

1 Supplemental materials

2
3 **Figure S1**
4
5
6

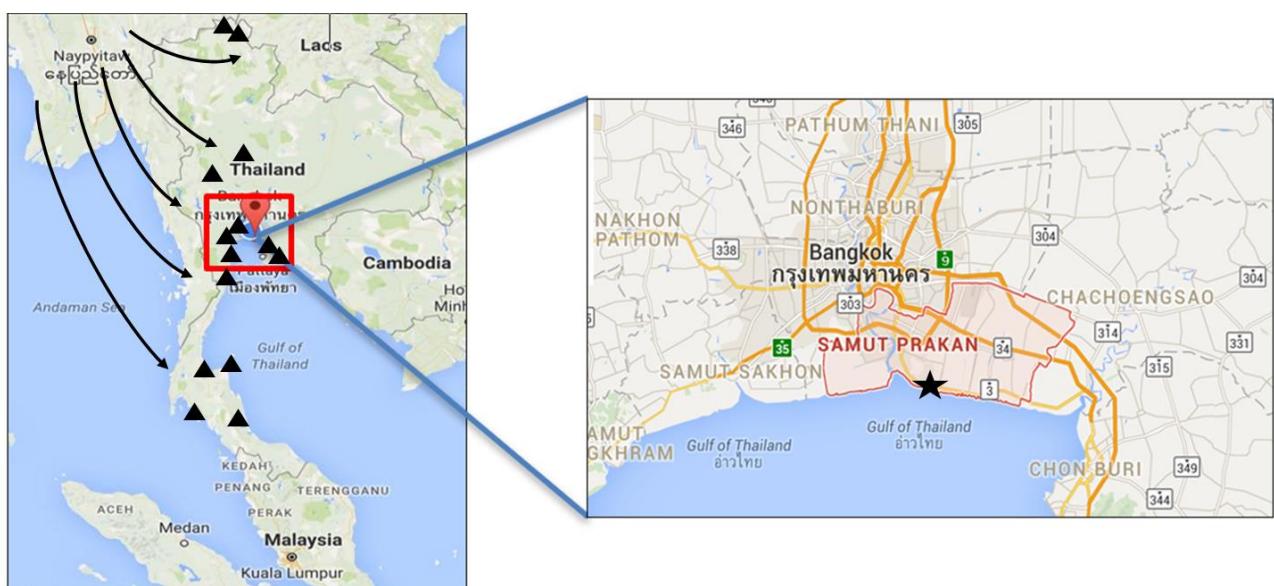


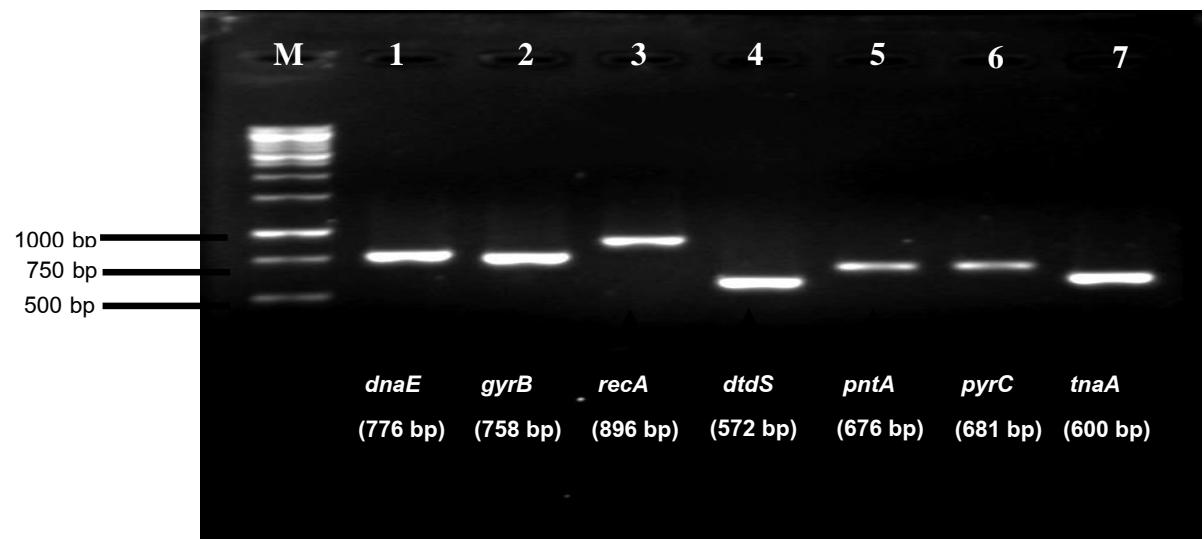
Figure S1 Map of sampling site at Bangpu Recreational Center (black star), Samut Prakan, Thailand. Arrows represent migratory direction of Brown-headed gull (*Chroicocephalus brunnicephalus*) from central Asia to inland lakes and coastal regions in southern Asia including Thailand. Triangles represent range map of *C. brunnicephalus* in Thailand. Data provided by Thai National Parks (2022).

16

Figure S2

17

18



19

20

21

22 **Figure S2 The 1% (w/v) agarose gel electrophoresis of *V. parahaemolyticus* PCR products**
23 **of 7 housekeeping genes positive control of strain VP902. Lane M: 1 kb ladder (Thermo**
24 **Scientific, USA); Lane 1-7: PCR products of 7 housekeeping genes including *dnaE* (776**
25 **bp), *gyrB* (758 bp), *recA* (896 bp), *dtdS* (572 bp), *pntA* (676 bp), *pyrC* (681 bp) and *tnaA***
26 **(600 bp), respectively**

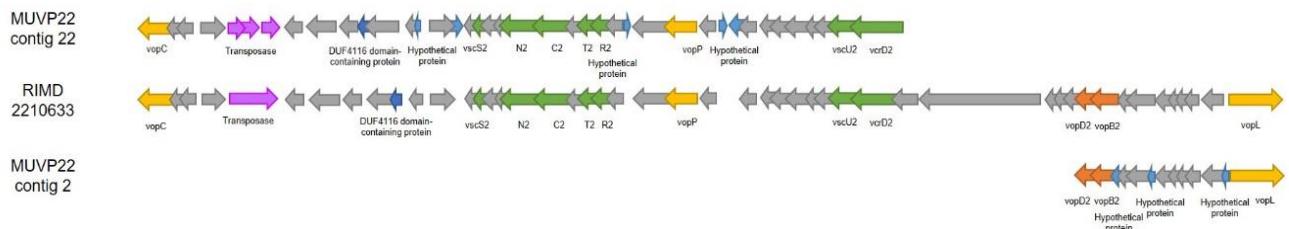
27

28

Figure S3

29

30



31 **Figure S3 Comparison of the gene organization of T3SS α -related genes in *Vibrio*
32 *parahaemolyticus* RIMD2210633 and MUVP22. Green arrows indicate the gene encoding
33 putative apparatus protein of T3SS2 α , yellow arrows indicate the gene encoding putative
34 effector protein of T3SS2 α , orange arrows indicate the gene encoding putative translocon
35 protein of T3SS2 α , purple arrows indicate transposase genes, dark blue arrows indicate
36 rearranged DUF4116 domain containing protein, light blue arrows indicated additional
37 hypothetical protein, and gray arrows indicate other proteins.**

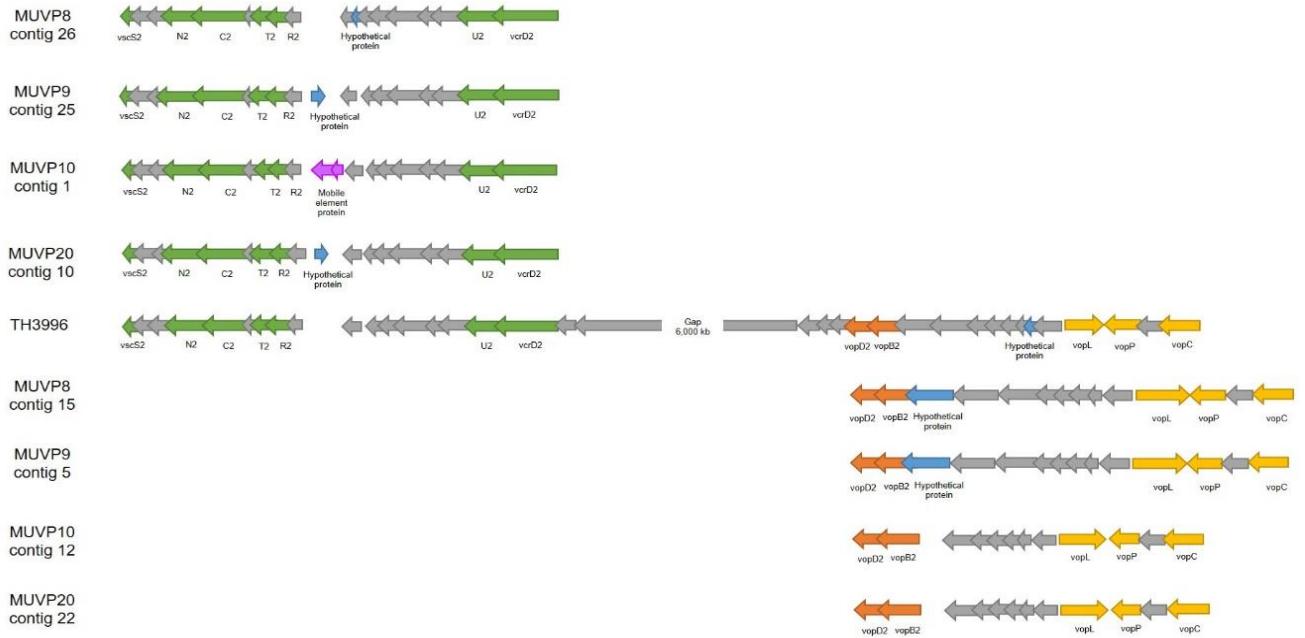
38

39

40

Figure S4

41



42

43

Figure S4 Comparison of the gene organization of T3SS2 β -related genes in *Vibrio parahaemolyticus* TH3996, MUVP8, MUVP9, MUVP10 and MUVP20. Green arrows indicate the gene encoding putative apparatus protein of T3SS2 β , yellow arrows indicate the gene encoding putative effector protein of T3SS2 β , orange arrows indicate the gene encoding putative translocon protein of T3SS2 β , purple arrows indicate mobile element protein, light blue arrows indicated additional hypothetical protein, and gray arrows indicate other proteins.

51

52 **Table S1 Nucleotide sequence of primers used for detection of species specific and**
 53 **virulence genes of *Vibrio parahaemolyticus***

54

Gene	Sequence (5' to 3')	Product size (bp)	Reference
<i>ldh</i> ^a	Forward 5'AAAGCGGATTATGCAGAAGCACTG3' Reverse 5'-GCTACTTTCTAGCATTCTCTGC-3'	450	(31)
<i>tdh</i> ^b	Forward 5'-GTACCGATATTTGCAAA-3' Reverse 5'-ATGTTGAAGCTGTACTTGA-3'	382	(33)
<i>trh</i> ^b	Forward 5'-CTCTACTTGCTTCAGT-3' Reverse 5'-TACGCTTATATAGGCGCTTA-3'	276	(34)
T3SS1	Forward 5'-TCGGTTAGCGAAGGCGTA-3' Reverse 5'-CCGCTGATAATGCCAGTA-3'	716	(16)
VP1680 ^c (<i>vopQ</i>)	Forward 5'-CACTGATGGCGTATCTGGTC-3' Reverse 5'-GTCAAGCTGAGCGCCAG-3'	386	(35)
T3SS2 α ^c (<i>vopP</i>)	Forward 5'-ATTCACCTTGACTTCTCTGC-3' Reverse 5'-ACCTAAAGATAACCGCTGCT-3'	379	(81)
T3SS2 β ^c (<i>vopC</i>)	Forward 5'-AACCAACTTGCAGACTAAATC-3' Reverse 5'-TCCCGACAGTTCTGCAC-3'	594	(32)

55 ^aspecies specific

56 *ldh* = lecithin dependent hemolysin

57 ^bvirulence genes

58 *tdh*= the mostable direct hemolysin

59 *trh*= the mostable direct hemolysin-related hemolysin

60 ^cType 3 secretion system genes

61 T3SS1 = Type 3 secretion system 1

62 T3SS2 α = Type 3 secretion system 2 α

63 T3SS2 β = Type 3 secretion system 2 β

64

65

66 **Table S2 List and properties of 49 *V. parahaemolyticus* isolates from aquatic bird fecal
67 samples**

No.	Isolate name	Date of isolation	Virulence genes				
			<i>tdh</i>	<i>trh</i>	T3SS1	T3SS2α	T3SS2β
1	MUVP1	22/08/2016	-	-	+	-	-
2	MUVP2	22/08/2016	-	-	+	-	-
3	MUVP3	22/08/2016	-	-	+	-	-
4	MUVP4	5/9/2016	-	-	+	-	-
5	MUVP5	5/9/2016	-	-	+	-	-
6	MUVP6	5/9/2016	-	-	+	-	-
7	MUVP7	5/9/2016	-	-	+	-	-
8	MUVP8	5/9/2016	-	+	+	-	+
9	MUVP9	3/10/2016	-	+	+	-	+
10	MUVP10	17/10/2016	-	+	+	-	+
11	MUVP11	17/10/2016	-	+	+	-	+
12	MUVP12	31/10/2016	-	-	+	-	-
13	MUVP13	31/10/2016	-	-	+	-	-
14	MUVP14	31/10/2016	-	-	+	-	-
15	MUVP15	31/10/2016	-	-	+	-	-
16	MUVP16	30/01/2017	-	-	+	-	-
17	MUVP17	30/01/2017	-	-	+	-	-
18	MUVP18	14/02/2017	-	-	+	-	-
19	MUVP19	14/02/2017	-	-	+	-	-
20	MUVP20	13/03/2017	+	+	+	-	+
21	MUVP21	27/03/2017	-	-	+	-	-
22	MUVP22	27/03/2017	+	-	+	+	-
23	MUVP23	27/03/2017	+	-	+	+	-
24	MUVP24	27/03/2017	+	-	+	+	-
25	MUVP25	27/03/2017	-	-	+	-	-
26	MUVP26	14/11/2016	-	-	+	-	-
27	MUVP27	14/11/2016	-	-	+	-	-
28	MUVP28	14/11/2016	-	-	+	-	-
29	MUVP29	14/11/2016	-	-	+	-	-
30	MUVP30	28/11/2016	-	-	+	-	-
31	MUVP31	28/11/2016	-	-	+	-	-
32	MUVP32	13/12/2016	-	-	+	-	-
33	MUVP33	13/12/2016	-	-	+	-	-
34	MUVP34	13/12/2016	-	-	+	-	-
35	MUVP35	13/12/2016	-	-	+	-	-
36	MUVP36	30/01/2017	-	-	+	-	-

Table S2 List and properties of 49 *V. parahaemolyticus* isolates from aquatic bird fecal samples (cont.)

No.	Isolates name	Date of isolation	Virulence genes				
			<i>tdh</i>	<i>trh</i>	T3SS1	T3SS2α	T3SS2β
37	MUVP37	30/01/2017	-	-	+	-	-
38	MUVP38	30/01/2017	-	-	+	-	-
39	MUVP39	30/01/2017	-	-	+	-	-
40	MUVP40	14/02/2017	-	-	+	-	-
41	MUVP41	27/02/2017	-	-	+	-	-
42	MUVP42	13/03/2017	-	-	+	-	-
43	MUVP43	13/03/2017	-	-	+	-	-
44	MUVP45	28/04/2017	-	-	+	-	-
45	MUVP46	28/04/2017	-	-	+	-	-
46	MUVP47	28/04/2017	-	-	+	-	-
47	MUVP48	28/04/2017	-	-	+	-	-
48	MUVP49	28/04/2017	-	-	+	-	-
49	MUVP50	28/04/2017	-	-	+	-	-

68

69

70