



Supplementary Materials for First Evidence of Co-Circulation of Emerging *Leishmania martiniquensis*, *Leishmania orientalis*, and *Crithidia* sp. in *Culicoides* Biting Midges (Diptera: Ceratopogonidae), the Putative Vectors for Autochthonous Transmission in Southern Thailand

Table S1. The BLASTn result of COI sequences obtained from representative samples of *Culicoides* Species in this study.

No.	Sample ID	Accession No.	Morphological Identification	Top BLASTn Hits and % Similarity
1.	CNR10	OP741195	<i>C. huffi</i>	<i>C. huffi</i> MLP218-11 (MZ191866), 96.20%
2.	CSP18	OP741196	<i>C. peregrinus</i>	<i>C. peregrinus</i> YYU_103 (KY433459), 99.52%
3.	CSP20	OP741197	<i>C. peregrinus</i>	<i>C. peregrinus</i> YYU_103 (KY433459), 100%
4.	CSP28	OP741198	<i>C. peregrinus</i>	<i>C. peregrinus</i> YYU_103 (KY433459), 100%
5.	CSP35	OP741199	<i>C. oxystoma</i>	<i>C. oxystoma</i> 87m045A211 (MW496264), 99.28%
6.	CSP38	OP741200	<i>C. oxystoma</i>	<i>C. oxystoma</i> O7 (MW496278), 99.52%
7.	CSP50	OP741201	<i>C. oxystoma</i>	<i>C. oxystoma</i> O8 (MW496279), 100%
8.	CSP51	OP741202	<i>C. oxystoma</i>	<i>C. oxystoma</i> 13m042A26 (MW496246), 99.05%
9.	CSP52	OP741203	<i>C. oxystoma</i>	<i>C. oxystoma</i> 12m016A22 (MW496245), 99.29%
10.	CNR3	OP741204	<i>C. mahasarakhamense</i>	<i>C. mahasarakhamense</i> MLP60-29 (MZ191855), 100%
11.	CNR4	OP741205	<i>C. mahasarakhamense</i>	<i>C. mahasarakhamense</i> MLP60-29 (MZ191855), 100%
12.	CSP61	OP741206	<i>C. mahasarakhamense</i>	<i>C. mahasarakhamense</i> MLP60-29 (MZ191855), 99.76%
13.	CSP62	OP741207	<i>C. mahasarakhamense</i>	<i>C. mahasarakhamense</i> MLP60-29 (MZ191855), 99.76%
14.	CHH95	OP741208	<i>C. orientalis</i>	<i>C. asiana</i> VNMC23 (MK760107), 99.76%
15.	CBF13	OP741209	<i>C. innoxius</i>	<i>C. innoxius</i> G6 (MZ189956), 99.76%
16.	CBF18	OP741210	<i>C. innoxius</i>	<i>C. innoxius</i> G6 (MZ189956), 99.76%
17.	CHH7	OP741211	<i>C. innoxius</i>	<i>C. innoxius</i> G6 (MZ189956), 99.27%
18.	CHH26	OP741212	<i>C. innoxius</i>	<i>C. innoxius</i> MLP251-31 (MZ191870), 100%
19.	CBF3	OP741213	<i>C. flaviscutatus</i>	<i>C. sinanoensis</i> RUXX03 (MK760243), 87%
20.	CBF5	OP741214	<i>C. flaviscutatus</i>	<i>C. sinanoensis</i> RUXX03 (MK760243), 87%
21.	CHH105	OP741215	<i>C. jacobsoni</i>	<i>C. jacobsoni</i> G9 (MK189959), 100%
22.	CHH45	OP741216	<i>C. fulvus</i>	<i>C. fulvus</i> ww08225 (KT352643), 100%
23.	CBF17	OP741217	<i>C. fulvus</i>	<i>C. fulvus</i> ww08225 (KT352643), 100%
24.	CBF28	OP741218	<i>C. Trithecoides</i> sp.	<i>Culicoides</i> sp. 3 YF-2021a G36 (MZ189973), 100%
25.	CBF42	OP741219	<i>C. Trithecoides</i> sp.	<i>C. tropicalis</i> CIRAD:ZA.21 (MF399798), 85.20%

Table S2. The BLASTn result of *Leishmania* ITS1 sequences amplified from field-caught *Culicoides* biting midges in this study.

No.	Location	Accession no.	Parasite Identification	Isolate	Host Species	Top BLASTn Matches and % Similarity
1.	Sadao/1 st patient house	OP698051	<i>L. martiniquensis</i>	CNR10	<i>C. huffi</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 98.84%
2.	Sadao/2 nd patient house	OP698052	<i>L. martiniquensis</i>	CSP2	<i>C. peregrinus</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 100%
3.	Sadao/2 nd patient house	OP698053	<i>L. martiniquensis</i>	CSP8	<i>C. peregrinus</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 100%

4.	Sadao/2 nd patient house	OP698054	<i>L. martiniquensis</i>	CSP9	<i>C. peregrinus</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 98.84%
5.	Sadao/2 nd patient house	OP698055	<i>L. martiniquensis</i>	CSP11	<i>C. peregrinus</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 99.61%
6.	Sadao/2 nd patient house	OP698056	<i>L. orientalis</i>	CSP12	<i>C. peregrinus</i>	<i>L. orientalis</i> 378_Trang (KY982674), 100%
7.	Sadao/2 nd patient house	OP698057	<i>L. martiniquensis</i>	CSP15	<i>C. peregrinus</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 99.61%
8.	Sadao/2 nd patient house	OP698058	<i>L. martiniquensis</i>	CSP35	<i>C. oxystoma</i>	<i>L. martiniquensis</i> SK4-1 (MK603826), 99.61%
9.	Sadao/2 nd patient house	OP698059	<i>L. orientalis</i>	CSP38	<i>C. oxystoma</i>	<i>L. orientalis</i> 378_Trang (KY982674), 98%
10.	Sadao/2 nd patient house	OP698060	<i>L. martiniquensis</i>	CSP41	<i>C. oxystoma</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 98.84%
11.	Sadao/2 nd patient house	OP698061	<i>L. martiniquensis</i>	CSP42	<i>C. oxystoma</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 99.61%
12.	Sadao/2 nd patient house	OP698062	<i>L. martiniquensis</i>	CSP44	<i>C. oxystoma</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 99.61%
13.	Sadao/2 nd patient house	OP698063	<i>L. martiniquensis</i>	CSP45	<i>C. oxystoma</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 99.61%
14.	Sadao/2 nd patient house	OP698064	<i>L. martiniquensis</i>	CSP48	<i>C. oxystoma</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 99.61%
15.	Sadao/2 nd patient house	OP698065	<i>L. martiniquensis</i>	CSP59	<i>C. mahasarakhamense</i>	<i>L. martiniquensis</i> SK4-1 (MK603826), 100%
16.	Rattaphum/livestock sheds	OP698066	<i>L. martiniquensis</i>	CHH10	<i>C. fordai</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 100%
17.	Rattaphum/livestock sheds	OP698067	<i>L. martiniquensis</i>	CHH45	<i>C. fulvus</i>	<i>L. martiniquensis</i> 770605_Trang (KY982650), 100%

Table S3. The BLASTn result of *Crithidia* SSU rRNA sequences derived from field-caught. Culicoides biting midges in this study.

No.	Location	Accession no.	Parasite Identification	Isolate	Host species	Top BLASTn Matches and % Similarity
1.	Sadao/2 nd patient house	OP698037	<i>Crithidia</i> sp.	CSP18	<i>C. peregrinus</i>	<i>C. thermophila/confusa/deanei</i> , 97.21%
2.	Sadao/2 nd patient house	OP698038	<i>Crithidia</i> sp.	CSP20	<i>C. peregrinus</i>	<i>C. thermophila/confusa/deanei</i> , 97.42%
3.	Sadao/2 nd patient house	OP698039	<i>Crithidia</i> sp.	CSP22	<i>C. peregrinus</i>	<i>C. thermophila/confusa/deanei</i> , 96.78%
4.	Sadao/2 nd patient house	OP698040	<i>Crithidia</i> sp.	CSP25	<i>C. peregrinus</i>	<i>C. thermophila/confusa/deanei</i> , 96.99%
5.	Sadao/2 nd patient house	OP698041	<i>Crithidia</i> sp.	CSP28	<i>C. peregrinus</i>	<i>C. thermophila/confusa/deanei</i> , 97.31%
6.	Sadao/2 nd patient house	OP698042	<i>Crithidia</i> sp.	CSP30	<i>C. peregrinus</i>	<i>C. thermophila/confusa/deanei</i> , 96.46%
7.	Sadao/2 nd patient house	OP698043	<i>Crithidia</i> sp.	CSP42	<i>C. oxystoma</i>	<i>C. thermophila/confusa/deanei</i> , 96.99%

8.	Sadao/2 nd patient house	OP698044	<i>Crithidia</i> sp.	CSP50	<i>C. oxystoma</i>	<i>C. thermophila/confusa/deanei</i> , 96.24%
9.	Rattaphum/livestock sheds	OP698045	<i>Crithidia</i> sp.	CHH64	<i>C. fordae</i>	<i>C. thermophila/confusa/deanei</i> , 97.31%
10.	Rattaphum/livestock sheds	OP698046	<i>Crithidia</i> sp.	CHH65	<i>C. fordae</i>	<i>C. thermophila/confusa/deanei</i> , 96.89%
11.	Rattaphum/livestock sheds	OP698047	<i>Crithidia</i> sp.	CHH95	<i>C. orientalis</i>	<i>C. thermophila/confusa/deanei</i> , 96.89%
12.	Rattaphum/livestock sheds	OP698048	<i>Crithidia</i> sp.	CHH113	<i>C. elbeli</i>	<i>C. thermophila/confusa/deanei</i> , 97.42%

Accession nos of *C. thermophila/confusa/deanei* references were KY264937, JF717837, and EU079129, respectively.