

Table S1. PCR primers and probes used in this study.

| Target organism | Target gene | Primer name | Primer sequence (5'-3') | Product size | Reference ¹ |
|---|--|--------------------------------------|--|--------------|------------------------|
| <i>Primers for amplifying, cloning and sequencing</i> | | | | | |
| Bacteria | 16S rRNA | 07F 1507R | AGAGTTTGATCMTGGCTCAG TACCTTGTTACGACTTCAC | 1.5 kb | [1] |
| <i>Burkholderia</i> strains | <i>gyrB</i> | <i>gyrB</i> 135F <i>gyrB</i> 850R | ACCGGTCTGCAYCACCTCGT TCGTTGTAGCTGTCGTTCCACTGC | 0.65 kb | modified from [2] |
| Insect (Stenocephalidae) | <i>cox1</i> | COI_long22 Pat_R | ACWAAYCATAAAGATATTGGMAC TCCAATGCACTAATCTGCCATATTA | 1.55 kb | [3] [4] |
| " | <i>nad1</i> (+ 2nd half of <i>cob</i>) | steno_ND1_F1 steno_ND1_R1 | GTAAAATGATTATGAGGRGGATT CTTAGGGATAACAGCGTAATTTTYYGG | 2.0 kb | [3] |
| " | <i>cob</i> (1st half) | steno_cytB_F1 steno_cytB_R3 | ATTTATATAGCAAGAGTAGCATCAAA TTCTGGTTGAATATGAACTGGTGT | 1.1 kb | [3] |
| " | <i>nad6</i> | steno_ND4_F2 steno_ND6_R1 | CCTAAACCCAAAGCACCTTCACA AATCCYCCTCATAATCATTTTAC | 1.35 kb | [3] |
| Target organism | Target gene | probe name | Probe sequence (5'-3') | Labeling dye | Reference |
| <i>Probes for fluorescence in situ hybridization</i> | | | | | |
| Bacteria | 16S rRNA | EUB338 | GCTGCCTCCCGTAGGAGT | Cy3 | [5] |
| EUB-Planctomycetes | 16S rRNA | EUB338 II | GCAGCCACCCGTAGGTGT | Cy3 | [5] |
| EUB-Verrucomicrobia | 16S rRNA | EUB338 III | GCAGCCACCCGTAGGTGT | Cy3 | [5] |
| - | 16S rRNA | NON338 | ACTCCTACGGGAGGCAGC | Cy3 | [6] |
| <i>Burkholderia</i> -symbiont of Stenocephalidae | 16S rRNA | StenoSymb440 | ACTAATACTGTGGGGGGA | Cy3 | [3] |

¹[1] Lane, D. J. 1991. In E. Stackenbrandt and M. Goodfellow (ed.), John Wiley and Sons, New York, NY. [2] 1. Spilker et al., 2009. J. Clin. Microbiol. 47: 2607–2610. [3]

This study. [4] Simon et al., 1994. Ann. Entomol. Soc. Am. 87: 651–702. [5] Daims et al., 1999. Syst. Appl. Microbiol. 22: 434–444. [6] Manz et al., 1992. Syst. Appl. Microbiol. 15: 593–600.

Table S2. The number of *gyrB* gene clones in each OTU subgroups of the *Burkholderia* symbionts.

| Species | Insect ID | Stenocephalidae-clade | | | | | SBE clade | | | | | PBE clade | Total | |
|----------------------------------|-----------|-----------------------|------|------|------|------|-----------|------|------|------|-------|-----------|-------|-----------|
| | | OTU1 | OTU2 | OTU3 | OTU4 | OTU5 | OTU6 | OTU7 | OTU8 | OTU9 | OTU10 | OTU11 | | |
| <i>Dicranocephalus agilis</i> | | | | | | | | | | | | | | |
| | Dag1 | 2 | - | - | - | - | - | - | - | - | - | - | - | 2 |
| | Dag2 | - | - | 3 | 1 | 4 | - | - | - | - | - | - | - | 8 |
| <i>Dicranocephalus medius</i> | | | | | | | | | | | | | | |
| | Dme1 | 1 | - | - | - | - | - | - | - | - | - | - | - | 1 |
| | Dme2 | 8 | - | - | - | - | - | - | - | - | - | - | - | 8 |
| | Dme3 | 2 | 5 | - | - | - | - | - | - | - | - | - | - | 7 |
| | Dme4 | - | 3 | - | - | - | - | - | - | - | - | - | - | 3 |
| | Dme5 | - | 2 | - | - | - | - | - | - | - | - | - | - | 2 |
| | Dme6 | 12 | - | - | - | - | - | - | - | - | - | - | - | 12 |
| <i>Dicranocephalus lateralis</i> | | | | | | | | | | | | | | |
| | Dla1 | - | - | - | - | - | 9 | 1 | 1 | - | - | - | - | 11 |
| | Dla2 | - | - | - | - | - | - | - | 3 | - | - | - | - | 3 |
| | Dla3 | - | - | - | - | - | - | - | 7 | 1 | - | - | - | 8 |
| | Dla4 | - | - | - | - | - | - | - | - | - | 10 | - | - | 10 |
| <i>Dicranocephalus albipes</i> | | | | | | | | | | | | | | |
| | Dal1 | - | - | - | - | - | - | - | - | - | - | 13 | - | 13 |
| | Dal2 | - | 3 | - | - | - | - | - | - | - | - | - | - | 3 |
| | OTU total | 25 | 13 | 3 | 1 | 4 | 9 | 1 | 11 | 1 | 10 | 13 | - | 91 |

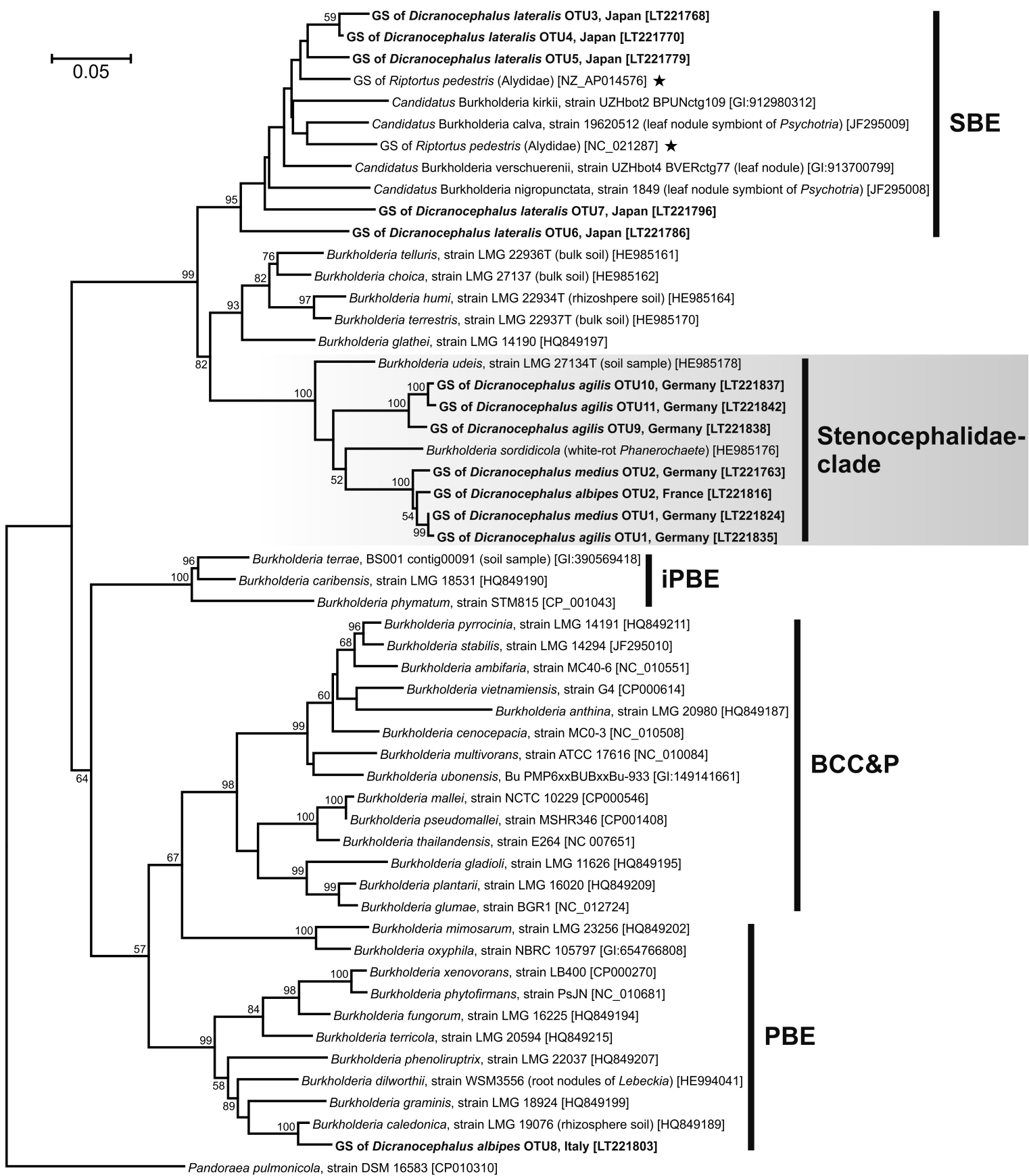


Fig. S1. Molecular phylogeny of the gut symbiotic bacteria of stenocephalid species based on the *gyrB* sequences. The tree displays a maximum likelihood (ML) phylogeny of eleven OTUs of the gut symbiotic bacteria (GS) identified from *Dicranocephalus albipes*, *D. agilis*, *D. lateralis* and *D. medius* together with selected representatives of the different *Burkholderia* groups. The alignment of 662 nucleotide sites of the *Burkholderia gyrB* gene was used. The gut symbionts of the stenocephalid species are shown in bold. The origins or sources of isolation of the *Burkholderia* strains/sequences are represented in parentheses. Accession numbers in the DNA database (DDBJ/EMBL/GenBank) are shown in square brackets. *Burkholderia* gut symbionts of other stinkbug families are marked with an asterisk. The major *Burkholderia* clades (BCC&P, SBE and PBE) including the subclade “insect-associated PBE (iPBE)” and the “Stenocephalidae-clade” here described are indicated on the right. Bayesian (MrBayes) and neighbor-joining (NJ) analyses gave essentially the same results (data not shown). Bootstrap values higher than 50% are depicted at the nodes.