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TECHNICAL NOTE

Development a Set of 46 SNP Markers for the Yellow Catfish (*Tachysurus fulvidraco*)

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Abstract

The whole genome resequencing was used to develop single nucleotide polymorphisms (SNP) markers for the yellow catfish (Tachysurus fulvidraco). A total of 46 SNP markers from 129 individuals were selected from 5550676 genotyping markers which distributed on 26 chromosomes. Of the 46 SNPs analyzed, 35 SNPs conformed to Hardy-Weinberg equilibrium. The observed and expected heterozygosity of these markers ranged from 0.2519 to 0.771 and from 0.265 to 0.5018, respectively. This set of markers will be of great useful for population genetics of the yellow catfish.

Introduction

The yellow catfish (*Tachysurus fulvidraco*), belonging to Bagridae (Nelson et al., 2016), is an important economic species of freshwater fish, widely distributed in China. Due to its delicious taste and high nutritional value, the yellow catfish was favored by the market. With the increasing market demand, this species has suffered over- exploitation and declined sharply of its natural stocks over the past few years (Guo et al., 2006; Yu et al., 2019). Therefore, it is essential to understand the population genetics of yellow catfish for the conservation and sustainable exploitation.

Precious genetic studies of this species have been explored using SSR markers (Feng et al., 2009; Hu et al., 2009; Zhang et al., 2009). Compared with the SSR marker, the SNP marker has significant advantages, e.g., high throughput, site-specific, low error-detection rate, which motived the development of SNP marker for the population genetics of the yellow catfish (Li et al., 2018; Meyer-Sand et al., 2018). Here we characterized a set of 46 SNP markers for the yellow catfish which were analyzed using whole genome resequencing.

Materials & methods

The genomic DNA from 129 individuals was sequenced using Illumina HiSeq™ X Ten platform (Zhou et al., 2021). After reads quality filter and mapping to the reference genome (Gong et al., 2018), 46 SNP markers were randomly selected from 5550676 genotyping markers which distributed on 26 chromosomes. The genetic parameters of these markers, including observed heterozygosity, expected

heterozygosity, and the Hardy–Weinberg equilibrium (HWE), were calculated using Popgene v 1.32 (Yeh et al., 1997).

The marker primers were designed by the flanking sequence using Oligo 7 (Rychlik, 2007) software and examined for their amplification efficiency. PCR were conducted in 50 µL reaction mixtures containing 200 ng template DNA, 5 µL 10 × buffer (TaKaRa, Dalian, China), 4 µL $MgCl_2$ (2.5 mol/L), 2.5 µL dNTP (2.5 mmol/L), 2 µL of each primer (5 µmol/L), and 1 U Taq DNA polymerase (25 U/µL, TaKaRa). PCR conditions were as follows: initial denaturation (95°C, 2 min), then 34 cycles of denaturation (94°C, 40 s), primer annealing (50 s), and elongation (72°C, 60 s) and a final extension (72°C, 10 min).

Results

All 46 SNP loci were amplified successfully. The observed heterozygosity of these markers varied from 0.2519 to 0.771, while the expected heterozygosity ranged from 0.265 to 0.5018 (Table 1). A total of 35 loci conformed to Hardy-Weinberg equilibrium (P>0.01) after Bonferroni's correction, ensuring the reliability of their application to evaluate larger groups.

Ethical Statement

The experimental protocol was established, according to the ethical guidelines of the Basel Declaration and was approved by the Experimental

Table 1. Characterization of 46 SNP markers in *Tachysurus fulvidraco*

SNP ID	Chr. NO.	Primer sequence (5′–3′)	Tm (℃)	Size (bp)	SNP position (bp)	SNP type	*Ho*	*He*	HWEP
55823	1	AGGAGCTTTACAAACACTCAC	52	591	331	A/C	0.3664	0.3285	NS
		CTGTTTGCCTAAAATGCTCCA	53						
2071	1	TTTAAATAAGCACAAATAGCAA	45	571	87	A/C	0.4504	0.3586	NS
		AAACTTCCAGAATGTCCCC	52						
58894	1	CCAAATAATGAAGCGAAAGCC	52	681	310	C/A	0.3969	0.4661	NS
		ACAATGCAGGAATGATAACCAG	53						
52924	1	CTGCCCATGGTCATAATTGGA	55	653	428	C/T	0.3817	0.4258	NS
		TGCCACTACCTTATTATCACT	50						
560625	2	AACCGTTAAAATTAGAGATCACAC	50	586	393	G/T	0.4198	0.3966	NS
		CAACTTAAGCACAACTCGGTA	52						
599487	2	GAATCTGATCACTGAGCTCGT	54	468	226	C/T	0.6870	0.4806	*
		TTTCAAGAACCAGCATGGAC	53						
638351	2	AGTTCTGCCTTCAACACC	52	877	378	T/C	0.2977	0.2955	NS
		TAGTGCCATAGAAAATAGCTT	48						
638434	2	TTCTTACCGTAACTCATGC	49	603	367	G/T	0.2519	0.3053	NS
		CACGTCTATGAATCTACTGC	50						
760278	3	CCAACGTGCTATGGTCT	52	584	278	G/C	0.4351	0.5014	NS
		ACAAACTGTCTTAAAGGGCTC	52						
938090	3	TCATTTTCCTCTTCATGCACT	51	565	351	A/G	0.6947	0.4851	*
		TAGCCAGACATCCCGAA	53						
1149005	4	CAACTCCACAGATGCACT	52	364	99	A/T	0.6183	0.4790	NS
		CAACATCCCCTAATTAATCCAC	51						
1149027	4	ATTTCAAGCCTTATTGCTC	47	694	186	A/G	0.5802	0.4574	NS
		CTTTTACAACCAAGTGCAT	47						
1487601	5	AAATAGGCTTCATGTGT	44	501	275	T/G	0.6183	0.4756	NS
		TCTTCTTTAACTGGCATC	46						
1798440	6	ACATATACATACGCACACGTT	51	498	367	T/C	0.5420	0.4990	NS
		CCATGTCTGTAATTTTCACGTT	50						
1855240	7	CAAGCTTATGTTAAAGGCTCT	50	570	125	A/G	0.6641	0.4756	*
		AACTAACGATGTCACACGCTA	53						
2265537	9	ATCGTCCATGACAAATCACC	52	620	149	C/T	0.5420	0.3966	*
		TCTGTTTATTCTCATAGTGCAA	48						
2477173	10	TAGTGAATAATCATAGGGCTT	47	540	231	G/T	0.3206	0.2906	NS
		TATTTCCACAACACGCTCA	51						
2597747	10	ATCCTAGCCAATGTATTGCTG	52	603	398	C/T	0.2977	0.2650	NS
		TGTTAGGTATCGCCACT	49						
2676641	11	GCCTTTGTTCTTATAGTGCAT	50	603	337	A/G	0.5954	0.4836	NS
		CACTGAGCTCCTGTTACACAC	56						
2941890	12	TTAGTTGCAGGTTCCTC	48	658	154	A/C	0.5802	0.4701	NS
		AAGTAAGCCTGTATTAGCC	48						
3171163	13	TTGCCACAAGGTATAACCGAA	54	419	327	A/T	0.4275	0.3931	NS
		GGCTCTAGTATTTTATCAAACCC	50						
3171175	13	AATCAGAGCGTTTGAACCAT	52	403	119	T/G	0.6107	0.5012	NS
		CAATTCCCTAATTTGCCCTT	50						
3171187	13	CAGAATCAGTTATGGCCCTC	53	693	317	A/G	0.5725	0.5014	NS
		TTACTTCTGGCTAATTACTGG	49						
3171263	13	ATCACACTCATTCGGCTT	51	558	305	T/C	0.2672	0.3147	NS
		AAGGCTATCATGCACCCAA	54						

Table 1. Continued

SNP ID	Chr. NO.	Primer sequence (5′–3′)	Tm (℃)	Size (bp)	SNP position (bp)	SNP type	H_O	H_e	HWEP
3171347	13	CAGCCATTAAAATATAGGGAG	47	451	129	A/G	0.6260	0.4773	NS
		CAACACAATTTCGCTGGA	50						
3319329	14	GCCTCCACCTAGACTCC	55	423	239	A/G	0.5344	0.4574	NS
		AGAATGCGATGTTTACTGCT	51						
3485859	14	CTAATTGTGCCTTGTATGCAG	51	380	89	G/T	0.5649	0.4619	NS
		TGTAAATAATCAGAGCGTGT	48						
3541374	15	ACGTGTACAACTGTTATTACGC	53	405	233	G/C	0.5573	0.4756	NS
		GTGTCCGCATCATAAACGAA	53						
3542145	15	CTAATCTCAGCACATCCG	50	450	157	C/T	0.5267	0.4228	NS
		AATCAGATGAAAGGCACAC	50						
3706234	16	TATTGGCAACAACCAGT	48	537	372	G/T	0.5573	0.4756	NS
		GCATAAGACACATAACGCATC	51						
4029832	17	GTACTGCCAATTTACACAAC	49	576	385	G/C	0.6794	0.4720	*
		AAAATTAAAAGCCTTATTAGTCCA	47						
4030097	17	ACGCTGTTGTCCTACCTG	55	426	182	A/G	0.6336	0.4681	*
		CCCATTTTCTAGGGGTCCTT	54						
4057632	17	ATGCTTCATTTCATCCGTTC	50	417	275	C/T	0.6794	0.5018	*
		ACCCGCCTTAAATTTGGAC	53						
4174143	18	GTGACCCGATTTTACATGACC	54	470	296	A/T	0.6183	0.4720	NS
		GCGTTGTGGAATTCTTATCACT	53						
4329563	19	CCTTCATCTTCTGGATTTTGCG	54	561	291	C/T	0.5115	0.4102	NS
		TGACTGCCAACAGATTACCCAA	56						
4540487	20	CTGTAGGAAAACACCGCTCT	55	653	330	A/G	0.6107	0.4701	NS
		TGCCAATCAACCTACCATGC	57						
590446	21	ACAGAGACATCAGTGCAA	50	418	194	A/T	0.6947	0.4902	*
		GACAAAACTCCTTCGGAT	49						
4931481	22	CCTTAAACACTAGGCTACCAC	53	659	392	C/T	0.6260	0.4914	NS
		GAGAAAAGATTTACGCTGCT	50						
4931542	22	ATAAGTTGCACCAGCGTA	51	522	105	G/T	0.5344	0.4528	NS
		GCTGGTCATTTAATAACTAACAC	49						
4931576	22	TCAGCCGTTACTAGGTT	49	657	359	A/G	0.6947	0.4902	*
		CCACAGACTGACGACCA	54						
4931603	22	CTGCCTCTATGCCCGAT	55	531	310	G/T	0.4885	0.4069	NS
		AACTGCTGCCTTTGAGACTG	55						
5028968	23	GAAAGCTGCTAGACACA	48	584	328	A/C	0.5344	0.4661	NS
		GTTGGTAATTTTAGCCCTC	48						
5078926	23	ATGCTAACCACTAAGCCA	50	631	453	C/T	0.6031	0.4821	NS
		CCAATTCACATATGCCTT	46						
5206582	24	ATCTTGGTGACTTTGCGCTT	55	539	246	A/G	0.6641	0.4790	*
		GCGGTTGCCACTATAATCTCA	55						
5232887	25	GCAAATAAGAAGCACGA	46	628	308	C/T	0.5725	0.4720	NS
		CAAGCACCAAGTTAGCTC	51						
5456365	26	TGGCTTACACCTGATATAGAGGAC	55	452	177	G/T	0.7710	0.4977	*
		CAGCGCAAAAGCTTAGCACCA	59						

H_O represents the observed heterozygosity, H_e represents the expected heterozygosity, HWEP represents the results for Hardy–Weinberg equilibrium, NS represents non-significant, *P<0.01.

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Author Contribution

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Huaxing Zhou, Guoqing Duan, Huan Wang and Tingshuang Pan. The first draft of the manuscript was written by Huaxing Zhou and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Conflict of Interest

The authors declare there are no competing interests.

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