

**Rapid, Specific and Sensitive Detection of *Vibrio Parahaemolyticus* in Seafood by  
Accelerated Strand Exchange Amplification**

Ritong Sun,<sup>a</sup> Jiao Chen,<sup>a</sup> Yingeng Wang,<sup>b</sup> Zheng Zhang,<sup>b</sup> Yong Li,<sup>a</sup> Fengmei Li,<sup>a</sup>

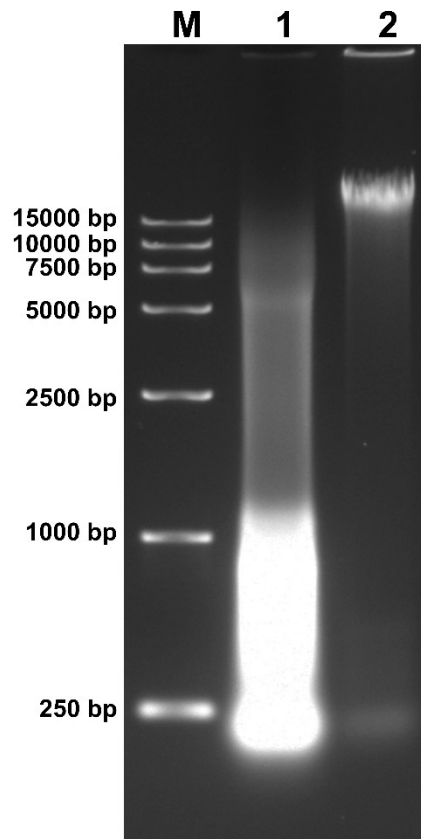
Cuiping Ma,<sup>a</sup> Qingxia Han,<sup>c</sup> and Yanjing Shi<sup>a,\*</sup>

<sup>a</sup> *Shandong Provincial Key Laboratory of Biochemical Engineering, Qingdao Nucleic Acid Rapid Detection Engineering Research Center, College of Marine Science and Biological Engineering, Qingdao University of Science and Technology, Qingdao, 266042, PR China.*

<sup>b</sup> *Yellow Sea Fisheries Research Institute, Chinese Academy of Fishery Sciences, Qingdao, 266071, PR China.*

<sup>c</sup> *Bin Zhou Polytechnic, Qingdao, 256600, PR China.*

\*Corresponding author. Tel (Fax): +86-0532-84022929; E-mail: [syj99@163.com](mailto:syj99@163.com)



**Fig. S1** The agarose electrophoresis of *V. parahaemolyticus* genomic DNA. Lane M represented a 15000 bp DNA ladder. Lane 1 represented genomic DNA generated by nucleic acid release agent and treated with RNase A. Lane 2 represented genomic DNA extracted by TIANamp Bacteria DNA Kit.

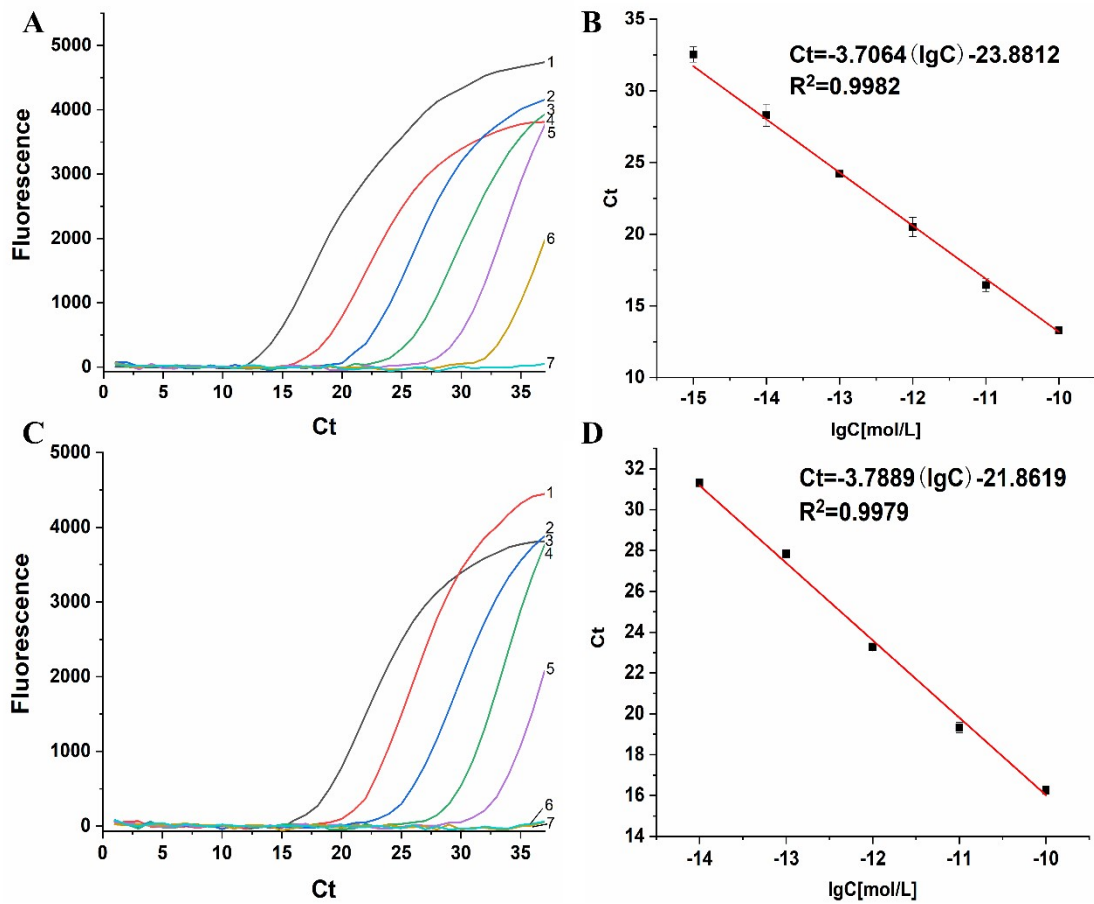


Fig. S2 The comparison of ASEA and real-time PCR in detecting pure *V. parahaemolyticus* genomic DNA. (A) The amplification curves of ASEA. (B) Relationship between the Ct values of ASEA reaction and the logarithmic values of the amount of genomic DNA targets. (C) The amplification curves of real-time PCR. (D) Relationship between the Ct values of real-time PCR reaction and the logarithmic values of the amount of genomic DNA targets in ASEA. Error bars showed the mean standard deviations of three determinations. 1-7 represented 10-fold dilution from the *V. parahaemolyticus* genomic DNA concentration of  $7.6 \times 10^{-11}$  M to  $7.6 \times 10^{-16}$  M as well as no template control, respectively.

**Table S1** List of *ToxR* Target region and SEA species-specific primers

Primer	Sequence (5'-3')
<i>Vibrio parahaemolyticus</i>	<u>CCTTACCTACTCTTGACATCCAGAGAACTTT</u>
( <sup>a</sup> HF564887.1, <sup>b</sup> 982-1027)	<u>CCAGAGATGGATTGG</u>
SEA-F	CCTTACCTACTCTTGACATCCAG
SEA-R	CCAATCCATCTCTGGAAAGTTC

<sup>a</sup>GenBank accession number, <sup>b</sup>the position of specific sequence in genomic DNA; the underlined sequence in the *ToxR* target region was used to design SEA-F primer, and the dotted sequence in the *ToxR* target region was the reverse complement sequence of the SEA-R primer.

**Table S2** ASEA and real-time PCR of 56 samples

Kind of samples <sup>#</sup>	Sample Number <sup>#</sup>	Cycle of threshold (Ct)	
		ASEA for	Real-time PCR for
		<i>V. parahaemolyticus</i>	<i>V. parahaemolyticus</i>
<b>Shrimps</b>	1-14	ND	ND
	15-26	ND	ND
<b>Sea fishes</b>	27	24.71	28.56
	28	25.12	29.43
	29-33	ND	ND
<b>Oysters</b>	34	29.18	31.66
	35-41	ND	ND
	42	27.49	30.34
<b>Scallops</b>	43-51	ND	ND
	52	ND	32.17
	53-56	ND	ND
<b>DNase/ RNase-free</b>	-	ND	ND

ND: not detected.

<sup>#</sup>The 56 samples came from 14 shrimps, 14 Sea fishes, 14 Oysters, 14 Scallops, and were numbered according to 1-56.