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Rapid communication: Thirty-eight polymorphic microsatellite markers for mapping in rainbow trout¹

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Species. *Oncorhynchus mykiss*.

Source and Description. Microsatellite repeats were identified by sequencing clones from microsatellite-enriched libraries constructed from genomic DNA. Microsatellite markers were developed by designing primers to amplify the repeats.

Primer Sequences. See Table 1.

Method of Detection. PCR conditions were optimized using rainbow trout DNA (Kamloop strain) and mini-prep DNA from the clone used to develop the marker. PCR reactions included 25 ng of DNA, 1.5 to 2 mM MgCl₂, 1× manufacturer's reaction buffer (Applied Biosystems, Foster City, CA), 2 mM dNTPs, 1 μM each of forward and reverse primer, and 0.05 units of AmpliTaq Gold (Perkin-Elmer, Norwalk, CT). Reactions were thermocycled as follows: 10 min at 94°C, 35 cycles of 94°C for 30 s, annealing temperature for 30 s, and 72°C for 30 s, and a final extension at 72°C for 5 min. PCR

products were analyzed by electrophoresis in 3% agarose stained with ethidium bromide and viewed using a ChemImager (Alpha Innotech, San Leandro, CA). Forward primers of successfully optimized primer pairs were resynthesized labeled with FAM, HEX, or NED fluorescein dyes. PCR was done to amplify the National Center for Cool and Cold Water Aquaculture "Polymorphism Identification and Mapping Panel" of 45 DNA samples that included 25 unrelated rainbow trout (OSU, Arlee, Swanson, Hot Creek, Clearwater, Housecreek, Kamloop, Redband, and Steelhead) and 20 representatives of other salmonids (two to four each, Cutthroat Trout, Sockeye Salmon, Chinook Salmon, Atlantic Salmon, Brown Trout, Brook Trout, and Arctic Char). PCR products were then combined for capillary electrophoresis on an ABI3700. Results were analyzed using Genotyper (ABI, Foster City, CA).

Description of Polymorphism. See Table 1.

Comments. OMM1032 had a 171-bp fragment for all rainbow trout genotyped. Two clones contained more than one microsatellite repeat and each was given an independent designation (OMM1101 and OMM1102, OMM1116 and OMM1117).

¹Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the USDA.

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Table 1. Microsatellite marker information for mapping in rainbow trout

Marker	Library	Repeat	Forward primer sequence	Reverse primer sequence	AT ^a	MgCl ₂ mM	No. of alleles	Size range	% Het ^b	GenBank
OMM1032	TAGA	(AG) ²²	GCGAGGAAGAGAAAGTAGTAG	CCCATCTTCCTCTGATTATG	58	2	9	200–222	5	AF352737
OMM1053	TAGA	(TAGA) ¹⁵	GACTGACACGGGTATTGAA	TATGATATATCCACGGTGTG	56	2	15	247–357	33	AF352738
OMM1054	TAGA	(TATC) ³⁸	AGCCCTTATCTCTGAC	ACATCATCAGTACCGTG	60	2	26	190–325	70	AF352739
OMM1055	TAGA	(AC) ¹⁴	GGCCAAAGCTTTAGTGTG	GCCAGCTTACTCTCATACC	58	2	4	213–220	26	AF352740
OMM1058	TAGA	(TAGA) ¹⁸	GTGTGTATGTGGTTTCAC	CCAATGAGAAAGCGTTAC	60	2	1	174–227	45	AF352741
OMM1059	ATG	(CAT) ⁹	CGCCAGATGATTAAACGA	GGGCTATTCACACGTTCA	58	2	6	182–193	35	AF352742
OMM1061	ATG	(ATC) ⁸	GCGGTCACTGTCAAGA	GGGAGACATGGATCATA	62	2	4	222–275	12	AF352743
OMM1064	TAGA	(GATA) ¹⁹	AGAATGCTACTGTGGTGTATTGTGA	TCTGAAAGACAGGTGGATGGTTCC	60	2	12	146–199	68	AF352744
OMM1073	ATG	(GTGA) ¹³	CATATGTCAAAGTGTGTCTCCA	CTATCAGTCAGCAGGCTTTCATGATC	58	2	6	134–174	24	AF352745
OMM1075	TAGA	(TATC) ⁷	ACGCAACCCAGACAGGTAAAGA	GCGCTGACAAGAAGAACAC	58	2	13	203–284	74	AF352746
OMM1076	TAGA	(TATC) ²⁶	AACCCGACACCTAGGACACCT	CTTACACCATGCCACGGAAGTTA	64	1.5	15	171–277	55	AF352747
OMM1077	TAGA	(GATA) ⁹	GGCTGACAGAGAAAGACTAGTTC	TGTTACGGTGTCTGACATGC	58	2	3	225–262	0	AF352748
OMM1078	TAGA	(TAGA) ²⁴	AACTCACGCCCTGACCAACCTAAC	GATTTCAAGTATTGGTCCGAGCC	64	2	20	184–298	65	AF352749
OMM1080	TAGA	(GATA) ¹⁴	GAGACTGACACGGGTATTGA	GTTATGTTGTCATGCTAGGG	58	2	15	206–303	52	AF352750
OMM1081	TAGA	(TATC) ¹⁷	CCGTTGTATAACCAATGACC	TCTTTACACAGAGGGTCTTAC	56	2	16	151–258	78	AF352751
OMM1082	TAGA	(GATA) ¹⁷	CAAGAGCACTAACGACCATGT	CGCAAGCAAGCTAACACA	58	2	13	166–223	86	AF352752
OMM1083	TAGA	(GATA) ²⁶	GCCCTGACCAACCTAACACA	TGCTGACATTCGGTTAGTAGTGG	58	2	17	122–232	41	AF352753
OMM1084	TAGA	(GATA) ²⁰	CGAGACAAGCAGCCAGATAGAG	CACCTGACTGTCTCTTGGCTATC	58	2	10	198–242	71	AF352754
OMM1086	TAGA	(TATC) ⁷	GTATGCTTTCACAAATTGCATGT	CCTGTTCAAGTCAAACTCAC	64	2	9	186–223	45	AF352755
OMM1087	TAGA	(TCTA) ¹³	GACGCAGAAAGTTTAGCTCT	TTACTGTCTCTCTGCAGCA	58	2	13	237–291	71	AF352756
OMM1088	TAGA	(GATA) ¹²	CTACAGGCCAACACTACAATC	CTATAAAGGGAATAGGCACCT	58	2	12	113–170	78	AF352757
OMM1089	TAGA	(GATA) ¹³	GCAGCTCCTGTTTCTATGTG	CTGAGATGCAGTGCCCTTAGAC	58	2	17	111–238	26	AF352758
OMM1090	TAGA	(AG) ⁹⁰ (TAGA) ¹⁰	TGCGGTAGGAAGGCTTTAGTG	AAATGGAGCAGCGCTGGTAT	64	2	11	253–394	26	AF352759
OMM1093	TAGA	(TO) ²⁶	CCGTTATCTGCCAGTTCACTCTC	CGGCTGCACCTGTGAGATAGAGA	58	2	10	269–303	16	AF352761
OMM1096	TAGA	(TATC) ³⁴	CTCGCTTTATTTGATCATGTTCTACTG	AGAGATCAGTGGCAGCTTAGGG	64	1.5	19	124–275	48	AF352762
OMM1097	TAGA	(GATA) ²⁶	CTAGCCATCCGAACACTG	AGAAATAGGTGCTGTATCTC	64	2	20	201–304	68	AF352763
OMM1100	TAGA	(GATA) ¹⁴	AGCTTGTCCCTTATCCTT	GCCATAGTTATGATCC	58	2	11	193–229	65	AF352764
OMM1101	TAGA	(ATAG) ³¹	CTGCCCTCTGATTGAGAACCATATC	CCGTGTCAGATGAATGGG	62	2	15	139–228	68	AF352765
OMM1102	TAGA	(ATAG) ³¹ (AC) ¹⁹	CGGCCCTCTGCTGTGATCCAATAT	CTGACTTCAATCTGAGCCGATGAG	64	2	14	261–298	32	AF352765
OMM1104	TAGA	(GATA) ¹⁵	AACAGGCCCTGATGAGTTTC	CTCTCTGTCTCGCTCCTATTG	58	2	15	166–227	77	AF352767
OMM1105	TAGA	(AGAC) ²³ (GATA) ¹⁶	GCACACTGTGCGGTAAGAGA	GACAGCCACACTAAACCA	62	2	12	131–200	36	AF352768
OMM1108	TAGA	(TCTA) ¹⁴	CACAGCTGTGAAACATGCCGTAAT	TCACAGTCGGTCCAGTCTCG	58	2	10	160–203	43	AF352769
OMM1109	TAGA	(AC) ³⁰	GGCAACAACCCACCCAAACCAATCTA	AGCACCAAGATCGAACTCC	62	2	2	113–119	9	AF352770
OMM1116	TAGA	(GA) ¹¹ (GATA) ¹⁹	GACAAAGACAGAGGGACGA	AGCAATGGGCTCTATGACTGAT	58	2	3	183–223	59	AF352771
OMM1117	TAGA	(GATA) ¹⁹	AAGCCAGAGGGGATAAGATG	TTGGTGTTCACAGGACAGTAA	62	2	12	217–251	19	AF352772
OMM1120	TAGA	(GATA) ¹¹	TTGAAGACAAAGTACGCGAGAG	CCTGCTATTGTACATGCTAC	62	2	8	121	0	AF352773
OMM1122	TAGA	(TAGA) ⁹	TACATCAACAGGTCATTTGTG	TTCTCATCCCATCTACCATCC	58	2	1	117–132	39	AF352774
OMM1125	TAGA	(AC) ⁴¹ (GT) ¹² (AG) ⁴⁹	GGAGATTGGGTGAGAGCTAAA		64	2	8			

^aAnnealing temperature.^bHeterozygosity.