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APPENDIX

APPENDIX A

The table of clinical and raw data.

No	Year	Age	Multiplex RT-PCR					
			PBMC	Plasma	Pellet	Saliva	Oral brush	Serum
1	47	9	D1	D1	N/A	N/A	N/A	N/A
2	47	14	D4	D4	N/A	N/A	N/A	N/A
3	47	13	D4	D4	N/A	N/A	N/A	N/A
4	47	14	D4	D4	N/A	N/A	N/A	D4
5	47	7	D1	D1	N/A	N/A	N/A	N/A
6	47	9	D4	N/A	N/A	N/A	N/A	N/A
7	47	8	D4	D4	N/A	N/A	N/A	N/A
8	47	10	D1+D3	D1	N/A	N/A	N/A	N/A
9	47	13	D1	D1	N/A	D1	D1	N/A
10	47	12	D2	D2	D2	N/A	N/A	N/A
11	47	8	N/A	N/A	D2	N/A	N/A	N/A
12	47	8	D1+D4	D4	N/A	N/A	N/A	N/A
13	47	6	D1	D1	N/A	N/A	D1	N/A
14	47	6	D1	D1	D1+D2+D4	D1	D1	N/A
15	47	10	D4	D4	N/A	N/A	N/A	N/A
16	47	14	D2	N/A	N/A	N/A	N/A	N/A
17	47	8	D4	D4	N/A	N/A	N/A	N/A
18	47	8	D3	D3	N/A	N/A	N/A	N/A
19	47	7	D2/D1	D1	N/A	N/A	N/A	N/A
20	47	25	D1	D1	N/A	N/A	N/A	N/A
21	47	7	D3	N/A	N/A	N/A	N/A	N/A
22	47	6	D4	D4	N/A	N/A	N/A	N/A
23	47	8	D4	D4	N/A	N/A	N/A	N/A
24	47	25	D1	N/A	N/A	N/A	N/A	N/A
25	47	5	D2+D4	D2	N/A	N/A	N/A	N/A
26	47	12	D4	N/A	N/A	N/A	N/A	N/A
27	47	8	D4	D4	N/A	N/A	N/A	N/A
28	47	12	D4	D4	N/A	N/A	N/A	N/A

No	Year	Age	Multiplex RT-PCR					
			PBMC	Plasma	Pellet	Saliva	Oral brush	Serum
29	47	11	D4	N/A	N/A	N/A	N/A	N/A
30	47	7	D1	D1	D4	N/A	D1+D4	N/A
31	47	9	D2+D4	D2+D4	N/A	N/A	N/A	N/A
32	47	15	D3	D3	N/A	N/A	N/A	N/A
33	47	9	D2	D2	N/A	N/A	D1+D4	N/A
34	47	11	D1	D1	N/A	D1	D1+D4	N/A
35	47	13	D2	D1+D2	N/A	N/A	N/A	N/A
36	47	13	D3	D3	N/A	N/A	N/A	N/A
37	47	10	D4	D4	N/A	N/A	N/A	N/A
38	47	10	D4	D4	N/A	N/A	N/A	N/A
39	47	12	D4	D4	N/A	N/A	N/A	N/A
40	47	6	D4	D4	N/A	N/A	N/A	N/A
41	47	12	D4	D4	N/A	N/A	N/A	N/A
42	47	9	N/A	N/A	N/A	N/A	N/A	N/A
43	47	10	D4	N/A	N/A	N/A	N/A	N/A
44	48	7	D4	N/A	N/A	N/A	N/A	N/A
45	48	8	D1	D1	N/A	N/A	N/A	N/A
46	48	8	D3	D3	N/A	N/A	N/A	N/A
47	48	13	N/A	N/A	N/A	N/A	N/A	N/A
48	48	6	D4	D4	N/A	N/A	N/A	N/A
49	47	25	D4	N/A	N/A	N/A	N/A	N/A
50	47	20	N/A	N/A	N/A	N/A	N/A	N/A
51	47	23	N/A	D1	N/A	N/A	N/A	N/A
52	47	30	D2	D2	D2	N/A	N/A	N/A
53	47	23	D2	D2	D2	N/A	N/A	N/A
54	47	35	D4	D4	D4	N/A	N/A	N/A
55	48	27	N/A	N/A	D1+D2+D4	N/A	N/A	D4
56	48	26	D1+D4	D1+D4	D1	D1	N/A	N/A
57	48	23	D1	N/A	N/A	N/A	N/A	N/A
58	48	32	D4	D4	N/A	N/A	N/A	N/A
59	48	37	D4	N/A	N/A	N/A	N/A	N/A

No	Year	Age	Multiplex RT-PCR					
			PBMC	Plasma	Pellet	Saliva	Oral brush	Serum
60	48	28	D4+D2	D4	N/A	N/A	N/A	N/A
61	48	34	D4+D2	D4	N/A	N/A	N/A	N/A
62	48	32	D4	D4	N/A	N/A	N/A	N/A
63	48	40	D4	D4	N/A	N/A	N/A	N/A
64	48	38	D3	D3	N/A	N/A	N/A	N/A
65	48	37	D4	D4	N/A	N/A	N/A	N/A
66	48	40	N/A	N/A	N/A	N/A	N/A	N/A
67	48	42	D3	D3	N/A	N/A	N/A	N/A
68	48	8	D3	D3	N/A	N/A	N/A	N/A
69	48	29	N/A	N/A	N/A	N/A	N/A	N/A
70	48	34	D4	D4	N/A	N/A	N/A	N/A
71	48	38	N/A	N/A	N/A	N/A	N/A	N/A
72	48	21	D4+D2	D4	N/A	N/A	N/A	N/A
73	48	37	D1	D1	N/A	N/A	D1	N/A
74	48	39	N/A	N/A	N/A	N/A	D3	N/A
75	48	28	D3	D3	N/A	N/A	N/A	N/A
76	48	40	D4	N/A	N/A	N/A	N/A	N/A
77	48	23	N/A	N/A	N/A	N/A	N/A	N/A
78	48	42	N/A	N/A	N/A	N/A	N/A	N/A
79	48	28	N/A	N/A	N/A	N/A	N/A	N/A
80	49	31	D1+D3+D4	D1+D3+D4	N/A	D4	D4	D1+D3+D4
81	49	20	D2	D2	N/A	D2+D3	D2	N/A
82	49	36	N/A	N/A	N/A	D2+D3	N/A	D2+D3
83	49	18	N/A	D2+D3	N/A	N/A	D2	D2
84	50	24	D2	D2	D2	N/A	D2	D2
85	50	13	D1	D1	D1	D1	N/A	N/A
86	50	9	D1	D1	N/A	N/A	N/A	N/A
87	50	14	D3	D3	N/A	N/A	N/A	N/A
88	50	13	D1	D1	N/A	D1	N/A	N/A

No	Year	Age	Multiplex RT-PCR					
			PBMC	Plasma	Pellet	Saliva	Oral brush	Serum
89	50	44	N/A	N/A	N/A	N/A	N/A	N/A
90	46	37	N/A	N/A	N/A	N/A	N/A	N/A
91	46	25	N/A	N/A	D4	N/A	N/A	N/A
92	46	6	D1	D1	D1	D1	N/A	N/A
93	47	9	N/A	N/A	D1	N/A	N/A	N/A
94	47	13	N/A	D1	D1+D3	N/A	N/A	N/A
95	47	9	N/A	N/A	D1	N/A	N/A	N/A

no	Nested RT-PCR						ELISA		CR
	PBMC	Plasma	Pellet	Saliva	Oral brush	Serum	IgM	IgG	
1	+ve	+ve	-ve	-ve	-ve	N/A	24	175	DF (secondary)
2	+ve	+ve	-ve	+ve	-ve	N/A	36	138	DF (secondary)
3	+ve	+ve	-ve	-ve	-ve	N/A	39	89	DHF (secondary)
4	+ve	+ve	-ve	+ve	-ve	+ve	25	110	DHFII (secondary)
5	+ve	+ve	+ve	-ve	-ve	-ve	93	20	DF (primary)
6	+ve	+ve	-ve	-ve	-ve	N/A	52	205	DF (secondary)
7	+ve	+ve	+ve	+ve	+ve	N/A	13	219	DHF III (secondary)
8	+ve	-ve	-ve	+ve	-ve	N/A	6	110	DF (secondary)
9	+ve	+ve	+ve	+ve	+ve	N/A	12	125	DHFII (secondary)
10	+ve	+ve	+ve	+ve	-ve	N/A	37	141	DSS (secondary)
11	N/A	N/A	+ve	+ve	N/A	N/A	33	226	DHFII (secondary)
12	+ve	+ve	N/A	N/A	N/A	N/A	14	101	secondary
13	+ve	+ve	-ve	+ve	-ve	N/A	31	215	DHFII (secondary)
14	+ve	+ve	-ve	+ve	+ve	N/A	119	18	DF (primary)
15	+ve	+ve	N/A	N/A	N/A	N/A	42	129	DHFIII (secondary)
16	+ve	-ve	N/A	N/A	N/A	N/A	39	132	DHFII (secondary)
17	+ve	+ve	N/A	N/A	N/A	N/A	39	106	DF (primary)
18	+ve	+ve	-ve	-ve	-ve	N/A	20	156	DF (secondary)
19	+ve	+ve	N/A	N/A	N/A	N/A	25	216	DF (secondary)
20	+ve	+ve	N/A	N/A	N/A	N/A	138	134	secondary
21	+ve	-ve	-ve	-ve	-ve	-ve	43	223	DHFII (secondary)
22	+ve	+ve	N/A	N/A	N/A	N/A	44	158	DHFII (secondary)
23	+ve	+ve	-ve	-ve	-ve	N/A	18	146	DHFII (secondary)
24	+ve	-ve	N/A	N/A	N/A	N/A	44	106	secondary
25	+ve	+ve	N/A	N/A	N/A	N/A	61	176	DHF (secondary)
26	+ve	-ve	N/A	N/A	N/A	N/A	55	177	DHFII (secondary)
27	+ve	+ve	N/A	N/A	N/A	N/A	79	185	DF (secondary)
28	+ve	+ve	N/A	N/A	N/A	N/A	61	213	DHFII (secondary)
29	+ve	-ve	N/A	N/A	N/A	N/A	26	246	DHF III (secondary)
30	+ve	+ve	+ve	-ve	+ve	N/A	91	3	DF (primary)

no	Nested RT-PCR						ELISA		CR
	PBMC	Plasma	Pellet	Saliva	Oral brush	Serum	IgM	IgG	
31	+ve	-ve	N/A	N/A	N/A	N/A	53	194	DF (primary)
32	+ve	+ve	N/A	N/A	N/A	N/A	32	157	DFI I(secondary)
33	+ve	+ve	N/A	N/A	N/A	N/A	12	190	DHFII (secondary)
34	+ve	+ve	N/A	-ve	-ve	N/A	39	106	DHFI (primary)
35	+ve	+ve	N/A	N/A	N/A	N/A	41	242	DHFII (secondary)
36	+ve	+ve	N/A	N/A	N/A	N/A	69	241	DHFII (secondary)
37	+ve	+ve	-ve	+ve	+ve	N/A	51	14	DF (primary)
38	+ve	+ve	-ve	-ve	-ve	-ve	37	106	DF (secondary)
39	+ve	+ve	N/A	+ve	-ve	N/A	35	123	DHFI (secondary)
40	+ve	+ve	N/A	N/A	N/A	N/A	42	112	DF (secondary)
41	+ve	+ve	N/A	N/A	N/A	N/A	20	168	DF (secondary)
42	N/A	N/A	+ve	+ve	+ve	N/A	78	0	primary
43	+ve	-ve	-ve	-ve	-ve	N/A	65	125	DF (secondary)
44	+ve	-ve	-ve	-ve	-ve	N/A	35	101	DHFII (secondary)
45	+ve	+ve	-ve	-ve	-ve	N/A	20	21	DHFII (secondary)
46	+ve	+ve	-ve	+ve	-ve	N/A	41	129	secondary
47	+ve	+ve	+ve	-ve	-ve	N/A	78	137	secondary
48	+ve	+ve	N/A	+ve	+ve	N/A	25	137	secondary
49	+ve	-ve	-ve	-ve	+ve	N/A	44	161	DHFII I(secondary)
50	-ve	-ve	+ve	-ve	-ve	N/A	35	237	DHFII (secondary)
51	-ve	+ve	+ve	-ve	-ve	N/A	13	125	DHFI (secondary)
52	+ve	+ve	+ve	-ve	+ve	N/A	58	224	DHFIII (secondary)
53	+ve	+ve	+ve	N/A	N/A	N/A	36	158	DHFII (secondary)
54	+ve	+ve	+ve	-ve	+ve	N/A	21	145	DHFII (secondary)
55	N/A	N/A	+ve	+ve	+ve	+ve	54	105	DHFIII (secondary)
56	+ve	+ve	-ve	-ve	N/A	N/A	33	144	DHFIII (secondary)
57	+ve	-ve	-ve	-ve	-ve	N/A	87	113	DHF I(secondary)
58	+ve	+ve	-ve	+ve	-ve	N/A	82	135	DHF I(secondary)
59	+ve	-ve	-ve	-ve	-ve	N/A	84	138	DHF III (secondary)
60	+ve	-ve	-ve	-ve	-ve	N/A	72	135	DHFII (secondary)

no	Nested RT-PCR						ELISA		CR
	PBMC	Plasma	Pellet	Saliva	Oral brush	Serum	Ig M	IgG	
61	+ve	+ve	-ve	-ve	-ve	N/A	62	125	DHF II (secondary)
62	+ve	+ve	-ve	+ve	+ve	N/A	74	148	DHFII (secondary)
63	-ve	+ve	+ve	+ve	+ve	N/A	81	150	DHF II(secondary)
64	+ve	+ve	-ve	-ve	-ve	N/A	90	155	DF (secondary)
65	+ve	+ve	-ve	+ve	-ve	N/A	41	137	DHF II (secondary)
66	+ve	-ve	-ve	-ve	-ve	N/A	10	148	secondary
67	+ve	+ve	+ve	-ve	+ve	N/A	1	133	DHF I (secondary)
68	+ve	-ve	-ve	+ve	+ve	N/A	26	137	DHF III
69	+ve	-ve	-ve	+ve	-ve	N/A	49	-	(secondary)
70	-ve	-ve	-ve	+ve	-ve	N/A	-	153	DHF I (secondary)
71	+ve	-ve	-ve	-ve	-ve	N/A	35	126	DF (secondary
72	-ve	-ve	-ve	+ve	+ve	N/A	64	155	DHF II (secondary)
73	+ve	+ve	+ve	+ve	+ve	N/A	29	175	DHFII (secondary)
74	-ve	-ve	-ve	+ve	+ve	N/A	37	128	DHF II (secondary)
75	+ve	+ve	-ve	+ve	+ve	N/A	33	149	DHF II (secondary
76	+ve	+ve	-ve	-ve	-ve	N/A	34	140	DHF I (secondary)
77	-ve	-ve	+ve	N/A	N/A	N/A	53	158	DHF II (secondary
78	-ve	-ve	-ve	+ve	-ve	N/A	51	29	DHFII (secondary)
79	+ve	-ve	N/A	-ve	N/A	N/A	10	154	DHF I (primary)
80	+ve	+ve	-ve	-ve	+ve	+ve	9	143	DHFIII (secondary)
81	+ve	+ve	-ve	+ve	+ve	+ve	30	134	DHF I (secondary)
82	+ve	+ve	+ve	+ve	-ve	+ve	40	144	DHF I (secondary)
83	+ve	+ve	+ve	-ve	+ve	+ve	34	119	DHF I (secondary)
84	+ve	+ve	+ve	+ve	+ve	+ve	19	142	DHF I(secondary)
85	+ve	+ve	+ve	+ve	+ve	N/A	48	8	DHF I (secondary)
86	+ve	+ve	-ve	-ve	-ve	N/A	22	120	DHF I (primary)
87	+ve	+ve	-ve	+ve	-ve	N/A	87	116	DF (secondary)
88	+ve	+ve	N/A	+ve	-ve	N/A	76	136	DF (secondary DF (secondary)

no	Nested RT-PCR						ELISA		CR
	PBMC	Plasma	Pellet	Saliva	Oral brush	Serum	IgM	IgG	
89	+ve	+ve	+ve	N/A	N/A	+ve	25	153	DHFII (secondary)
90	+ve	+ve	+ve	N/A	N/A	N/A	77	120	DHF II (secondary)
91	+ve	+ve	+ve	N/A	N/A	N/A	57	170	DHF II (secondary)
92	+ve	+ve	+ve	+ve	-ve	N/A	48	123	DHF (secondary)
93	+ve	+ve	+ve	+ve	-ve	N/A	29	215	DSS (secondary)
94	+ve	+ve	+ve	-ve	-ve	N/A	39	221	DHF (secondary)
95	+ve	N/A	+ve	+ve	N/A	N/A	65	145	secondary

Years	Patients	Not detected	DENV-1	DENV-2	DENV-3	DENV-4	Multiple serotypes
2546	pediatric	0	1/1 (100%)	-	-	-	-
	Adult	1	-	-	-	1/1 (100%)	-
(%)			100%			50%	
2547	pediatric	2	5/37 (13.51%)	3/37 (8.11%)	4/37 (10.81%)	14/37 (37.84%)	11/37 (29.73%)
	Adult	1	4/7 (57.14%)	2/7 (28.57%)	-	1/7 (14.29%)	-
(%)			9/44 (20.45%)	5/44 (11.36%)	4/37 (10.81%)	15/44 (34%)	11/37 (29.72%)
2548	pediatric	0	1/4 (25%)	-	1/4 (25%)	2/4 (50%)	-
	Adult	5	2/21 (9.52%)	-	5/21 (23.81%)	9/21 (42.86%)	5/21 (23.81%)
(%)			3/25 (12%)	-	6/25 (24%)	10/25 (40%)	5/21 (24%)
2549	pediatric	0	-	-	-	-	-
	Adult	0	-	-	-	-	4/4 (100%)
(%)							4/4 (100%)
2550	pediatric	0	3/4 (75%)	-	1/4 (25%)	-	-
	Adult	1	-	1/1(100%)	-	-	-
(%)			3/5 (60%)	1/5 (20%)	1/5 (20%)		
Total	92	10	16/85 (18.82%)	6/85 (7.06%)	11/85 (12.94%)	32/85 (37.65%)	20/85 (23.53%)

Year	Patient	Multiple DENV serotypes infected						
		D1+D2	D1+D3	D1+D4	D2+D3	D2+D4	D1+D2+D4	D1+D3+D4
2546	Children							
	Adult							
2547	Children	2 /11 (18.18%)	2/11 (18.18%)	4/11 (36.36%)		2 /11 (18.18%)	1/11 (9.1%)	
	Adult							
2548	Children							
	Adult			1/5 (20%)		3/5 (60%)	1/5 (20%)	
2549	Children							
	Adult				3/4 (75%)			1/4 (25%)
2550	Children							
	Adult							
Total		2 /20 (10%)	2/20 (10%)	5/20 (25%)	3/20 (15%)	5/20 (25%)	2/20 (10%)	1/20 (5%)

Patient	DEN-1		DEN-2		DEN-3		DEN-4		Multi-serotypes		No Clinical data	Total
	DF	DHF	D F	DHF	DF	DHF	DF	DHF	DF	DHF		
children	5	7	0	3	3	2	7	9	5	5	4	47
Adult	0	3	0	3	1	4	1	9	0	10	3	31
Total	5	10	0	6	4	6	8	18	5	16	7	78
	6.33 %	12.65 %	0	7.6 %	5.06 %	7.6 %	10.12 %	21.52 %	7.6 %	21.52 %		

patient	Multiplex RT-PCR					No detected	Total
	DEN-1	DEN-2	DEN-3	DEN-4	Multi-serotypes		
Children	10 (19.23%)	3 (5.77%)	6 (11.53%)	17 (32.7%)	11(21.15%)	2	52
Adult	6(18.18%)	3(9.1%)	5(15.15%)	15(45.45%)	9(27.27%)	8	33
Total	16(18.82%)	6(7.06%)	11(12.94%)	32 (37.65%)	20(23.53%)	10	85



APPENDIX B

Buffer and Reagent Preparation

1. 10 TBE buffer

Tris base	54	g
Boric acid	27.5	g
0.5 M EDTA (pH8)	20	ml

Adjust pH to pH 8. Add ddH₂O to 1 liter and sterilized by autoclaving

2. PBS buffer

NaCl	8	g
KCl	0.2	g
Na ₂ HPO ₄	1.44	g
KH ₂ PO ₄	0.24	g

Adjust pH 7.4 and add DEPC incubate at 37°C Overnight and sterilize by autoclaving

3. Oral buffer

0.1 M EDTA	200	ml
2 M Tris	5	ml
5 M NaCl	4	ml
DEPC	791	ml

Sterilize by autoclaving

BIOGRAPHY



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Original articles

Wanla Kulwichit*1, Sumanee Nilgate2, Tanittha Chatsuwan2, Sunisa Krajiw1 Accuracies of *Leuconostoc* phenotypic identification: a comparison of API systems and conventional phenotypic assays. *BMC* 7: 69 (2007).