westslope or Yellowstone cutthroat trout genetic contribution to the population. The Okaga Lake population, therefore, is almost undoubtedly rainbow trout.

Although the frequency of *LDH-B2*76* is on the low border of the range usually observed in redband trout populations and the frequency of *sSOD-1*152* is well within the range usually observed in redband trout populations (TABLE 3), we do not believe the Lake Okaga population is native redband trout. We have never detected the *sSOD-1*152* allele in redband trout populations in the East Fork Yaak River drainage and among the six redband trout populations in the drainage from which we have data the frequency of *LDH-B2*76* ranges from 1.000 to 0.800 (mean 0.919; Sage et al. 1992). Thus, the frequencies of *LDH-B2*76* and *sSOD-1*152* in the Lake Okaga population are well outside the range observed in redband trout populations native to the drainage.

We also strongly suspect the Lake Okaga population is hybridized with coastal rainbow trout. Among 29 populations of redband trout from which we have comparable electrophoretic data, average expected heterozygosity ranges from 0.008 to 0.057 ($\bar{x}$ = 0.028, TABLE 4) and the proportion of polymorphic loci ranges from 0.025 to 0.286 ($\bar{x}$ = 0.103, TABLE 4). These two estimates of the amount of genetic variation in the Lake Okaga population (TABLE 4), therefore, are well outside the range observed in redband trout populations. The simplest explanation for the unusually high levels of genetic variation in the Lake Okaga population is hybridization between the genetically very divergent coastal rainbow and redband trout.

Sincerely,

Robb Leary

Robb Leary

LITERATURE CITED

Currens, K.P., C.B. Schreck, and H.W. Li. 1990. Allozyme and morphological divergence of rainbow trout *(Oncorhynchus mykiss)* above and below waterfalls in the Deschutes River, Oregon. Copeia 1990:730-746.

Leary, R.F. 2000. Letter to Buddy Drake, Drake and Associates, May 26, 2000.

Sage, G.K., R.F. Leary, and F.W. Allendorf. 1992. Genetic analysis of 45 trout populations in the Yaak River drainage, Montana. University of Montana Wild Trout and Salmon Genetics Laboratory Report 92/3.

Williams, R.N., R.F. Leary, and K.P. Currens. 1997. Localized genetic effects of a long-term hatchery stocking program on resident rainbow trout in the Metolius River, Oregon. North American Journal of Fisheries Management 17:1079-1093.

TABLE 1

Enzymes and loci examined. Tissues: E = eye, L = liver, M = muscle.

Enzyme	Loci	Tissue
Adenylate kinase	*AK-1*, AK-2**	M
Alcohol dehydrogenase	*ADH**	L
Aspartate aminotransferase	*sAAT-1*, sAAT-2**	L
	*sAAT-3,4**	M
Creatine kinase	*CK-A1*, CK-A2**	M
Dipeptidase	*PEPA-1*, PEPA-2**	E
Glucose-6-phosphate isomerase	*GPI-A*, GPI-B1*, GPI-B2**	M
Glyceraldehyde-3-phosphate dehydrogenase	*GAPDH-3*, GAPDH-4**	E
Glycerol-3-phosphate dehydrogenase	*G3PDH-1*, G3PDH-2**	L
N-acetyl-beta-glucosaminidase	*bGLUA**	L
Iditol dehydrogenase	*IDDH**	L
Isocitrate dehydrogenase	*mIDHP-1*, mIDHP-2**	M
	*sIDHP-1,2**	L

TABLE 1 — *continued*

Enzyme	Loci	Tissue
Lactate dehydrogenase	*LDH-A1*, LDH-A2**	M
	LDH-B1, LDH-B2*, LDH-C**	E
Malate dehydrogenase	*sMDH-A1,2**	L
	*sMDH-B1,2**	M
Malic enzyme	*mMEP-2**	M
	*sMEP-1,2**	L
Phosphoglucomutase	*PGM-1*, PGM-2**	M
	*PGM-1r**	L
Phosphogluconate dehydrogenase	*PGDH**	M
Superoxide dismutase	*sSOD-1**	L
Tripeptide aminopeptidase	*PEPB**	E
Xanthine dehydrogenase	*XDH**	L

TABLE 2

Diagnostic loci between rainbow and Yellowstone cutthroat trout and between rainbow and westslope cutthroat trout. When more than one allele exists at a locus within a taxon the most common allele is listed first.

	Characteristic alleles			Characteristic alleles	
Locus	Rainbow	Yellowstone	Locus	Rainbow	Westslope
sAAT-1*	100	165	sAAT-1*	100	200, 250
CK-A2*	100	84	CK-A2*	100	84
CK-C1*	100,38	38	GPI-A*	100	92,100
mIDHP-1*	100	-75	IDDH*	100,200,40	40,100
sMEP-1*	100	90	sIDHP-1,2*	100,114,71 40	86,114,100 71,40,20
sMEP-2*	100	110	mMEP-1*	null	88
PEPA-1*	100,115	101			
PEPB*	100,135	135			
PGM-1*	100,null	null			

Note: In westslope cutthroat trout, the frequency of sIDHP-1,2*86 is usually 0.500. In a hybridized population with rainbow trout, therefore, the proportion of westslope cutthroat trout alleles at these loci is the observed 86 allele frequency divided by 0.500.

TABLE 3

Allele frequencies at the loci showing evidence of genetic variation in a sample from what appears to be a hybridized population of coastal rainbow and redband trout in Lake Okaga. H_e = average expected heterozygosity. P = proportion of polymorphic loci.

Locus	Alleles	Allele frequencies
*sAAT-1**	*100*	0.929
	null	0.071
*sAAT-3,4**	*100*	0.922
	95	0.078
*CK-A1**	*100*	0.948
	75	0.052
*bGLUA**	*100*	0.648
	70	0.352
*IDDH**	*100*	0.854
	40	0.146
*mIDHP-2**	*100*	0.839
	140	0.161
*sIDHP-1,2**	*100*	0.741
	114	0.017
	71	0.190
	40	0.052

TABLE 4

Average expected heterozygosity (H_e) and proportion of polymorphic loci (P) in samples from 29 redband trout populations and what appears to be a hybridized population of redband and coastal rainbow trout in Lake Okaga.

Drainage and Population	H_e	P
Upper Columbia River, Washington		
Tonata Creek	0.008	0.056
Barnaby Creek	0.014	0.028
North Fork Deadman Creek	0.014	0.056
Deadman Creek	0.017	0.111
Lane Creek	0.023	0.083
Nancy Creek	0.024	0.083
North Fork Trout Creek	0.025	0.054
West Fork Trout Creek	0.028	0.054
South Fork Chewelah Creek	0.051	0.216
Kootenai River, Idaho and Montana		
Saddle Creek	0.022	0.050
North Callahan Creek	0.025	0.100
South Callahan Creek	0.028	0.100
Callahan Creek	0.031	0.150
Grass Creek	0.035	0.125

TABLE 4 — *continued*

Drainage and Population	H_e	P
Yaak River, Montana		
Basin Creek	0.014	0.050
Royal Creek	0.015	0.025
East Fork Basin Creek	0.016	0.075
West Fork Basin Creek	0.017	0.050
North Fork Yaak River	0.017	0.125
Porcupine Creek	0.019	0.075
East Fork Yaak River	0.021	0.050
Salmon River, Idaho		
Fish Creek	0.020	0.086
Lower Five Mile Creek	0.047	0.286
Upper Five Mile Creek	0.057	0.167
Snake River, Idaho		
Sinker Creek	0.041	0.189
Columbia Creek	0.043	0.118
Cabin Creek	0.048	0.154
King Hill Creek	0.050	0.176
Doby George Creek	0.054	0.147
Means	0.028	0.105
East Fork Yaak River, Montana		
Lake Okaga	0.079	0.368

18 Yellowstone Street
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October 3, 2000

Kenneth Bartz
R.R. #1, 432 Lake Okaga Road
Troy, MT 59935

COPY

Dear Mr. Bartz:

I finally received the genetic analysis results of the fish we collected from Lake Okaga on May 2, 2000. Dr. Robb Leary of the University of Montana Wild Trout and Salmon Genetics Laboratory did the analysis. He determined that most of the genes were from Columbia River redband trout. Redband trout are as native to much of the Yaak River system as well as much of the Kootenai River drainage downstream of Libby Dam.

The balance of the genetic material was of coastal rainbow trout origin and specifically, from Montana's Arlee brood stock. The Arlee rainbow trout strain has existed since the late 1940s and is housed at the states Jocko River Trout Hatchery. I believe that at least two of the sources that provided fish for Okaga Lake in the 1980-1994 era had Arlee-origin brood stock. These hatcheries were Sikokanee Springs and Crystal Lake. The Arlee fish carry a genetic marker that is specific to that strain of rainbow trout. The Arlee strain is actually a mix between Donaldson rainbow and Missouri rainbow.

The greatest value of the genetic testing was that it provided strong evidence that the original fish plant in 1938 was of redband trout. The first owner of Okaga was an Elmer Phillips and he got the first fish from Kenneth G. Drew's "Rainbow Ranch" located at Troy. I also strongly believe that Drews' fish originally came from Kilbrennen Lake. The Drews had a license to take both rainbow trout and brook trout eggs from Kilbrennen Lake from at least 1921 through 1933. Genetic testing done in the mid-1990s showed that Kilbrennen Lake and Kilbrennen Creek rainbow trout were mostly redband trout, not rainbow. Some fish were hybrid redband x rainbow crosses while a couple were redband x rainbow x westslope crosses. Coastal rainbow trout were planted in Kilbrennen in 1945 and 1950 while westslope were stocked in 1988.

Sincerely yours,

Joe E. Huston

cc: Mike Hensler