

ASSESSING THE LIKELIHOOD OF MEROPENEM RESISTANCE ACQUISITION IN
BURKHOLDERIA PSEUDOMALLEI VIA LATERAL GENE TRANSFER FROM
BURKHOLDERIA UBONENSIS

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ABSTRACT

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The disease melioidosis is caused by the environmental bacterium *Burkholderia pseudomallei*. Treatment for this disease can be complex, as *B. pseudomallei* is intrinsically resistant to multiple broad-spectrum antimicrobials. However, decades of research have resulted in a more effective treatment regimen, with meropenem reserved for patients in Intensive Care Units suffering from severe melioidosis. *Burkholderia ubonensis*, a distantly related non-pathogenic bacterium, is often co-isolated with *B. pseudomallei* from soil. It has been suggested that ~50% of *B. pseudomallei* strains are naturally competent and both species can acquire new genomic content via lateral gene transfer (LGT) and can also display resistance to a variety of antimicrobials, with *B. ubonensis* occasionally displaying high levels of intrinsic resistance to meropenem. The goal of this study was to determine if *B. ubonensis* can serve as a source for meropenem resistance in *B. pseudomallei* via LGT.

Meropenem resistance was evaluated in a set of 122 phylogenetically and spatially diverse environmental isolates of *B. pseudomallei* using a broth microdilution method. All strains had an MIC of ≤ 2 $\mu\text{g/ml}$ and were determined to be susceptible. A series of transformation experiments was conducted using an attenuated strain of *B. pseudomallei* and a genetically modified strain of *B. ubonensis*. No growth was observed when *B. pseudomallei* was exposed to *B. ubonensis* DNA for a short period of time and subsequently plated onto agar plates containing

subinhibitory levels of meropenem (20 µg/ml). In addition, clinically significant increases in meropenem resistance were not observed in generated *B. pseudomallei* mutants containing the specific genes that confer meropenem resistance in *B. ubonensis*.

Environmental isolates of *B. pseudomallei* are largely susceptible to meropenem and cannot acquire and express meropenem resistance genes from environmental *B. ubonensis* DNA.

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CHAPTER 1: INTRODUCTION

Burkholderia pseudomallei, a member of the *Burkholderia pseudomallei* complex (Bpc) within the genus *Burkholderia*, is a Gram-negative soil-dwelling organism that causes the disease melioidosis. Melioidosis largely affects immunocompromised individuals [1-3]; with patients often suffering from various co-morbidities, such as alcoholism or diabetes mellitus [4-7].

Disease complications can result from delayed diagnosis due to numerous diverse symptoms that can also be attributed to various other diseases, such as cancer, tuberculosis, osteomyelitis, or septic arthritis [8-11].

In addition to being difficult to diagnose, melioidosis can also be difficult to treat. Because there is no vaccine for this disease, treatment consist of antimicrobial therapy. *B. pseudomallei* is an intracellular bacterium in humans and, due to its lifestyle in the environment, is intrinsically resistant to many broad-spectrum antibiotics, such as gentamycin and penicillin [2] [and Hall 2023 (FDC paper)]. It is also resistant to colistin, a cyclic polypeptide antibiotic typically reserved for multi-drug resistant bacteria [12]. Decades of research have resulted in an effective treatment regimen; the current guidelines consist of a combinatory therapy of 10-14 days of ceftazidime or a carbapenem, such as meropenem (MEM), administered intravenously, followed by a 3-6 month course of trimethoprim-sulfamethoxazole taken orally [13, 14]. Within this regimen MEM is usually reserved for severe cases of melioidosis patients in Intensive Care Units (ICUs) [15]. Notably, melioidosis can sometimes exhibit mortality rates as high as 50%, even with medical intervention [3].

Although almost all tested environmental isolates of *B. pseudomallei* have been shown to be susceptible to MEM [16-18], several studies have documented rare, acquired MEM resistance

(MEM^r) in some clinical isolates. And in one study from Australia, some baseline levels of MEM^r (1-4 µg/ml) were observed in clinical and also environmental isolates [19]. In other studies, increases in MEM^r correlated with prolonged antibiotic therapy in the treatment of melioidosis [20, 21]. Although the mechanism for acquired MEM^r has been elucidated in clinical isolates of *B. pseudomallei* [20, 21], the mechanism for apparent intrinsic resistance in the aforementioned environmental isolates from Australia has not been investigated. To further assess the frequency of MEM^r in environmental *B. pseudomallei*, we used a broth microdilution method (BMD) to obtain the MEM MIC for 122 diverse isolates.

Several species in the *Burkholderia cepacia* complex (Bcc) within the *Burkholderia* genus are intrinsically resistant to MEM [22-25]. One Bcc member, *B. ubonensis*, can exhibit MEM resistance as high as >32 µg/ml [24, 25]. No reports of human illnesses have been attributed to this organism, which is unsurprising as it has been demonstrated to be non-pathogenic in murine models [25]. Nevertheless, MEM^r in this bacterial species has been thoroughly investigated due to concerns that it might be a resistance reservoir for *B. pseudomallei*; the high level of MEM^r observed in *B. ubonensis* is attributed to the *penR-penB* expression cassette [18].

B. pseudomallei is naturally competent, with one study suggesting that ~50% of strains are naturally transformable [26]. We further investigate this claim by assessing the presence of genes associated with competency. *B. pseudomallei* has a very large accessory genome [27, 28] and a high recombination rate [29], and spurious gene acquisition has been documented in *B. pseudomallei* from a few related species in the *Burkholderia* genus [30]. Additionally, a separate study demonstrated that *penB*_{Bu278} can be induced in MEM-susceptible *B. ubonensis* and *B. thailandensis*, another member of the Bpc, when challenged with a β-lactam [18]. Importantly, *B. pseudomallei* and *B. ubonensis* apparently occupy the same environmental niche, as they are

often co-isolated together [24, 25, 31, 32]. When considering the frequency and inducibility of MEM^r in Bcc species, the high rates of LGT within the *Burkholderia* genus, and the co-habitation of these two species, there is a question of whether *B. pseudomallei* can acquire genomic content from *B. ubonensis*. In this study, we explore the possibility of *B. pseudomallei* acquiring MEM^r genes from *B. ubonensis* by way of lateral gene transfer (LGT).

CHAPTER 2: MATERIALS AND METHODS

Bacterial strains, growth media and conditions. Bacterial strains used in this study are listed in Table 1. Strains were routinely grown in low-salt L-broth [33] at 37°C overnight. LB used to grow Bp82 and its derivatives was supplemented with 80 µg/ml adenine (LBA). Adenine (80 µg/ml) was also added to the Mueller Hinton agar used in the ETEST® procedures (bioMérieux, Marcy l'Étoile, France).

Table 1. Strain and plasmid details

Strain/plasmid	Description	Reference
<i>B. pseudomallei</i>		
Bp82	Attenuated version of strain 1026b, $\Delta purM$ mutant, exempt from CDC Tier 1 Select Agent regulations	Propst 2010
Bp82.27	Gentamycin-susceptible mutant of Bp82 created by deletion of the <i>amrAB-oprA</i> operon encoding the AmrAB-OprA efflux pump	Choi 2008, Webb 2018
Bp82:: <i>glmS2</i> mini-Tn7- <i>penR</i> ⁺ <i>penB</i> ⁺ _(Bu278)	Bp82:: <i>glmS2</i> mini-Tn7- <i>penR</i> ⁺ <i>penB</i> ⁺ _(Bu278)	This study
Bp82:: <i>glmS2</i> mini-Tn7- <i>rpsL</i> ⁺ _(Bp82) <i>penB</i> ⁺ _(Bu278)	Bp82:: <i>glmS2</i> mini-Tn7- <i>rpsL</i> ⁺ _(Bp82) <i>penB</i> ⁺ _(Bu278)	This study
<i>B. ubonensis</i>		
Bu278	Soil isolate from Puerto Rico that exhibits naturally high levels of MEM ^r (MIC ≥ 32 µg/ml); alternate ID: Bp8955	Price 2017, Hall 2019

Bu597	$\Delta purM$ mutant generated from Bu278	This study
<i>E. coli</i>		
DH5 α	Strain used for plasmid cloning experiments	Liss L. 1987
RHO3	Strain used to facilitate plasmid transfer between species of <i>Burkholderia</i> and <i>E. coli</i> . Requires diaminopimelic acid (DAP) for growth.	López 2009
RHO3/pTNS3	Transposase-producing strain used in mini-Tn7 system.	Choi 2008
Plasmids		
pEDL1005 $\Delta purM$	Plasmid containing 890 bp fragment upstream from <i>purM</i> _{Bu278} gene and the 836 bp fragment downstream from the <i>purM</i> _{Bu278} gene	This study
pPS2280 (<i>penK</i> ⁺ <i>penB</i> ⁺ _(Bu278))	PenB expression from natural promoter	This study
pPS2280 <i>P_{rpsL}</i> (Bp82) <i>penB</i> ⁺ _(Bu2)	PenB expression from <i>rpsL</i> promoter	This study

Overcoming challenges regarding Bp82. Although Bp82 was chosen to simplify laboratory experiments, some challenges had to be managed beforehand. The reasons Bp82 was chosen for the transformation experiment are twofold- it is an exempt strain from the Tier 1 Select Agent Regulations and its parent strain 1026b has been shown to be naturally transformable [26, 34, 35]. The attenuation in Bp82 is due to the deletion of *purM*; this mutant has been proven to be avirulent in murine models due to the disruption in the biochemical pathways that lead to the production of the adenine and thiamine [35]. Therefore, the Bp82 reacquisition of the analogous *purM* from *B. ubonensis* during the transformation experiment could theoretically reverse its avirulent phenotype. Therefore, it was necessary to generate a *B. ubonensis* mutant also containing a *purM* deletion, Bu597, to preclude the possibility of producing a MEM^r strain of Tier 1 SA strain of *B. pseudomallei*. IBC approval was required for these experiments.

Construction of Bu278 $\Delta purM$ mutant. Briefly, genomic DNA was extracted from Bu278 using a QIAamp DNA extraction kit (Qiagen). The upstream and downstream flanking regions of the *purM* gene were PCR amplified using Upstream-F and Upstream-R, yielding a 890 bp amplicon, and Downstream-F and Downstream-R, yielding a 836 bp amplicon, respectively (Table 2).

pEDL1005 was enzymatically digested with NotI and EcoRI (New England Biolabs, Ipswich, MA). The PCR fragments and linearized plasmid were Gibson assembled using the HiFi DNA Assembly Protocol (New England Biolabs, Ipswich, MA). The resulting plasmid, pEDL1005 Δ *purM*, was transformed into DH5 α cells using the heat shock transformation protocol, described below. Transformants were selected on LB plates containing trimethoprim (Tp) (100 μ g/ml) and X-Gal (50 μ g/ml). Plasmid DNA was extracted using a Miniprep kit (Qiagen) and transformed into RHO3 using the heat shock transformation protocol. RHO3 (pEDL1005 Δ *purM*) transformants were selected on LB plates containing 100 μ g/ml Tp and 400 μ g/ml diaminopimelic acid (DAP). Equal volumes of liquid cultures containing Bu278 and RHO3 (pEDL100 Δ *purM*) were plated onto LB plates containing 100 μ g/ml Tp and incubated at 37°C overnight. The resulting trimethoprim resistant (Tp^R) colonies were *B. ubonensis* merodiploids, which were resolved using the I-SceI system [36, 37]. The absence of *purM* was confirmed by PCR using the primers Upstream-F and Downstream-R (Table 2, Figure 1) The strain was renamed Bu597 after the transposon-genome junctions on the plasmid-cured mutant was confirmed with Sanger Sequencing.

Heat shock transformation protocol. Aliquots containing 100 μ l of DH5 α cells were thawed on ice. The Gibson assembled reaction or plasmid DNA to were added to thawed cells. The cell mixture was incubated on ice for 20 minutes without agitation. Afterwards, the 1.5 ml microcentrifuge tube was incubated at 42°C for 45 seconds, then immediately put on ice for 2 minutes. 900 μ l LB broth was added to the tube and subsequently incubated at 37°C for 1 hour with aeration. Aliquots of 50 μ l and 100 μ l were plated onto appropriate media at 37°C overnight.

Table 2. Primers used in this study

Primer name	Sequence (5'→3') *
Upstream-F	<u>cctgttatccctaccgggccaatgcgctcaatctcgc</u>
Upstream-R	<u>gcattccccagcaggtgtggaagatcg</u>
Downstream-F	<u>cacacctgctgggggaatgcaagaaggtc</u>
Downstream-R	<u>gggataacagggtaatcccgggaaatgggtcagcaggttaag</u>
penRpenB-F	aattcgatcatgcatgagctAAACTGTGGACAAAGCGG
penRpenB-R	ttcgcgaggtaccgggccaTGACGCAAGTGGTTGATC
PrpsL-F	ccccgggctgcaggCTCGCCTTCGCATTCCGG
PrpsL-R	gagtaggtcatTGTTTATTCCTGAAATTGACCAAATCGACAC
penB PrpsL-F	AGGAATAACAatgacctactcatcgcaacgtcgaac
penB PrpsL-R	gaggtaccgggcccacgctcaaccgagcgc

*Underlined sequences indicate overlap regions used in Gibson assembly and subsequent transposon mutagenesis reaction. All PCR amplicons were amplified using the Q5® High-Fidelity Taq DNA Polymerase High GC enhancer protocol (New England Biolabs, Ipswich, MA).

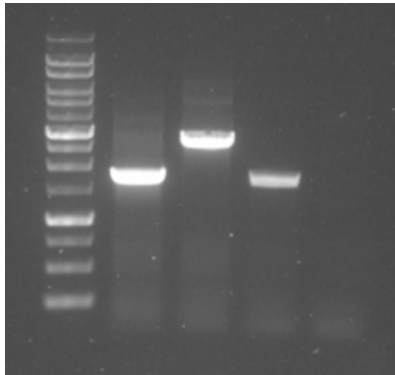


Figure 1. Agarose gel confirmation of *purM* deletion in Bu278 following I-SceI counterselection. Lane 1: 1 kb PLUS DNA Ladder (Goldbio™), Lane 2: *pEDL1005ΔpurM*, Lane 3: Bu278, Lane 4: Bu597, Lane 5: Bp82

Transformation experiment. Experiments were performed as described previously [38]. Liquid cultures of *B. pseudomallei* mutants were grown overnight at 37°C with aeration. Cells were made competent by adding 60 µl of culture to 2 ml of defined media (DM; 0.25X M63 supplemented with 0.2% glucose, 0.4% glycerol, 1 mM MgSO₄, 1 µg/ml thiamine-HCl, and 40 µg/ml each of leucine, isoleucine, valine, tryptophan, glutamic acid, and glutamine) and

incubating at 37°C with aeration until the OD₆₀₀ was ~0.5 (approximately 4-5 hrs.) One ml of Bp82 or Bp82.27 was pelleted, washed, resuspended in 100 µl of fresh broth, and inoculated with 150 ng, 250 ng, or 350 ng of genomic DNA from Bu597 or Bp82, depending on the experiment. The mixtures were incubated at room temperature without agitation for 30 minutes, inoculated with 2 ml of fresh DM, and then incubated overnight at 37°C with aeration. The cultures were centrifuged at 10,000 x g for 1 minute, washed with 1 ml LBA, resuspended in 250 ml of fresh LBA, and inoculated onto LBA plates. The plates used for Bp82 and Bp82.27 were supplemented with MEM (20 µg/ml) and gentamycin (Gm) (15 µg/ml), respectively. Plates were incubated overnight at 37°C.

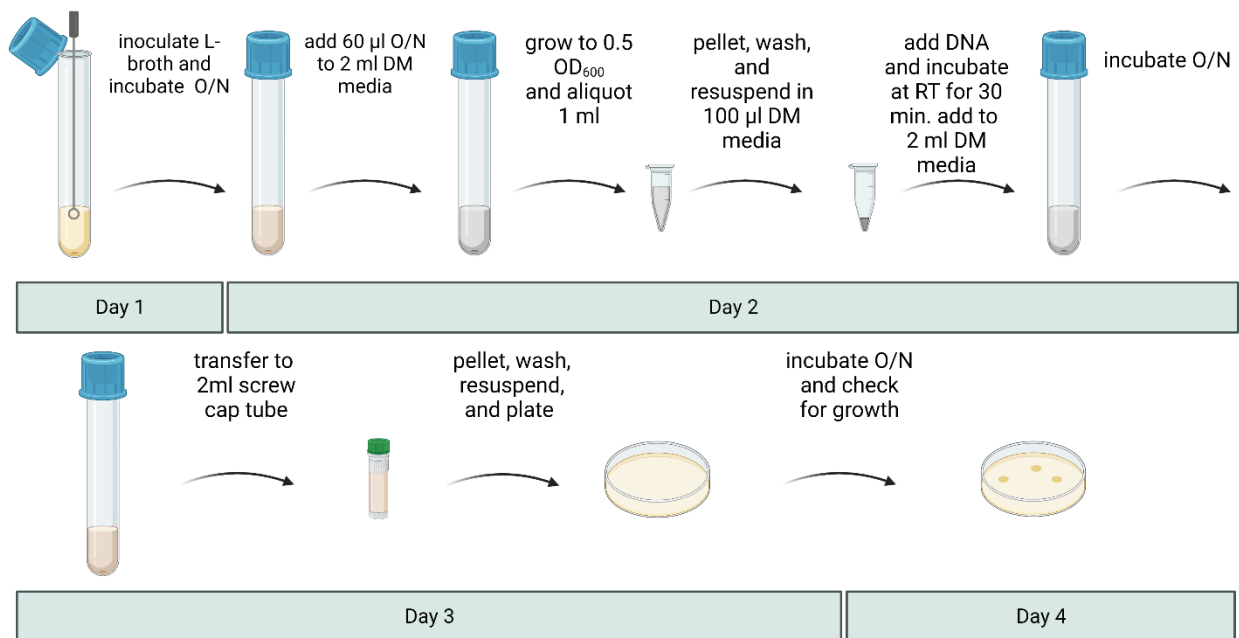


Figure 2. Transformation experiment workflow.

Generation of positive control insertion mutants using a mini-Tn7 system. The 2051 bp *penR-penB* fragment was amplified from Bu278 using primers penRpenB-F and penRpenB-R (Table 2). pPS2280 was enzymatically digested with HindIII and SacI (New England Biolabs,

Ipswich, MA). The amplicon and linearized plasmid were Gibson assembled using the NEBuilder HiFi DNA assembly master mix, resulting in the plasmid pPS2280 ($penR^+penB^+$ (Bu278)). The 231 bp *rpsL* promoter region (P_{rpsL}) was PCR amplified from Bp82 using PrpsL-F and PrpsL-R (Table 2). The 924 bp *penB* gene was PCR amplified from Bu278 using primers penB PrpsL-F and penB PrpsL-R (Table 2). pPS2280 was enzymatically digested with HindIII and EcoRI. The two amplicons and linearized plasmid were Gibson assembled using the NEBuilder HiFi DNA assembly master mix, resulting in the plasmid pPS2280 ($P_{rpsL(Bp82)}penB^+$ (Bu278)). Each plasmid was transformed into DH5 α cells and then subsequently into RHO3 cells using the heat shock transformation protocol. One ml of overnight cultures of Bp82, RHO3 containing the plasmid, and RHO3/pTNS3 were centrifuged at 10,000 x g for 1 minute, washed with 1 ml LB, and resuspended in 50 ml of fresh LB. A mixture containing 25 μ L of each suspension was plated onto LBA supplemented with DAP (400 μ g/ml) and then incubated overnight at 37°C. Bp82 mutants were selected at 37°C on LBA containing kanamycin (1000 μ g/ml).

Both insertion mutants were confirmed by PCR to contain only one copy of their respective gene cluster, and their *glmS*-associated *attTn7* sites were identified using methods described previously [34]. The amplicon insertion sites were determined by PCR using primers that were described in the same study: P_{Tn7L} and BPGLMS1, BPGLMS2, or BPGLMS3 [34].

Meropenem susceptibility testing. Meropenem susceptibility testing was performed using ETEST® strips and following manufacturer's guidelines (bioMérieux, Marcy l'Étoile, France).

Broth microdilution method for screening of environmental *B. pseudomallei* isolates. A set of 122 phylogenetically and spatially diverse environmental isolates of *B. pseudomallei* were

screened for MEM susceptibility using a broth microdilution method, as described elsewhere [39], and in accordance to the Clinical and Laboratory Standards Institute guidelines[40] . Briefly, an isolation streak of *B. pseudomallei* was incubated on LB agar for 48-72 hours at 37°C. Single colonies were diluted in 0.85% saline to reach a 0.5 McFarland Standard. The inoculated saline solution was added to cation-adjusted Mueller-Hinton broth (CAMHB) in a 1/100 dilution. An aliquot of 100 µl of the inoculated CAMHB was added to a pre-dispensed MICRONAUT-S 96-well plate from Merlin Diagnostics (Germany) containing 1-64 µg/ml of MEM, and then incubated for 18-24 hours at 37°C. The absorbance was measured using the accuSkan FC spectrophotometer plate reader (Fisher Scientific) at a wavelength of 620 nm. Positive and negative growth controls were included on every screened plate.

Bioinformatic analysis of homologous recombination sites. The 5 kb flanking regions upstream and downstream of the *penR-penB* expression cassette was obtained from the publicly available genome sequence for *B. ubonensis* strain Bp8955 (accession number ASM385310v1). Each 5 kb region was queried against the publicly available genome for *B. pseudomallei* strain 1026b (accession number ASM95912v1), the parent strain of Bp82, using nucmer v3.1 [41], with the distance mapping extension parameter set to allow 50 mismatches.

Assessment of competency genes in *B. pseudomallei*. Large-scale blast score ratio (LS-BSR) analysis [42] was performed using the -genes parameter. Briefly, 16 genes previously identified [43] as potentially important for natural competency, including both *comE* and *crp*, were queried against 454 publicly available global strains of *B. pseudomallei* [44] using BLAT v.36 [45]. Individual gene presence, absence, and diversity were identified across all samples for each gene using blast scores that ranged between 0-1; a score of 0 indicated complete absence, whereas a score of 1 indicated a perfect match of the gene in that particular strain. BSR cutoffs

were used as described previously [42]. Genes were considered present, and a match, if the BSR value was >0.8 . Gene homologs were characterized by BSR values between 0.8 and 0.4. Genes were considered absent if the BSR value was <0.4 . We chose to visualize the LS-BSR matrix data in the form of heatmap, with yellow representing a score of 0 and indigo representing score of 1. [42]

CHAPTER 3: RESULTS

Transformation of Bp82 did not result in detectable MEM resistance. To ascertain whether *B. pseudomallei* could acquire MEM^r from the environment, we performed several transformation experiments using competent Bp82 cells with varying amounts of Bu597 DNA, which we subsequently plated on LBA containing MEM (20 $\mu\text{g/ml}$). In theory, any transformants containing the MEM^r should be able to overcome this level of carbapenem, in the same manner that *B. ubonensis* can handle $>32 \mu\text{g/ml}$ of MEM. However, these attempts yielded no colonies. In contrast, in a parallel experiment transforming competent Bp82.27 cells with varying amounts of Bp82 DNA, we observed approximately 60 gentamycin resistant colonies (Table 3). Several colonies were streaked for purification and DNA was extracted using a QIAamp DNA extraction kit. Re-acquisition of the *amrAB-oprA* genes in Bp82.27 was confirmed via PCR. This result shows that the transformation methodology works.

Table 3. Transformation experiment results.

Column 2 are the results from the Bp82- Bu597 transformation experiment. Column 3 are the results from the Bp82.27- Bp82 transformation experiment.

Amount of DNA (ng)	No. of MEM-resistant colonies	No. of Gm-resistant colonies
0 -negative control	0	0
150	0	7
250	0	3
350	0	~50

***B. pseudomallei* does not exhibit MEM^r after the addition of Bu278 resistance genes.**

Bp82::*glmS2* mini-Tn7-*penR*⁺*penB*⁺_(Bu278) and Bp82::*glmS2* mini-Tn7-*P_{rpsL}*_(Bp82)*penB*⁺_(Bu278) exhibited little to no increases in MEM^r following the gene introductions. MEM MIC in Bp82::*glmS2* mini-Tn7-*P_{rpsL}*_(Bp82)*penB*⁺_(Bu278) only increased 2.7-fold as compared to its parent strain (Table 4, Figure 2).

Table 4. ETEST® results for Bp82 positive controls.

Reported MIC values are the mode of three biological replicates.

Strain	MEM MIC (µg/ml)
Bp82	0.75
Bp82:: <i>glmS2</i> mini-Tn7- <i>penR</i> ⁺ <i>penB</i> ⁺ _(Bu278)	0.75
Bp82:: <i>glmS2</i> mini-Tn7- <i>P_{rpsL}</i> _(Bp82) <i>penB</i> ⁺ _(Bu278)	2

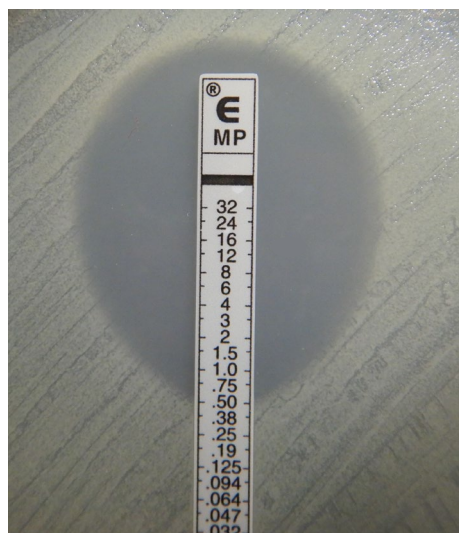


Figure 3. ETEST® result for Bp82.

MEM^r is extremely rare in environmental *B. pseudomallei* isolates. All 122 environmental *B. pseudomallei* were susceptible to MEM (Table 5.) Strains were considered susceptible to MEM if the MIC was ≤ 4 $\mu\text{g/ml}$, per the (M100) CLSI guidelines for Bcc. Data from the Merlin plates were not used if the OD₆₂₀ of the growth control was < 0.2 , or if the negative broth control had an OD₆₂₀ of ≥ 0.2 ; BMD screenings were rerun in those cases.

Sites for homologous recombination are present, but not perfect. Bioinformatic analyses revealed that on either side of the *penR-penB* gene cluster in *B. ubonensis*, there is some limited homology in the 5 kb flanking regions with *B. pseudomallei*, representing potential sites for recombination. Upstream from *penR-penB*, *B. pseudomallei* maps to *B. ubonensis* starting at 2029 bp (out of 5 kb) and breaks the alignment at position 4172. This accounts for 42% (2,143bp) homology of the 5 kb flanking region present in *B. pseudomallei*. Downstream of *penR-penB*, *B. pseudomallei* maps to *B. ubonensis* starting at 1999 (out of 5 kb) but breaks the alignment at position 2837. This accounts for 24.14% (1,207 bp) homology of the second 5k flanking region present in *B. pseudomallei*. Both flanking regions hit on *B. pseudomallei* chromosome 1. Flanking region 1 alignment is between positions 2069769 to 2071936. Flanking region 2 aligns between positions 2061714 to 2062554 as well as 2061188 to 2061555. Thus, although not a contiguous 1,207 base alignment, homologous recombination was not out of the realm of possibility.

BSR data reveals high prevalence of competency genes. The resulting heatmap (Table 6) indicates that these 16 genes are found, in near perfect match, in 98.4% of *B. pseudomallei* strains. Gene homologs and missing genes were found in 1.4% and 0.2% of strains, respectively.

CHAPTER 4: DISCUSSION

Natural Competency is likely found in more than 50% of *B. pseudomallei* strains. The ability of bacteria to take up free DNA from the environment is the defining characteristic of natural competence. Though there are different hypotheses to explain why this phenomenon occurs, there is no question as to whether this occurs in *B. pseudomallei*. It has been suggested that ~50% of *B. pseudomallei* are naturally competent, but the often-cited experiment by Kang et al. [26] is perhaps taken out of context. In that study, a PCR product containing green fluorescent protein was used to assess whether a small set of *B. pseudomallei* strains could acquire and express exogenous DNA. Out of 20 strains, nine were able to express fluorescence. They concluded that the remaining 11 strains were presumed not to be competent. From this result, subsequent researchers routinely cite that roughly half of *B. pseudomallei* strains are naturally competent. However, only 10 wildtype clinical isolates, and no environmental isolates, were included; the rest were mutants generated from those same isolates.

Though it is not uncommon to see variation in competence between strains within the same species [46, 47], it is likely that 50% competency in *B. pseudomallei* is an underestimation. A set of 16 genes have been identified in naturally competent strains of *B. pseudomallei* [43], and a subset of those have been confirmed to have a positive correlation with competency. Heacock-Kang et al. [48] experimentally confirmed that two genes from that list, *comE* and *crp*, were associated with naturally competent strains and were also able to confer competency to strains that could not undergo natural transformation. Additionally, they bioinformatically confirmed that *comE* and *crp* were found in 99 and 198 available *B. pseudomallei* genomes, respectively.

Although our team did not attempt to replicate their findings in the laboratory, we set out to verify their bioinformatics results. Our attempts were ultimately unsuccessful due to the unavailability of their genome list. We conducted our own LS-BSR analysis which resulted in a heatmap of the 16 genes queried against 454 available genomes of *B. pseudomallei*. Because majority of our data points were greater than 0.8, we presume it is highly likely that these genes will be present in any given strain of *B. pseudomallei*.

We propose that majority, not just ~50%, of *B. pseudomallei* strains are naturally competent. If we consider what has been established by the Norris and Heacock-Kang studies, along with our own LS-BSR analysis, we contend that percentage of naturally transformable strains is much higher. In other bacterial species, the presence of specific genes has been identified as the determinant factor for competence [49-51], so it is reasonable to conclude about the effect these genes might have on *B. pseudomallei*. Although we can bioinformatically assess the likelihood of natural competence, the gold standard would be laboratory confirmation, which would be significant undertaking.

***B. pseudomallei* is unlikely to acquire MEM^r from the environment.**

The focus of this study was to investigate the likelihood of *B. pseudomallei* acquiring MEM^r genes from exogenous DNA from *B. ubonensis*. However, based on the lack of evidence for LGT between the two species in this study, we conclude that such events are unlikely to occur. To strengthen this finding, we performed an additional experiment to ensure the validity of the transformation experiment. We utilized Bp82.27, a gentamycin-susceptible mutant of Bp82 created by deletion of the *amrAB-oprA* genes, exposed to genomic Bp82 DNA. PCR analysis of several colonies recovered from the LBA plates supplemented with Gm (15 µg/ml) showed that

the Bp82.27 cells re-incorporated the *amrAB-oprA* genes into its chromosome, thus demonstrating that resistance can be re-acquired via this method. Because Bp82.27 proved capable of DNA uptake, we can conclude that our results observed in the Bp82-Bu597 transformation experiment were true negatives.

The generation of positive control strains allowed us to assess any effects PenB might have on *B. pseudomallei*. PenB is transcribed by *penB*_(Bu278), which is under transcriptional control of PenR, a LysR regulator encoded by *penR*_(Bu278) [18, 52, 53]. The translation of PenR is initiated when the cell experiences peptidoglycan cell wall degradation caused by a beta lactam challenge, in our case MEM [18, 52, 53]. The Bp82 insertion mutant containing the native Bu278 resistance cassette, *penR*⁺-*penB*⁺_(Bu278), displayed no increase in MEM MIC compared to the wild-type Bp82 strain. These results were contrary to our initial assumption that introducing the resistance genes into Bp82 would result in MEM^f levels comparable to Bu278 (MIC >32 µg/ml). These data suggest that *B. pseudomallei* would not exhibit MEM^f despite acquiring the *penR*⁺-*penB*⁺_(Bu278) resistance cassette.

Given that the expression of PenB is dependent on PenR, we generated a *B. pseudomallei* mutant with a constitutive promoter preceding *penB*. We created a control strain of Bp82 containing a resistance cassette consisting of *P*_{rpsL}, a promoter that is transcriptionally independent of MEM levels in the cytoplasm [54], paired with *penB*_(Bu278). *P*_{rpsL} is a relatively strong species-specific promoter, in our case, for *B. pseudomallei* [55]; utilizing this promoter in conjunction with *penB*_(Bu278) should have established whether *B. pseudomallei* can overcome high levels of MEM in the cytoplasm. However, the observed MIC values in this positive control was only 2.7 times more than Bp82, the parent strain of these mutants. These results indicate that *penB*_(Bu278) can be active within *B. pseudomallei* but at a low level, which are currently attributed to an unknown

factor(s). With that said, these results are in keeping with the MEM susceptibility levels we observed in our screening of 122 environmental *B. pseudomallei* strains, as well as the abovementioned Australian study [19]

Although we have determined that MEM^r is highly unlikely to be conferred to *B. pseudomallei* via transformation, there is evidence to suggest that LGT is possible between *B. pseudomallei* and *B. ubonensis*. Intra-species transfer of genomic content has been observed numerous times between Bpc members *B. pseudomallei* and *B. thailandensis* [29, 56-59], giving us reason to presume that transfers might occur between *B. pseudomallei* and *B. ubonensis*. However, perhaps the lack of evidence for LGT in these two species is best explained by their placement within the *Burkholderia* phylogeny [28]. It is possible that these genetic differences are too insurmountable for any meaningful gene transfers to occur.

We also explored an explanation for the induction of carbapenemase activity in *B. ubonensis* and *B. thailandensis* [18] that is not observed in *B. pseudomallei*. A gene content analysis showed 5 genes common to both *B. ubonensis* and *B. thailandensis*, but not found in *B. pseudomallei*: D-xylose ABC transporter, D-xylose-binding, xylose isomerase, mercuric reductase, integrase core domain protein, and arabinose ABC transporter substrate-binding. A sixth gene, an IS256 family insertion element, has been implicated in maintaining antibiotic resistance in other bacterial species [60-64]. It is observed in many *Burkholderia* spp. but is only found in 3 public *B. pseudomallei* genomes. Perhaps the low occurrence of this gene in *B. pseudomallei*, including 1026B/Bp82, contributes to the difficulty we observed in activating carbapenemase activity. With that being said, the presence of this transposable element does not tell the entire story; insertion location can also influence gene expression.

Meropenem resistance is important for some members of the Bcc, but maybe not the Bpc.

As previously mentioned, MEM^r is found throughout the Bcc. It is evident to see the advantage this adaption can provide to pathogenic members of the Bcc, such as *B. cepacia* or *B. multivorans*, as well as the soil-dwelling species in the group, such as *B. ubonensis*. An interesting point to consider is the differences we see in MEM^r profiles between spatially distinct populations of *B. ubonensis*. For example, there are significant differences in MEM susceptibility profiles in the Australian population compared to the Puerto Rican populations [19, 24, 25]. There are no details for the low resistance seen in Australia versus the high resistance seen in Puerto Rico, although the mechanism for carbapenem resistance in the latter population has been well described [18].

Conversely, the opposite is true of *B. pseudomallei* with regards to MEM^r. Again, with the exception of a few Australian isolates, all environmental strains seem to be susceptible to MEM (Supplemental File 1) [24]. It should be noted, however, that the mechanisms for MEM^r in these two species differ considerably. In Bcc, resistance is imparted by way of generating a carbapenemase, such as PenB. Whereas in the Bpc, resistance is due to mutations in the extended spectrum beta lactamases (ESBL) [65, 66] or mutations in resistance-nodulation-division (RND) efflux pump regulators [20]. Though having MEM^r obviously has its advantages in a competitive setting, like soil, *B. pseudomallei* can survive and persist quite sufficiently without it. We presume, then, that this resistant phenotype is therefore not critical to the survival and persistence of other *Burkholderia spp* in the same niche.

CHAPTER 5: CONCLUDING REMARKS

Melioidosis is estimated to kill roughly 89,000 people annually [67], and the complexity surrounding *B. pseudomallei* only serves to confound the situation. Antimicrobial resistance

(AMR) presents intricate challenges in the treatment of this disease. Fortunately, we can exploit the meropenem susceptibility we see in *B. pseudomallei*. From what we can deduce, there is not yet a mechanism by which it can acquire clinically significant resistance to this invaluable antibiotic. In short, the mechanism previously proposed is not a factor of resistance; this bacteria should remain susceptible.

The idea that *B. pseudomallei* could acquire resistance from *B. ubonensis* is a valid concern we believe we have addressed. With AMR projected to be the leading cause of human mortality by the year 2050 [68], it is imperative that we continue to investigate any and all mechanisms that may contribute to the resistome of the *Burkholderiaceae* family. Though we may not have identified a potential source of AMR for *B. pseudomallei*, the contribution of this work to the field should not go unnoticed.

Table 5. MEM MIC of 122 environmental *B. pseudomallei* isolates.
S=Susceptible.

Culture NAU ID	Original ID	Source	Country	Year	MEM MIC	MEM call
Bp6738	MSHR1435	Environmental	AUSTRALIA	2002	≤1	S
Bp9714	MSHR1588WGS	Environmental	AUSTRALIA	2003	≤1	S
Bp9715	MSHR2575WGS	Environmental	AUSTRALIA	2007	≤1	S
Bp8783	MSHR4033WGS	Environmental	AUSTRALIA	2010	≤1	S
Bp8784	MSHR4034WGS	Environmental	AUSTRALIA	2010	≤1	S
Bp8047	MSHR5310WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp8048	MSHR5311WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp8049	MSHR5913WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp8050	MSHR5916WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp8051	MSHR5922WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp8051-P20	MSHR5922WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp8052	MSHR5930WGS	Environmental	AUSTRALIA	2011	1	S
Bp8052-P20	MSHR5930WGS	Environmental	AUSTRALIA	2011	2	S
Bp8053	MSHR6540WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp8054	MSHR6541WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp8055	MSHR6542WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp8055-P20	MSHR6542WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp8056	MSHR6652WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp8056-P20	MSHR6652WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp8057	MSHR6725WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp8058	MSHR6919WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp9701	MSHR5998WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp9704	MSHR7145WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp9685	MSHR7797WGS	Environmental	AUSTRALIA	2013	2	S
Bp6746	MSHR1895	Environmental	AUSTRALIA	2004	≤1	S
Bp9610	MSHR1869WGS	Environmental	AUSTRALIA	2004	≤1	S
Bp9700	MSHR2188WGS	Environmental	AUSTRALIA	2005	≤1	S
Bp9611	MSHR2572WGS	Environmental	AUSTRALIA	2007	≤1	S
Bp9737	MSHR2495WGS	Environmental	AUSTRALIA	2007	≤1	S
Bp9738	MSHR2852WGS	Environmental	AUSTRALIA	2007	≤1	S
Bp6609	MSHR4378	Environmental	AUSTRALIA	2010	≤1	S
Bp9612	MSHR4304WGS	Environmental	AUSTRALIA	2010	2	S
Bp9613	MSHR4305WGS	Environmental	AUSTRALIA	2010	≤1	S
Bp9614	MSHR4306WGS	Environmental	AUSTRALIA	2010	≤1	S
Bp9615	MSHR4307WGS	Environmental	AUSTRALIA	2010	≤1	S
Bp9616	MSHR4308WGS	Environmental	AUSTRALIA	2010	≤1	S

Bp9617	MSHR4343WGS	Environmental	AUSTRALIA	2010	2	S
Bp9618	MSHR4461WGS	Environmental	AUSTRALIA	2010	≤1	S
Bp9619	MSHR4652WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9620	MSHR4690WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9621	MSHR5069WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9622	MSHR5593WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9623	MSHR5929WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9624	MSHR5931WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9625	MSHR5932WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9626	MSHR5933WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9627	MSHR5934WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9628	MSHR5935WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9629	MSHR5936WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9630	MSHR5937WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9631	MSHR5938WGS	Environmental	AUSTRALIA	2011	2	S
Bp9637	MSHR7951WGS	Environmental	AUSTRALIA	2011	≤1	S
Bp9632	MSHR6146WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp9633	MSHR7146WGS	Environmental	AUSTRALIA	2012	2	S
Bp9634	MSHR7147WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp9635	MSHR7148WGS	Environmental	AUSTRALIA	2012	2	S
Bp9636	MSHR7149WGS	Environmental	AUSTRALIA	2012	≤1	S
Bp9647	MSHR9684WGS	Environmental	AUSTRALIA	2016	2	S
Bp9648	MSHR9685WGS	Environmental	AUSTRALIA	2016	2	S
Bp9649	MSHR9711WGS	Environmental	AUSTRALIA	2016	≤1	S
Bp9711	MSHR9360WGS	Environmental	AUSTRALIA	2016	≤1	S
Bp9707	MSHR9933WGS	Environmental	AUSTRALIA	2017	≤1	S
Bp9708	MSHR9934WGS	Environmental	AUSTRALIA	2017	≤1	S
Bp9709	MSHR9980WGS	Environmental	AUSTRALIA	2017	≤1	S
Bp9673	MSHR11761WGS	Environmental	AUSTRALIA	2018	2	S
Bp9674	MSHR11763WGS	Environmental	AUSTRALIA	2018	2	S
Bp9675	MSHR11793WGS	Environmental	AUSTRALIA	2018	≤1	S
Bp9676	MSHR11878WGS	Environmental	AUSTRALIA	2018	2	S
Bp0044	2002721629	Environmental	KENYA	1992	≤1	S
Bp0045	2002721628	Environmental	MADAGASCAR	1977	≤1	S
Bp9039	PR_site23_hole07_PIG S_plate1_sub1	Environmental	PUERTO RICO	2017	≤1	S
Bp9039-P20	PR_site23_hole07_PIG S_plate1_sub1	Environmental	PUERTO RICO	2017	≤1	S
Bp9110	PRsite23hole09try2P1sub1	Environmental	PUERTO RICO	2017	≤1	S
Bp0037	2002721785	Environmental	THAILAND	1965	≤1	S
Bp0046	2002721784	Environmental	THAILAND	1965	≤1	S
Bp0058	2002721789	Environmental	THAILAND	1965	≤1	S
Bp0064	2002721788	Environmental	THAILAND	1965	≤1	S
Bp0374	RF8-BP5	Environmental	THAILAND	2007	≤1	S

Bp0607	RF44-BP52	Environmental	THAILAND	2007	≤1	S
Bp0622	RF49-BP5	Environmental	THAILAND	2007	2	S
Bp1355	RF87-BP2	Environmental	THAILAND	2007	≤1	S
Bp8884	INT2-BP270	environmental	THAILAND	2007	≤1	S
Bp8885	INT2-BP12	Environmental	THAILAND	2007	2	S
Bp8890	INT2-BP108	environmental	THAILAND	2007	≤1	S
Bp8892	INT2-BP123	environmental	THAILAND	2007	≤1	S
Bp8867	Bp1761	Environmental	THAILAND	-	≤1	S
Bp8870	Bp1798	Environmental	THAILAND	-	≤1	S
Bp8872	Bp1826	Environmental	THAILAND	-	≤1	S
Bp8874	Bp1837	Environmental	THAILAND	-	≤1	S
Bp8876	Bp1864	Environmental	THAILAND	-	≤1	S
Bp8877	Bp1921	Environmental	THAILAND	-	≤1	S
Bp8878	Bp1951	Environmental	THAILAND	-	≤1	S
Bp8879	Bp1954	Environmental	THAILAND	-	≤1	S
Bp8881	Bp1972	Environmental	THAILAND	-	≤1	S
Bp8882	Bp1978	Environmental	THAILAND	-	≤1	S
Bp8883	Bp2001	Environmental	THAILAND	-	≤1	S
Bp0245	RF4-BP80	Environmental	THAILAND	2007	2	S
Bp0347	RF6-BP26	Environmental	THAILAND	2007	≤1	S
Bp0371	RF8-BP2	Environmental	THAILAND	2007	≤1	S
Bp0619	RF49-BP2	Environmental	THAILAND	2007	≤1	S
Bp0624	RF49-BP7	Environmental	THAILAND	2007	≤1	S
Bp0704	NRF57-BP67	Environmental	THAILAND	2007	≤1	S
Bp0755	RF61-BP2	Environmental	THAILAND	2007	≤1	S
Bp0938	RF68-BP5	Environmental	THAILAND	2007	≤1	S
Bp10019	180-10-P1-S2	environmental	United States	2020	≤1	S
Bp10023	213-18-P1-S1	environmental	United States	2020	≤1	S
Bp10043	219-14-P1-S1	environmental	United States	2020	≤1	S
Bp10068	209-03-P1-S1	environmental	United States	2020	≤1	S
Bp10077	210-11-P1-S1	environmental	United States	2020	≤1	S
Bp10081	211-06-P1-S1	environmental	United States	2020	≤1	S
Bp0054	2002721787	Environmental	UNKNOWN	-	≤1	S
Bp0055	2002721786	Environmental	UNKNOWN	-	≤1	S
Bp8832	3946	Environmental	UNKNOWN	-	≤1	S
Bp8832-P20	3946	Environmental	UNKNOWN	-	≤1	S
Bp8833	4102	Environmental	UNKNOWN	-	≤1	S
Bp8833-P20	4102	Environmental	UNKNOWN	-	≤1	S
Bp8834	4117	Environmental	UNKNOWN	-	≤1	S
Bp8834-P20	4117	Environmental	UNKNOWN	-	2	S
Bp8835	4259	Environmental	UNKNOWN	-	≤1	S
Bp8835-P20	4259	Environmental	UNKNOWN	-	≤1	S

Bp8838	7406	Environmental	UNKNOWN	-	≤ 1	S
Bp8838-P20	7406	Environmental	UNKNOWN	-	≤ 1	S

Table 6. LS-BSR heat map.

BSR values >0.8 represented genes that were present and matching. BSR values between 0.8 and 0.4 indicated gene homologs. BSR values <0.4 represented missing genes.

	BP1026B_I0804	BP1026B_II2056	BP1026B_I0809	BP1026B_I0819	BP1026B_I0820	BP1026B_I2363	BP1026B_I2369	BP1026B_I2371	BP1026B_I2562	BP1026B_I2563	BP1026B_I2566	BP1026B_I2574	BP1026B_I2578	BP1026B_II2062	BP1026B_II2080	BP1026B_II2082
GCA_0019764 75.1_Burkhold eria_pseudomal lei_4027a_Cont ig_STnovel	1	1	0.99	1	1	1	1	0.99	1	1	1	0.71	1	0.95	0.99	1
GCA_0019764 15.1_Burkhold eria_pseudomal lei_200272174 0_Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1
GCA_0019784 45.1_Burkhold eria_pseudomal lei_NAU14A_4 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019808 95.1_Burkhold eria_pseudomal lei_942a_Conti g_ST17	0.99	1	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
BP1026B_gene s	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1

GCA_0019782 05.1_Burkholderia_pseudomallei_MSHR0776 Contig_ST150	1	1	1	1	1	0.99	0.99	0.99	0.99	0.99	1	0.99	1	0.95	1	1
GCA_0007559 45.1_Burkholderia_pseudomallei_MSHR5858 _Complete_Genome_ST562	0.99	1	0.99	1	1	0.99	0.99	1	1	1	1	0.7	1	0.95	1	1
GCA_0014459 15.1_Burkholderia_pseudomallei_RF6BP15 _Contig_ST178	1	1	1	0.99	1	1	1	0.99	1	1	1	0.99	1	1	0.99	1
GCA_0009321 05.1_Burkholderia_pseudomallei_QCMRI_BP18_Scaffold_ST252	1	0.99	1	1	1	0.99	1	1	0.99	0.99	1	1	1	0.95	0.99	1
GCA_0007732 35.1_Burkholderia_pseudomallei_ABCPW_107_Contig_ST1024	1	0.99	0.99	1	1	1	0.99	0.99	0.99	1	1	0.99	1	0.95	0.99	1
GCA_0019780 15.1_Burkholderia_pseudomallei_Ubon_P19_Bp11_Contig_ST670	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1
GCA_0019778 75.1_Burkholderia_pseudomallei_INT2_BP175_Contig_ST60	0.98	1	1	1	0.99	0.99	1	0.99	1	1	1	0.7	1	1	1	1

GCA_0019751 45.1_Burkhold eria_pseudomal lei_MSHR3763 _Complete_Ge nome ST36	0.99	0.99	1	1	1	0.99	0.99	1	1	1	1	0.99	1	0.99	0.99	1
GCA_0019763 85.1_Burkhold eria_pseudomal lei_200272178 6_Contig ST84	0.99	1	1	1	1	1	0.99	0.99	0.67	1	1	0.7	1	1	0.99	1
GCA_0012330 45.1_Burkhold eria_pseudomal lei_23_96_Con tig ST51	0.99	1	0.99	0.99	0.99	1	0.99	0.99	0.99	1	1	0.7	1	1	0.99	1
GCA_0019775 75.1_Burkhold eria_pseudomal lei_NRF60_BP 3_Contig_STno vel	0.99	1	1	1	1	0.99	1	1	1	1	1	0.99	1	1	1	1
GCA_0014458 55.1_Burkhold eria_pseudomal lei_Gu1909a_C ontig_STtrunca ted	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.7	1	1	0.99	1
GCA_0028882 85.1_Burkhold eria_pseudomal lei_MSHR1079 _Contig ST111	1	0.99	1	1	0.99	0.99	1	1	1	1	1	1	1	0.95	0.99	1
GCA_0019803 05.1_Burkhold eria_pseudomal lei_MSHR4483 _Contig ST109	0.99	1	0.99	1	0.99	1	0.98	0.99	0.99	0.99	1	0.99	1	1	1	1
GCA_0019783 65.1_Burkhold	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1

eria_pseudomalei_NAU13B_1_Contig_ST617																
GCA_001976585.1_Burkholderia_pseudomalei_4112a_Contig_STnovel	1	1	1	1	1	1	0.99	0.99	0.99	1	1	1	1	0.95	0.99	1
GCA_001980245.1_Burkholderia_pseudomalei_MSHR4250_Contig_ST109	1	1	0.99	1	0.99	1	0.98	0.99	0.99	0.99	1	0.99	1	1	1	1
GCA_001979545.1_Burkholderia_pseudomalei_MSHR0443_Contig_ST243	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_001978175.1_Burkholderia_pseudomalei_Ubon_P45_Bp25_Contig_STnovel	1	1	0.99	1	0.99	0.75	1	1	1	0.86	1	1	1	0.95	0.99	1
GCA_000350505.1_Burkholderia_pseudomalei_NCTC_13392_Contig_ST23	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.99	1	0.95	0.96	1
GCA_000755925.1_Burkholderia_pseudomalei_HBPUB10134a_Complete_Genome_ST228	1	1	0.99	1	1	1	0.99	0.99	1	1	1	1	1	1	0.99	1
GCA_001445875.1_Burkholderia_pseudomalei	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.71	1	1	0.99	1

lei_RF67BP1_Contig_STtruncated																	
GCA_000773155.1_Burkholderia_pseudomallei_MSHR5609_Contig_ST1029	1	1	1	1	1	1	1	1	1	1	1	1	0.99	1	0.98	0.99	1
GCA_000259735.1_Burkholderia_pseudomallei_1026a_Contig_ST102	1	0.38	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
GCA_001977745.1_Burkholderia_pseudomallei_RF87_BP2_Contig_STnovel	1	1	1	1	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
GCA_001978725.1_Burkholderia_pseudomallei_NAU20B_8_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	0.99	1
GCA_002921005.1_Burkholderia_pseudomallei_UM129_Contig_ST1342	1	1	1	0.99	1	1	0.99	1	0.99	1	1	0.99	1	0.95	0.99	0.99	1
GCA_001980965.1_Burkholderia_pseudomallei_984a_Contig_ST70	1	1	1	1	1	0.99	0.99	0.99	1	1	1	1	0.71	1	1	0.99	1
GCA_002110925.1_Burkholderia_pseudomallei_200272168	1	0.99	0.99	1	1	0.99	1	1	0.99	0.99	1	0.99	1	0.95	0.99	0.99	1

4_Chromosome ST297																	
GCA_0007751 15.1_Burkhold eria_pseudomal lei_TSV44_Co ntig ST276	1	1	0.99	1	1	1	0.99	0.99	1	0.99	1	0.99	1	0.95	0.99	1	
GCA_0007561 65.1_Burkhold eria_pseudomal lei_MSHR1655 _Complete_Ge nome ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1	
GCA_0019785 05.1_Burkhold eria_pseudomal lei_NAU14A_8 Contig ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1	
GCA_0019788 35.1_Burkhold eria_pseudomal lei_NAU22A_5 Contig ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1	
GCA_0019769 65.1_Burkhold eria_pseudomal lei_RF62_BP4 1_Contig_ST11 4	1	0.99	0.99	1	1	1	0.99	0.99	1	0.99	1	0.99	1	0.95	1	1	
GCA_0005216 45.1_Burkhold eria_pseudomal lei_MSHR146_ Complete_Gen ome ST617	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1	
GCA_0007756 85.1_Burkhold eria_pseudomal lei_A79D_Cont ig ST667	1	1	0.99	1	1	1	0.99	0.99	0.99	1	1	0.99	1	1	0.99	1	

lei_NAU35A3_Contig_ST326																
GCA_0019799 75.1_Burkholderia_pseudomallei_MSHR0347_Contig_ST617	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	0.74	1	0.95	0.99	1
GCA_0009592 85.1_Burkholderia_pseudomallei_K96243_Complete_Genome_ST10	1	1	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
GCA_0019807 55.1_Burkholderia_pseudomallei_316c_Contig_ST17	1	1	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
GCA_0019762 75.1_Burkholderia_pseudomallei_3904b_Contig_ST206	1	1	0.99	1	0.99	1	1	1	0.99	1	1	0.99	1	1	0.99	1
GCA_0009812 85.2_Burkholderia_pseudomallei_cm_manipal_Contig_ST1478	0.98	1	0.99	0.99	1	0.82	1	0.99	0.99	0.99	1	0.62	1	0.95	1	1
GCA_0019773 75.1_Burkholderia_pseudomallei_4210a_Contig_ST169	0.99	1	0.99	1	0.99	1	0.99	1	0.99	1	1	0.71	1	1	0.99	1
GCA_0019794 95.1_Burkholderia_pseudomallei_MSHR0376_Contig_ST243	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1

GCA_0019799 65.1_Burkhold eria_pseudomal lei_MSHR0474 _Contig_ST135 6	1	0.99	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	0.99	0.99	1
GCA_0019806 05.1_Burkhold eria_pseudomal lei_MSHR5093 _Contig_ST881	0.99	0.99	1	0.99	0.99	0.99	0.99	0.99	1	1	1	0.99	1	1	1	1
GCA_0007742 65.1_Burkhold eria_pseudomal lei_MSHR4377 _Scaffold_ST1 020	1	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	0.99	0.99	1
GCA_0019778 05.1_Burkhold eria_pseudomal lei_INT2_BP10 2_Contig_ST93	1	1	0.99	1	1	1	0.99	1	1	1	1	1	1	1	1	1
GCA_0007570 35.2_Burkhold eria_pseudomal lei_Pasteur_52 237_Complete_ Genome_ST41 1	0.99	1	1	1	1	1	0.99	0.99	1	1	1	1	1	1	0.99	1
GCA_0021112 05.1_Burkhold eria_pseudomal lei_201374677 7_Complete_G enome_ST518	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.98	1	0.95	0.99	1
GCA_0019800 75.1_Burkhold eria_pseudomal lei_MSHR0120 _Contig_ST259	1	1	1	1	1	0.99	0.98	1	0.99	0.87	1	0.99	1	0.94	1	1

GCA_0019775 45.1_Burkholderia_pseudomallei_NRF57_BP64_Contig_ST670	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1
GCA_0019764 65.1_Burkholderia_pseudomallei_2002721789_Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1
GCA_0019761 75.1_Burkholderia_pseudomallei_2002721712_Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	0.99	1	0.67	1	1	0.99	1
GCA_0028879 55.1_Burkholderia_pseudomallei_MSHR2543_Contig_STnovel	1	0.99	1	0.99	1	0.99	0.99	1	0.89	0.99	1	1	1	0.95	1	1
GCA_0019802 65.1_Burkholderia_pseudomallei_MSHR3974_Contig_ST846	1	1	1	1	1	1	1	0.99	0.99	1	1	0.99	1	1	1	1
GCA_0019772 25.1_Burkholderia_pseudomallei_Ubon_P19_Bp45_Contig_ST670	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1
GCA_0007705 35.1_Burkholderia_pseudomallei_A79A_Complete_Genome_ST667	1	1	0.99	1	1	1	0.99	0.99	0.99	1	1	0.99	1	1	0.99	1

lei_NAU2B_8 Contig ST326																	
GCA_0019786 75.1_Burkhold eria_pseudomal lei_NAU20B_5 Contig ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	0.99	1
GCA_0019793 35.1_Burkhold eria_pseudomal lei_MSHR3042 Contig ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	0.99	1
GCA_0019808 15.1_Burkhold eria_pseudomal lei_577ci_Cont ig ST708	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.71	1	1	0.99	0.99	1
GCA_0019805 65.1_Burkhold eria_pseudomal lei_NAU33A_4 Contig ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	0.99	1
GCA_0019806 75.1_Burkhold eria_pseudomal lei_MSHR5105 Contig ST881	0.99	0.99	1	0.99	0.99	0.99	0.99	0.99	1	1	1	0.99	1	1	1	1	1
GCA_0014461 55.1_Burkhold eria_pseudomal lei_RNS7Bp6_ Contig ST60	0.99	1	1	1	0.99	0.99	1	0.99	0.99	1	1	0.71	1	1	1	1	1
GCA_0019806 25.1_Burkhold eria_pseudomal lei_MSHR5086 Contig ST658	0.99	1	0.99	0.99	0.99	1	0.99	0.99	1	1	1	0.7	1	1	0.99	0.99	1
GCA_0009593 45.1_Burkhold eria_pseudomal	1	1	0.99	1	1	1	0.99	0.99	0.99	1	1	0.99	1	0.95	0.99	0.99	1

lei_PB0829801 0_Complete_G enome_ST426																	
GCA_0019775 25.1_Burkhold eria_pseudomal lei_NRF57_BP 62_Contig_ST6 70	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1	
GCA_0028809 25.1_Burkhold eria_pseudomal lei_DL35_Cont ig_ST507	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.7	1	1	0.99	1	
GCA_0004108 95.1_Burkhold eria_pseudomal lei_406e_Conti g_ST211	1	1	0.99	1	0.99	0.99	0.99	1	0.99	1	1	0.71	1	1	0.99	1	
GCA_0007561 45.1_Burkhold eria_pseudomal lei_1106a_Com plete_Genome_ ST70	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.7	1	1	0.99	1	
GCA_0016370 15.1_Burkhold eria_pseudomal lei_NAU21B_1 3_Contig_ST17 4	0.99	1	0.99	1	1	0.99	1	1	1	1	1	1	1	0.95	1	1	
GCA_0019791 75.1_Burkhold eria_pseudomal lei_MSHR1048 Contig_ST131	1	0.68	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0	0	0.13	
GCA_0014459 75.1_Burkhold eria_pseudomal	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1	

lei_NAU14B6_Contig_ST326																
GCA_0028436 45.1_Burkholderia_pseudomallei_MSHR1435_Complete_Genome_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0002605 15.1_Burkholderia_pseudomallei_1026b_Complete_Genome_ST102	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
GCA_0019811 25.1_Burkholderia_pseudomallei_3964d_Contig_ST671	1	1	0.99	1	1	0.99	1	1	1	0.86	1	1	1	0.95	0.99	1
GCA_0019765 45.1_Burkholderia_pseudomallei_4108a_Contig_ST158	1	1	1	1	1	0.99	1	1	1	1	1	1	1	1	0.99	1
GCA_0019781 85.1_Burkholderia_pseudomallei_MSHR0465A_Contig_ST132	1	1	0.99	1	0.99	1	0.99	0.99	0.99	0.99	1	0.99	1	0.95	0.99	1
GCA_0028880 55.1_Burkholderia_pseudomallei_MSHR146_Contig_ST617	1	1	1	1	0.99	0.99	0.99	0.96	0.99	1	1	1	1	0.95	0.99	1
GCA_0019805 15.1_Burkholderia_pseudomallei_DW10_Contig_ST333	1	0.36	0.99	0.99	1	0.99	0.99	1	1	1	1	1	1	0.95	0.99	1

GCA_0021112 45.1_Burkhold eria_pseudomal lei_201374687 7_Complete_G enome_ST1038	1	0.99	0.99	1	0.99	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0019770 85.1_Burkhold eria_pseudomal lei_INT4_BP18 Contig_ST60	0.99	1	1	1	0.99	0.99	1	0.99	1	1	1	0.71	1	1	1	1
GCA_0019805 45.1_Burkhold eria_pseudomal lei_DW7_Conti g_STnovel	1	0.36	0.99	0.99	1	0.99	0.99	1	0.99	0.99	1	0.99	1	0.95	0.99	1
GCA_0019772 75.1_Burkhold eria_pseudomal lei_Ubon_P45_ Bp30_Contig_ ST671	1	1	0.99	1	0.99	0.99	1	1	1	0.86	1	1	1	0.95	0.99	1
GCA_0028879 65.1_Burkhold eria_pseudomal lei_MSHR338_ Contig_ST243	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_0019788 75.1_Burkhold eria_pseudomal lei_NAU24B_3 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0014459 55.1_Burkhold eria_pseudomal lei_MSHR465a Contig_ST132	1	1	0.99	1	0.99	1	0.99	0.99	0.99	0.99	1	0.99	1	0.95	0.99	1
GCA_0028810 15.1_Burkhold eria_pseudomal	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1

lei_STW_214_Contig_ST84																
GCA_0028879 75.1_Burkholderia_pseudomallei_MSHR730_Contig_ST103	1	0.99	1	0.99	0.99	0.99	0.99	1	0.99	0.84	1	1	1	0.95	1	1
GCA_0004547 05.1_Burkholderia_pseudomallei_NCTC_13392_morphotype_8_Contig_ST23	1	1	0.99	1	0.99	0.99	0.99	1	1	0.78	1	0.99	1	0.95	0.96	1
GCA_0019791 15.1_Burkholderia_pseudomallei_MSHR0800_Contig_ST114	1	0.99	0.99	1	1	1	0.99	0.99	1	0.99	1	0.99	1	0.95	1	1
GCA_0019771 25.1_Burkholderia_pseudomallei_RNS7_Bp6_Contig_ST60	0.99	1	1	1	0.99	0.99	1	0.99	0.99	1	1	0.71	1	1	1	1
GCA_0019768 85.1_Burkholderia_pseudomallei_RF8_BP5_Contig_ST178	1	1	1	0.99	1	1	1	0.99	1	1	1	0.99	1	1	0.99	1
GCA_0019787 95.1_Burkholderia_pseudomallei_NAU21B_11_Contig_ST617	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019787 65.1_Burkholderia_pseudomallei_NAU21B_3_Contig_ST617	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1

GCA_0019808 35.1_Burkhold eria_pseudomal lei_577d_Conti g_ST708	1	0.51	0.99	1	0.99	0.99	0.99	1	1	1	1	0.7	1	1	0.99	1
GCA_0019776 45.1_Burkhold eria_pseudomal lei_RF67_BP1 _Contig_STtru ncated	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.71	1	1	0.99	1
GCA_0009592 05.1_Burkhold eria_pseudomal lei_MSHR491_ Complete_Gen ome_ST126	1	0.99	0.99	0.99	0.99	1	0.99	0.99	0.99	1	1	0.99	1	1	0.99	1
GCA_0028811 35.1_Burkhold eria_pseudomal lei_MSHR952_ Contig_ST114	1	0.99	0.99	1	1	1	0.99	0.99	1	0.99	1	0.99	1	0.95	1	1
GCA_0019779 45.1_Burkhold eria_pseudomal lei_INT2_BP21 7_Contig_ST58	1	1	1	1	1	0.99	1	0.99	1	1	1	1	1	1	1	1
GCA_0019767 85.1_Burkhold eria_pseudomal lei_3997a_Cont ig_STnovel	1	0.99	0.99	1	1	0.99	0.99	0.99	1	1	1	0.99	1	1	1	1
GCA_0019767 35.1_Burkhold eria_pseudomal lei_4191a_Cont ig_ST209	1	1	1	1	1	1	1	1	1	1	1	0.99	1	1	1	1
GCA_0019768 05.1_Burkhold eria_pseudomal	1	1	1	0.99	1	1	1	0.99	1	1	1	0.99	1	1	0.99	1

lei_RF6_BP26 Contig_ST178																
GCA_0007732 95.1_Burkhold eria_pseudomal lei_MSHR4868 _Contig_ST102 6	1	0.99	1	1	1	0.99	0.99	1	1	0.99	1	0.99	1	0.95	1	1
GCA_0021111 85.1_Burkhold eria_pseudomal lei_201374677 6_Chromosome ST297	1	1	1	1	1	0.99	1	1	1	1	1	0.99	1	0.95	0.99	1
GCA_0021109 85.1_Burkhold eria_pseudomal lei_201175629 6_Chromosome ST297	1	0.99	1	1	1	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0019767 25.1_Burkhold eria_pseudomal lei_4187a_Cont ig_ST17	1	1	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
GCA_0007560 85.1_Burkhold eria_pseudomal lei_BGR_Com plete_Genome_ ST102	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
GCA_0019794 05.1_Burkhold eria_pseudomal lei_MSHR1888 Contig_ST245	1	0.99	1	1	1	1	0.99	0.99	0.99	1	1	0.99	1	0.99	0.99	1
GCA_0007561 85.1_Burkhold eria_pseudomal lei_576_Comp	1	1	0.99	1	1	1	1	0.99	1	0.99	1	0.99	1	0.95	1	1

ete_Genome_S T501																	
GCA_0019801 45.1_Burkhold eria_pseudomal lei_MSHR3499 Contig_ST862	1	0.99	0.99	1	1	1	0.99	1	1	1	1	0.99	1	0.97	0.99	1	
GCA_0019789 25.1_Burkhold eria_pseudomal lei_NAU35A_2 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1	
GCA_0028883 65.1_Burkhold eria_pseudomal lei_INT2_Bp18 4 Contig_ST93	1	1	0.99	1	1	1	0.99	1	1	1	1	1	1	1	1	1	
GCA_0028880 15.1_Burkhold eria_pseudomal lei_MSHR491_ Contig_ST126	1	0.99	0.99	0.99	0.99	1	0.99	0.99	0.81	1	1	0.99	1	1	0.99	1	
GCA_0007559 05.1_Burkhold eria_pseudomal lei_HBPUB103 03a_Complete_ Genome_ST48	1	1	0.99	1	1	1	1	1	1	1	1	0.71	1	1	0.99	1	
GCA_0016370 05.1_Burkhold eria_pseudomal lei_NAU33A_5 _Contig_ST100 5	0.99	1	1	1	1	0.99	1	1	0.99	1	1	0.71	1	1	0.99	1	
GCA_0019784 85.1_Burkhold eria_pseudomal lei_NAU14A_1 0_Contig_ST32 6	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1	

GCA_0029210 15.1_Burkholderia_pseudomallei_UMC108_Contig_ST1342	1	1	1	0.99	1	1	0.99	1	1	1	1	0.99	1	0.95	0.99	1
GCA_0019772 65.1_Burkholderia_pseudomallei_Ubon_P23_Bp21_Contig_ST208	1	1	1	1	1	1	1	0.99	1	1	1	1	1	1	1	1
GCA_0000159 25.1_Burkholderia_pseudomallei_I106a_Complete_Genome_ST70	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.71	1	1	0.99	1
GCA_0019776 55.1_Burkholderia_pseudomallei_RF68_BP5_Contig_STnovel	0.99	1	1	1	0.99	1	0.99	0.99	0.99	1	1	0.71	1	1	0.99	1
GCA_0019772 85.1_Burkholderia_pseudomallei_Ubon_P23_Bp20_Contig_ST208	1	1	1	1	1	1	1	0.99	1	1	1	1	1	1	1	1
GCA_0019779 15.1_Burkholderia_pseudomallei_INT2_BP214_Contig_ST58	1	1	1	1	1	0.99	1	0.99	1	1	1	1	1	1	1	1
GCA_0019774 35.1_Burkholderia_pseudomallei_RF44_BP52_Contig_ST498	1	1	1	1	1	1	0.99	1	0.99	1	1	1	1	1	0.99	1

GCA_0028809 65.1_Burkhold eria_pseudomal lei_DL34_Cont ig_ST197	1	1	1	1	1	1	1	1	1	1	1	1	0.71	1	1	1	1
GCA_0019761 65.1_Burkhold eria_pseudomal lei_200272173 8_Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	1	1	1	0.71	1	1	0.99	1
GCA_0009320 85.1_Burkhold eria_pseudomal lei_QCMRI_B P13_Contig_S T1041	1	0.99	1	1	1	0.99	0.99	0.99	0.99	0.99	1	1	1	1	1	0.99	1
GCA_0004948 55.1_Burkhold eria_pseudomal lei_NCTC_131 79_Complete_ Genome_ST61 3	1	1	1	1	0.99	0.99	0.98	0.99	0.99	1	1	0.99	1	0.99	0.99	0.99	1
GCA_0019796 95.1_Burkhold eria_pseudomal lei_MSHR1683 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	0.99	1
GCA_0019800 65.1_Burkhold eria_pseudomal lei_MSHR0296 Contig_ST260	1	0.99	1	1	1	1	0.99	0.99	0.99	1	1	0.99	1	0.95	1	1	1
GCA_0019768 65.1_Burkhold eria_pseudomal lei_RF8_BP2_ Contig_ST227	1	1	1	1	1	1	0.99	1	0.99	1	1	0.71	1	0.95	0.99	0.99	1
GCA_0009592 65.1_Burkhold	1	1	0.99	1	0.99	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	0.99	1

3_Contig_STtruncated																
GCA_0005119 15.1_Burkholderia_pseudomallei_NAU20B_16_Complete_Genome_ST617	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0006483 55.1_Burkholderia_pseudomallei_4900CFPatient1_Contig_ST92	1	1	1	1	1	0.99	1	0.99	0.97	1	1	0.99	1	0.98	0.99	1
GCA_0021112 85.1_Burkholderia_pseudomallei_3000047530_Complete_Genome_ST92	1	0.99	1	1	1	0.99	1	1	1	1	1	0.99	1	0.98	0.99	1
GCA_0019747 45.1_Burkholderia_pseudomallei_MSHR6522_Chromosome_ST437	1	1	0.99	0.99	0.99	1	1	0.99	1	0.99	1	0.99	1	1	0.99	1
GCA_0019761 95.1_Burkholderia_pseudomallei_2002721183_Contig_ST11	1	1	0.99	1	0.99	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0019797 65.1_Burkholderia_pseudomallei_MSHR2845_Contig_ST243	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_0007750 85.1_Burkholderia_pseudomallei	1	1	1	1	0.99	1	0.99	1	1	0.99	1	0.99	1	0.94	0.99	1

lei_TSV28_Contig_ST815																
GCA_0007733 45.1_Burkholderia_pseudomallei_MSHR3997_Contig_STnovel	1	1	1	1	1	1	0.99	0.99	0.99	0.99	0.98	0.99	1	1	1	1
GCA_0019806 55.1_Burkholderia_pseudomallei_MSHR5104_Contig_ST881	1	0.99	1	0.99	0.99	0.99	0.99	0.99	1	1	1	0.99	1	1	1	1
GCA_0004530 05.1_Burkholderia_pseudomallei_HBPUB10303a_Contig_ST48	1	1	0.99	1	1	1	1	1	1	1	1	0.7	1	1	0.99	1
GCA_0019791 65.1_Burkholderia_pseudomallei_MSHR2053_Contig_ST1356	1	0.99	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	1	1
GCA_0019799 95.1_Burkholderia_pseudomallei_MSHR1079_Contig_ST111	1	0.99	1	1	0.99	0.99	1	1	1	1	1	1	1	0.95	0.99	1
GCA_0019779 55.1_Burkholderia_pseudomallei_INT2_BP241_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019792 15.1_Burkholderia_pseudomallei	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1

lei_MSHR1218 Contig_ST131																	
GCA_0019797 45.1_Burkhold eria_pseudomal lei_MSHR1685 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1	
GCA_0006483 35.1_Burkhold eria_pseudomal lei_PB0829801 0_Contig_ST42 6	1	1	0.99	1	1	1	0.99	0.99	0.98	1	1	0.99	1	0.95	0.99	1	
GCA_0014460 75.1_Burkhold eria_pseudomal lei_NCTC_131 79_Contig_ST6 13	1	1	1	1	0.99	0.99	0.98	0.99	0.99	1	1	0.99	1	0.99	0.99	1	
GCA_0019807 75.1_Burkhold eria_pseudomal lei_402a_Conti g_ST60	0.99	1	1	1	0.99	0.99	1	0.99	1	1	1	0.71	1	1	1	1	
GCA_0019768 95.1_Burkhold eria_pseudomal lei_RF49_BP2 Contig_ST526	0.99	1	0.99	1	0.99	1	1	1	1	0.78	1	0.99	1	1	0.99	1	
GCA_0019795 65.1_Burkhold eria_pseudomal lei_MSHR1670 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1	
GCA_0019801 05.1_Burkhold eria_pseudomal lei_MSHR0911 Contig_ST112	1	0.99	1	0.99	0.99	1	0.99	1	0.99	1	1	0.99	1	1	0.99	1	

GCA_0021110 25.1_Burkhold eria_pseudomal lei_201383305 5_Complete_G enome_ST297	1	0.99	1	1	1	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0028880 45.1_Burkhold eria_pseudomal lei_MSHR511_ Contig_ST617	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0021110 85.1_Burkhold eria_pseudomal lei_200272118 4_Complete_G enome_ST11	1	1	0.99	1	0.99	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0019785 15.1_Burkhold eria_pseudomal lei_NAU14B_1 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019798 35.1_Burkhold eria_pseudomal lei_MSHR3106 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0007558 25.1_Burkhold eria_pseudomal lei_BSR_Comp lete_Genome_S T211	1	1	0.99	1	0.99	0.99	0.99	1	0.99	1	1	0.7	1	1	0.99	1
GCA_0004546 85.1_Burkhold eria_pseudomal lei_NCTC_133 92_morphotype _9_Contig_ST2 3	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.99	1	0.95	0.96	1

GCA_0007733 55.1_Burkholderia_pseudomallei_MSHR7498 Contig_ST766	1	0.99	0.99	1	1	0.99	0.99	1	1	1	1	0.99	1	0.95	0.99	1
GCA_0019764 05.1_Burkholderia_pseudomallei_2002721785 Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1
GCA_0019785 65.1_Burkholderia_pseudomallei_NAU14B_3 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019770 45.1_Burkholderia_pseudomallei_INT2_BP61 Contig_ST93	1	1	0.99	1	1	1	0.99	1	0.75	1	1	1	1	1	1	1
GCA_0019778 65.1_Burkholderia_pseudomallei_INT2_BP127 Contig_ST93	1	1	0.99	1	1	1	0.99	1	1	1	1	1	1	1	1	1
GCA_0019782 45.1_Burkholderia_pseudomallei_NAU2B_1 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019795 95.1_Burkholderia_pseudomallei_MSHR1677 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0014460 35.1_Burkholderia_pseudomallei_NAU44A6 Contig_ST266	1	1	0.99	0.99	1	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1

Bp11_Contig_ST177																
GCA_0018851 95.1_Burkholderia_pseudomallei_VB976100_Complete_Genome_ST436	1	0.99	0.99	1	0.99	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0019789 05.1_Burkholderia_pseudomallei_NAU33A_6_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0009591 45.1_Burkholderia_pseudomallei_406e_Complete_Genome_ST211	1	1	0.99	1	0.99	0.99	0.99	1	0.99	1	1	0.7	1	1	0.99	1
GCA_0004529 45.1_Burkholderia_pseudomallei_MSHR5858_Contig_ST562	0.99	1	0.99	1	1	0.99	0.99	1	1	1	1	0.7	1	0.95	1	1
GCA_0014459 85.1_Burkholderia_pseudomallei_NAU20B16_Contig_ST617	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019802 05.1_Burkholderia_pseudomallei_MSHR3876_Contig_ST842	1	1	1	1	1	0.99	1	1	1	0.99	1	1	1	0.94	0.99	1
GCA_0019797 25.1_Burkholderia_pseudomallei_MSHR1684_Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1

GCA_0028878 55.1_Burkholderia_pseudomallei_MSHR503_Contig_ST149	1	1	1	1	1	1	0.99	1	0.99	0.99	1	1	1	0.95	0.99	1
GCA_0014460 05.1_Burkholderia_pseudomallei_NAU24B3_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019764 95.1_Burkholderia_pseudomallei_4032a_Contig_ST177	0.99	1	1	1	1	1	1	1	1	1	1	0.7	1	1	1	1
GCA_0004545 85.1_Burkholderia_pseudomallei_NCTC_13392_morphotype_6_Contig_ST23	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.99	1	0.95	0.96	1
GCA_0004529 65.1_Burkholderia_pseudomallei_MSHR5848_Contig_ST553	1	0.99	0.99	1	0.99	0.99	0.99	0.99	1	1	1	0.99	1	0.95	1	1
GCA_0019772 45.1_Burkholderia_pseudomallei_Ubon_P23_Bp05_Contig_ST670	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1
GCA_0028809 55.1_Burkholderia_pseudomallei_MSHR87_Contig_ST131	1	0.68	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0	0	0.12
GCA_0021111 05.1_Burkhold	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.98	1	0.95	0.99	1

eria_pseudomalei_2002734728_Complete_Genome_ST518																
GCA_001976485.1_Burkholderia_pseudomalei_4069a_Contig_ST48	1	1	0.99	1	1	0.99	1	1	1	1	1	0.71	1	1	1	1
GCA_000959225.1_Burkholderia_pseudomalei_MSHR2543_Complete_Genome_ST573	1	0.99	1	0.99	1	0.99	0.99	1	1	0.99	1	1	1	0.95	1	1
GCA_001887575.1_Burkholderia_pseudomalei_Burk178_Type2_Complete_Genome_ST553	1	0.99	0.99	1	0.99	0.99	0.99	0.99	1	1	1	0.99	1	0.95	1	1
GCA_001977325.1_Burkholderia_pseudomalei_MSHR0730_Contig_ST103	1	0.99	1	0.99	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	1	1
GCA_001977015.1_Burkholderia_pseudomalei_INT2_BP38_Contig_ST93	1	1	0.99	1	1	1	0.99	1	1	1	1	1	1	1	1	1
GCA_001978525.1_Burkholderia_pseudomalei_NAU14B_2_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_000454565.1_Burkholderia_pseudomalei	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.99	1	0.95	0.96	1

lei_NCTC_13392_morphotype_1_Contig_ST23																	
GCA_001978045.1_Burkholderia_pseudomallei_Ubon_P19_Bp34_Contig_ST670	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1	
GCA_002900645.1_Burkholderia_pseudomallei_VL_Contig_ST46	0.99	1	0.99	1	1	0.99	1	1	1	0.99	1	0.7	1	1	0.99	1	
GCA_000954175.1_Burkholderia_pseudomallei_vgh07_Complete_Genome_ST58	1	1	1	1	1	0.99	1	0.99	1	0.99	1	1	1	1	1	1	
GCA_000445385.1_Burkholderia_pseudomallei_MSHR338_Contig_ST243	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1	
GCA_001979645.1_Burkholderia_pseudomallei_MSHR1678_Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1	
GCA_001979945.1_Burkholderia_pseudomallei_MSHR0295_Contig_ST617	0.87	1	1	1	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1	
GCA_001976645.1_Burkholderia_pseudomallei	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.71	1	1	0.99	1	

lei_NAU2B_9_Contig_ST326																
GCA_0007730 75.1_Burkholderia_pseudomallei_MSHR7504_Contig_ST1032	0.99	0.94	1	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	0.95	0.99	1
GCA_0019803 15.1_Burkholderia_pseudomallei_MSHR4504_Contig_ST1022	1	0.99	1	0.99	0.99	1	0.99	0.99	1	1	1	0.99	1	0.99	1	1
GCA_0005838 35.1_Burkholderia_pseudomallei_MSHR520_Complete_Genome_ST36	0.99	0.99	1	1	1	0.99	0.99	1	0.98	1	1	0.99	1	0.99	0.99	1
GCA_0019802 15.1_Burkholderia_pseudomallei_MSHR3998_Contig_ST848	1	1	1	1	1	1	0.99	0.99	0.99	1	1	0.99	1	1	1	1
GCA_0019774 95.1_Burkholderia_pseudomallei_NRF57_BP22_Contig_ST670	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1
GCA_0019795 05.1_Burkholderia_pseudomallei_MSHR1660_Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0007561 05.1_Burkholderia_pseudomallei_MSHR346	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1

Chromosome_ ST243																
GCA_0019807 35.1_Burkhold eria_pseudomal lei_5598ci_Con tig ST78	1	1	0.99	1	0.99	1	1	0.99	1	1	1	0.99	1	1	0.99	1
GCA_0029006 05.1_Burkhold eria_pseudomal lei_TOML_Co ntig ST369	0.99	1	0.99	0.99	0.99	1	0.99	0.99	0.99	1	1	0.71	1	1	1	1
GCA_0019774 25.1_Burkhold eria_pseudomal lei_RF4_BP80 Contig ST392	1	1	0.99	1	1	0.99	0.99	1	1	0.99	1	0.99	1	1	0.99	1
GCA_0019773 45.1_Burkhold eria_pseudomal lei_MSHR0644 Contig ST122	1	0.99	1	0.99	0.99	1	0.99	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019780 55.1_Burkhold eria_pseudomal lei_Ubon_P23_ Bp38_Contig_ ST208	1	1	1	1	1	1	1	0.99	1	1	1	1	1	1	1	1
GCA_0019796 75.1_Burkhold eria_pseudomal lei_MSHR1682 Contig ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019774 65.1_Burkhold eria_pseudomal lei_RF49_BP5 Contig ST309	1	1	1	0.99	1	0.99	1	1	1	1	1	1	1	1	0.99	1
GCA_0019778 15.1_Burkhold	0.99	1	1	1	0.99	0.99	1	0.99	1	1	1	0.7	1	1	1	1

eria_pseudomalei_INT2_BP105_Contig_ST60																
GCA_001975105.1_Burkholderia_pseudomalei_MSHR5864_Complete_Genome_ST975	0.99	1	1	1	1	1	1	1	1	1	1	0.99	1	0.99	0.99	1
GCA_002860065.1_Burkholderia_pseudomalei_14M0960418_Complete_Genome_ST70	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.7	1	1	0.99	1
GCA_001978325.1_Burkholderia_pseudomalei_NAU2B_11_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_000770455.1_Burkholderia_pseudomalei_B03_Complete_Genome_ST667	1	1	0.99	1	1	1	0.99	0.99	0.99	1	1	0.99	1	1	0.99	1
GCA_000439695.1_Burkholderia_pseudomalei_MSHR305_Complete_Genome_ST36	0.99	0.99	1	1	1	0.99	0.99	1	1	1	1	0.99	1	0.99	0.99	1
GCA_001186165.1_Burkholderia_pseudomalei_MSHR0169_Scaffold_ST284	1	1	0.99	1	1	1	0.99	0.99	1	0.99	1	0.99	1	0.99	1	1
GCA_001976675.1_Burkhold	1	0.92	1	1	1	0.99	0.99	1	1	0.77	1	1	1	0.99	0.99	1

eria_pseudomalei_4263b_Contig_STnovel																
GCA_0019790_85.1_Burkholderia_pseudomalei_MSHR0099_Contig_ST135	1	0.99	1	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	0.95	0.99	1
GCA_0019796_15.1_Burkholderia_pseudomalei_MSHR1676_Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0014461_25.1_Burkholderia_pseudomalei_RNS3Bp1_Contig_ST93	1	1	0.99	1	1	1	0.99	1	1	1	1	1	1	1	1	1
GCA_0019811_85.1_Burkholderia_pseudomalei_4226b_Contig_ST306	1	1	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
GCA_0029006_25.1_Burkholderia_pseudomalei_TOMS_Contig_ST369	0.99	1	0.99	0.99	0.99	1	0.99	0.99	0.99	1	1	0.7	1	1	1	1
GCA_0007732_55.1_Burkholderia_pseudomalei_MSHR5492_Contig_ST862	1	0.99	0.99	1	1	1	0.99	1	1	1	1	0.99	1	0.97	0.99	1
GCA_0019810_55.1_Burkholderia_pseudomalei_2381c_Contig_ST4	0.99	1	1	1	1	0.99	1	1	1	1	1	1	1	1	0.99	1
GCA_0019769_25.1_Burkhold	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.71	1	1	0.99	1

eria_pseudomallei_NRF57_BP67_Contig_ST70																	
GCA_0019790_15.1_Burkholderia_pseudomallei_NAU35B_4_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1	
GCA_0019776_05.1_Burkholderia_pseudomallei_RF63_BP1_Contig_ST17	1	1	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1	
GCA_0019804_95.1_Burkholderia_pseudomallei_MSHR1141_Contig_ST333	1	1	0.99	0.99	1	0.99	0.99	1	0.99	0.99	1	0.99	1	0.95	0.99	1	
GCA_0018875_55.1_Burkholderia_pseudomallei_Burk178_Type1_Complete_Genome_ST553	1	0.99	0.99	1	0.99	0.99	0.99	0.99	1	0.99	1	0.99	1	0.95	1	1	
GCA_0007734_15.1_Burkholderia_pseudomallei_MSHR7500_Contig_ST1031	1	0.91	1	1	1	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1	
GCA_0009530_95.1_Burkholderia_pseudomallei_3921_Complete_Genome_ST670	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1	
GCA_0019781_65.1_Burkhold	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1	

eria_pseudomalei_MSHR0487_Contig_ST243																
GCA_002887875.1_Burkholderia_pseudomalei_MSHR487_Contig_ST243	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_001975065.1_Burkholderia_pseudomalei_MSHR7929_Complete_Genome_ST437	1	1	0.99	0.99	0.99	1	1	0.99	1	0.99	1	0.99	1	1	0.99	1
GCA_001980915.1_Burkholderia_pseudomalei_975d_Contig_ST873	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.71	1	1	0.99	1
GCA_001978985.1_Burkholderia_pseudomalei_NAU35B_3_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_001977615.1_Burkholderia_pseudomalei_RF63_BP2_Contig_ST177	0.99	1	1	1	1	0	0	0	1	1	1	1	1	1	0.99	1
GCA_002880985.1_Burkholderia_pseudomalei_DL17_Contig_ST48	1	1	0.99	1	1	0.99	1	1	1	1	1	0.7	1	1	1	1
GCA_001446175.1_Burkholderia_pseudomalei_Songkhla34W2_Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1

GCA_0021113 65.1_Burkholderia_pseudomallei_2008724758_Complete_Genome_ST550	1	1	0.99	0.99	1	1	1	0.99	1	0.99	1	0.99	1	1	0.99	1
GCA_0013182 45.1_Burkholderia_pseudomallei_Bp1651_Complete_Genome_ST880	1	0.99	1	0.99	0.99	1	0.82	0.99	1	0.99	1	0.99	1	1	0.99	1
GCA_0019781 25.1_Burkholderia_pseudomallei_Ubon_P44_Bp34_Contig_ST177	0.99	1	1	1	1	1	0.99	0.99	1	1	1	1	1	1	0.99	1
GCA_0028809 15.1_Burkholderia_pseudomallei_DL2_Contig_ST225	1	1	1	1	1	0.99	1	1	1	1	1	1	1	1	1	1
GCA_0019809 05.1_Burkholderia_pseudomallei_979bi_Contig_ST60	0.99	1	1	1	0.99	0.99	1	0.99	1	1	1	0.71	1	1	1	1
GCA_0002597 75.1_Burkholderia_pseudomallei_1258b_Contig_ST221	1	0.58	1	1	1	1	0.99	1	1	1	1	0.99	1	1	1	1
GCA_0013204 25.1_Burkholderia_pseudomallei_AH3_Scaffold_ST51	0.99	1	0.99	0.99	0.99	1	0.99	0.99	0.99	1	1	0.71	1	1	0.99	1
GCA_0019762 45.1_Burkhold	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1

GCA_0012778 75.1_Burkholderia_pseudomallei_vgh16R_Complete_Genome_ST1001	1	0.99	0.99	1	1	0.99	1	0.99	0.99	1	1	0.99	1	1	0.99	1
GCA_0019751 25.1_Burkholderia_pseudomallei_MSHR4083_Complete_Genome_ST36	0.99	0.99	1	1	1	0.99	0.99	1	1	1	1	0.99	1	0.99	0.99	1
GCA_0019769 05.1_Burkholderia_pseudomallei_RF43_BP22_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0002597 95.1_Burkholderia_pseudomallei_354e_Contig_ST78	1	0.57	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
GCA_0009591 85.1_Burkholderia_pseudomallei_MSHR840_Complete_Genome_ST257	1	0.99	0.99	0.99	1	1	0.99	1	0.99	0.99	1	0.99	1	0.95	1	1
GCA_0019784 05.1_Burkholderia_pseudomallei_NAU14A_1_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0004546 45.1_Burkholderia_pseudomallei_NCTC_13392_morphotype	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.99	1	0.95	0.96	1

_5_Contig_ST23																
GCA_0019806 45.1_Burkholderia_pseudomallei_MSHR5100_Contig_ST658	0.99	1	0.99	0.99	0.99	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1
GCA_0019794 35.1_Burkholderia_pseudomallei_GU_1909A_Contig_STtruncated	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.7	1	1	0.99	1
GCA_0028878 35.1_Burkholderia_pseudomallei_MSHR296_Contig_ST260	1	0.99	1	1	1	1	0.99	0.99	0.99	1	1	0.99	1	0.95	1	1
GCA_0019799 05.1_Burkholderia_pseudomallei_MSHR0200_Contig_ST617	1	0.67	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019787 45.1_Burkholderia_pseudomallei_NAU21B_4_Contig_ST617	1	1	1	1	0.99	0.99	0.99	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0016115 85.1_Burkholderia_pseudomallei_PtBps01_Scaffold_ST376	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.7	1	1	0.99	1
GCA_0021113 85.1_Burkholderia_pseudomallei_3000015486_Complete_Genome_ST92	1	1	0.99	1	0.77	0.99	1	0.99	1	1	1	0.98	1	0.95	0.99	1

GCA_0019793 85.1_Burkhold eria_pseudomal lei_MSHR1088 Contig_ST259	1	1	1	1	1	0.99	0.98	1	0.99	0.86	1	1	1	0.94	1	1
GCA_0019782 65.1_Burkhold eria_pseudomal lei_NAU2B_6_ Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0007730 85.1_Burkhold eria_pseudomal lei_MSHR5608 _Contig_ST102 8	1	1	1	1	1	1	0.99	0.99	1	1	1	0.99	1	0.99	0.99	1
GCA_0019783 85.1_Burkhold eria_pseudomal lei_NAU2B_12 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019793 25.1_Burkhold eria_pseudomal lei_MSHR1300 Contig_ST333	1	1	0.99	0.99	1	0.99	0.99	1	0.99	0.99	1	0.99	1	0.95	0.99	1
GCA_0019763 45.1_Burkhold eria_pseudomal lei_3943a_Cont ig_ST300	1	1	1	1	1	0.99	0.99	1	1	1	1	0.99	1	1	0.99	1
GCA_0019786 65.1_Burkhold eria_pseudomal lei_NAU14B_1 0_Contig_ST32 6	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019791 35.1_Burkhold eria_pseudomal	1	0.99	0.99	1	1	1	0.99	0.99	1	0.99	1	0.99	1	0.95	1	1

lei_MSHR0952 Contig_ST114																
GCA_0019798 85.1_Burkhold eria_pseudomal lei_MSHR1289 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0007559 65.1_Burkhold eria_pseudomal lei_MSHR5848 _Complete_Ge nome_ST553	1	0.99	0.99	1	0.99	0.99	0.99	0.99	1	1	1	0.99	1	0.95	1	1
GCA_0007732 15.1_Burkhold eria_pseudomal lei_MSHR5569 Contig_ST885	1	1	0.99	0.99	1	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_0019806 95.1_Burkhold eria_pseudomal lei_5598b_Con tig_ST78	1	0.71	0.99	1	0.99	1	1	0.99	1	1	1	0.99	1	1	0.99	1
GCA_0012778 95.1_Burkhold eria_pseudomal lei_vgh16W_C omplete_Geno me_ST1001	1	0.99	0.99	1	1	0.99	1	0.99	0.99	1	1	0.99	1	1	0.99	1
GCA_0021113 05.1_Burkhold eria_pseudomal lei_300046597 2_Complete_G enome_ST436	1	0.99	0.99	1	0.99	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0028811 15.1_Burkhold eria_pseudomal lei_INT2_Bp24 _Contig_ST58	1	1	1	1	1	0.99	1	0.99	0.93	1	1	1	1	1	1	1

GCA_0007645 75.1_Burkhold eria_pseudomal lei_NAU35A_3 _Complete_Ge nome_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019809 85.1_Burkhold eria_pseudomal lei_979bii_Con tig_ST60	0.99	1	1	1	0.99	0.99	1	0.99	1	1	1	0.71	1	1	1	1
GCA_0001697 15.1_Burkhold eria_pseudomal lei_305_Contig ST36	0.99	0.99	1	1	1	0.99	0.99	1	1	1	1	0.99	1	0.99	0.99	1
GCA_0019750 85.1_Burkhold eria_pseudomal lei_MSHR6755 _Complete_Ge nome_ST975	0.99	1	1	1	1	1	1	1	1	1	1	0.99	1	0.99	0.99	1
GCA_0021139 45.1_Burkhold eria_pseudomal lei_201400281 6_Contig_ST10 53	1	0.99	0.99	1	1	0.99	1	0.99	1	1	1	0.98	1	0.99	0.99	1
GCA_0028881 35.1_Burkhold eria_pseudomal lei_INT4_Bp18 Contig_ST60	0.99	1	1	1	0.99	0.99	1	0.99	1	1	1	0.71	1	1	1	1
GCA_0021111 45.1_Burkhold eria_pseudomal lei_201000750 9_Complete_G enome_ST518	1	0.99	1	1	0.99	0.99	1	0.99	1	1	1	0.97	1	1	0.99	1

GCA_0007570 15.2_Burkholderia_pseudomallei_PHL5_112_Complete_Genome_ST15	1	1	1	1	1	1	0.99	1	0.99	1	1	1	1	1	0.99	1
GCA_0019762 55.1_Burkholderia_pseudomallei_2002721772_Contig_ST418	1	1	0.99	1	1	0.99	1	1	0.99	1	1	0.71	1	0.95	1	1
GCA_0021110 45.1_Burkholderia_pseudomallei_3000015237_Complete_Genome_ST951	1	0.99	1	1	1	0.99	1	0.99	1	1	1	0.98	1	0.95	0.99	1
GCA_0019766 85.1_Burkholderia_pseudomallei_4175a_Contig_ST197	1	1	1	1	1	1	1	1	1	1	1	0.71	1	1	1	1
GCA_0019781 05.1_Burkholderia_pseudomallei_Ubon_P44_Bp26_Contig_ST177	0.99	1	1	1	1	1	0.99	0.99	1	1	1	1	1	1	0.99	1
GCA_0005006 55.1_Burkholderia_pseudomallei_MSHR6137_Contig_ST325	1	1	1	1	0.99	0.99	1	0.99	1	0.99	1	0.99	1	0.95	0.99	1
GCA_0019794 85.1_Burkholderia_pseudomallei_MSHR0391_Contig_ST243	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1

GCA_0014458 65.1_Burkhold eria_pseudomal lei_MSHR1950 _Contig_ST107 4	1	1	1	0.99	0.99	0.99	0.99	0.56	0.99	0.99	1	0.99	0.99	0.99	1	1
GCA_0019784 55.1_Burkhold eria_pseudomal lei_NAU14A_5 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019802 75.1_Burkhold eria_pseudomal lei_MSHR4301 _Contig_ST104 4	1	1	1	1	0.99	0.99	1	0.99	0.99	0.99	1	0.99	1	0.95	0.99	1
GCA_0019784 15.1_Burkhold eria_pseudomal lei_NAU14A_2 Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0007756 75.1_Burkhold eria_pseudomal lei_A79C_Cont ig_ST667	1	1	0.99	1	1	1	0.99	0.99	0.99	1	1	0.99	1	1	0.99	1
GCA_0021113 45.1_Burkhold eria_pseudomal lei_200272117 1_Complete_G enome_ST12	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0019771 65.1_Burkhold eria_pseudomal lei_RNS8_BP1 _Contig_STnov el	1	1	0.99	0.99	1	0.99	0.99	1	0.86	1	1	1	1	1	0.99	1

GCA_0019763 35.1_Burkholderia_pseudomallei_4151a_Contig_ST288	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.95	0.99	1
GCA_0034454 15.1_Burkholderia_pseudomallei_9_Complete_Genome_ST72	0.99	1	1	1	1	0.99	1	1	1	1	1	0.99	1	0.95	1	1
GCA_0019809 95.1_Burkholderia_pseudomallei_2374a_Contig_ST174	0.99	1	0.99	1	1	0.99	1	1	1	1	1	1	1	0.95	1	1
GCA_0019789 65.1_Burkholderia_pseudomallei_NAU35B_2_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.89	1	0.99	0.99	1
GCA_0019798 65.1_Burkholderia_pseudomallei_MSHR3030_Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019805 85.1_Burkholderia_pseudomallei_MSHR5089_Contig_ST881	1	0.99	1	0.99	0.99	0.99	0.99	0.99	1	1	1	0.99	1	1	1	1
GCA_0019777 25.1_Burkholderia_pseudomallei_RF85_BP37_Contig_ST70	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.7	1	1	0.99	1
GCA_0029006 65.1_Burkholderia_pseudomallei	0.99	1	0.99	1	1	0.99	1	1	1	0.99	1	0.71	1	1	0.99	1

GCA_0029209 45.1_Burkholderia_pseudomallei_UM137_Contig_ST1342	1	1	1	0.99	1	1	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_0019779 75.1_Burkholderia_pseudomallei_INT2_BP235_Contig_ST1005	0.99	1	1	1	1	0.99	1	1	0.99	1	1	0.7	1	1	0.99	1
GCA_0019789 35.1_Burkholderia_pseudomallei_NAU35A_4Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0007750 95.1_Burkholderia_pseudomallei_TSV5_Contig_ST814	1	1	1	1	1	1	0.99	1	0.99	1	1	0.99	1	0.94	0.99	1
GCA_0019803 65.1_Burkholderia_pseudomallei_MSHR4638Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0004646 85.1_Burkholderia_pseudomallei_NCTC_13392_morphotype_10_Contig_ST23	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.99	1	0.95	0.96	1
GCA_0007741 85.1_Burkholderia_pseudomallei_MSHR4304_Scaffold_ST995	1	1	1	0.99	1	1	0.99	0.99	0.99	1	1	1	1	0.95	0.99	1

GCA_0009591 25.1_Burkholderia_pseudomallei_1026b_Complete_Genome_ST102	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
GCA_0019788 25.1_Burkholderia_pseudomallei_NAU22A_3_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0028810 75.1_Burkholderia_pseudomallei_Smith_002025_Contig_ST418	1	1	0.99	1	1	0.99	1	1	0.99	1	1	0.71	1	0.95	1	1
GCA_0019795 85.1_Burkholderia_pseudomallei_MSHR1672_Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019786 15.1_Burkholderia_pseudomallei_NAU14B_8_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0007704 95.1_Burkholderia_pseudomallei_TSV_48_Complete_Genome_ST252	1	0.99	1	1	1	0.99	1	1	0.99	0.99	1	1	1	0.95	0.99	1
GCA_0019773 65.1_Burkholderia_pseudomallei_MSHR0503_Contig_ST149	1	1	1	1	1	1	0.99	1	0.99	0.99	1	1	1	0.95	0.99	1
GCA_0019794 55.1_Burkhold	1	1	1	1	1	1	0.99	1	0.99	1	1	1	1	1	0.99	1

eria_pseudomallei_GU_2143A Contig_ST498																
GCA_00292105.1_Burkholderia_pseudomallei_UM136_Contig_ST1342	1	1	1	0.99	1	1	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_001978005.1_Burkholderia_pseudomallei_INT2_BP264_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_000756125.1_Burkholderia_pseudomallei_Mahidol_1106a_Complete_Genome_ST70	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.7	1	1	0.99	1
GCA_000756065.1_Burkholderia_pseudomallei_MSHR5855_Complete_Genome_ST553	1	0.99	0.99	1	0.99	0.99	0.99	0.99	1	1	1	0.99	1	0.95	1	1
GCA_001981025.1_Burkholderia_pseudomallei_2374b_Contig_ST304	1	1	1	1	1	0.99	1	0.99	1	1	1	1	1	1	1	1
GCA_000182195.1_Burkholderia_pseudomallei_576_Contig_ST501	1	1	0.99	1	1	1	1	0.99	1	0.99	1	0.99	1	0.95	1	1
GCA_002888275.1_Burkholderia_pseudomallei_MSHR2053	1	0.99	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	1	1

_Contig_ST1356																
GCA_000755765.1_Burkholderia_pseudomallei_BDP_Complete_Genome_ST36	0.99	0.99	1	1	1	0.99	0.99	1	1	1	1	0.99	1	0.99	0.99	1
GCA_002887885.1_Burkholderia_pseudomallei_MSHR139_Contig_STnovel1	1	0.99	1	1	1	1	0.99	1	1	1	1	0.99	1	0.94	0.99	1
GCA_001979045.1_Burkholderia_pseudomallei_NAU44A_6_Contig_ST266	1	1	0.99	0.99	1	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_001981195.1_Burkholderia_pseudomallei_4226c_Contig_ST306	1	1	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
GCA_000452985.1_Burkholderia_pseudomallei_HBPUB10134a_Contig_ST228	1	1	0.99	1	1	1	0.99	0.99	1	1	1	1	1	1	0.99	1
GCA_001320645.1_Burkholderia_pseudomallei_48_Scaffold_ST423	0.99	1	1	1	1	1	0.99	0.99	0.99	1	1	0.71	1	1	0.99	1
GCA_002888115.1_Burkholderia_pseudomallei_DL28_Contig_ST70	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.71	1	1	0.99	1

GCA_0019794 15.1_Burkholderia_pseudomallei_MSHR1879 Contig_ST332	1	0.99	1	1	1	1	0.99	1	0.99	0.86	1	0.99	1	0.95	0.99	1
GCA_0019767 55.1_Burkholderia_pseudomallei_3944b_Contig_ST9	1	1	0.99	1	0.99	0.99	1	1	1	1	1	0.99	1	1	1	1
GCA_0019771 35.1_Burkholderia_pseudomallei_RNS3_Bp1 Contig_ST93	1	1	0.99	1	1	1	0.99	1	1	1	1	1	1	1	1	1
GCA_0019807 25.1_Burkholderia_pseudomallei_316a_Contig_ST17	1	1	0.99	1	0.99	1	1	1	1	1	1	0.99	1	1	0.99	1
GCA_0014456 35.1_Burkholderia_pseudomallei_RF43Bp22_Contig_ST326	1	1	1	1	1	0.99	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0019801 25.1_Burkholderia_pseudomallei_MSHR2825 Contig_ST642	1	1	1	1	1	1	0.99	0.99	0.99	0.99	1	1	1	0.95	0.99	1
GCA_0019808 05.1_Burkholderia_pseudomallei_577cii_Scaffold_ST708	1	0.59	0.99	1	0.99	0.99	0.99	1	1	1	1	0.71	1	1	0.99	1
GCA_0019789 95.1_Burkholderia_pseudomallei_NAU44A_2_Contig_ST266	1	1	0.99	0.99	1	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1

GCA_0012779 75.1_Burkholderia_pseudomallei_982_Complete_Genome_ST1477	1	1	1	1	1	1	0.99	0.99	1	1	1	1	1	1	0.99	1
GCA_0019761 85.1_Burkholderia_pseudomallei_2002721784_Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1
GCA_0004546 05.1_Burkholderia_pseudomallei_NCTC_13392_morphotype_3_Contig_ST23	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.99	1	0.95	0.96	1
GCA_0021113 25.1_Burkholderia_pseudomallei_2002721123_Complete_Genome_ST92	1	1	1	1	1	0.99	1	1	1	1	1	0.99	1	0.95	0.99	1
GCA_0019811 05.1_Burkholderia_pseudomallei_3013c_Contig_ST874	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.91	1	1	0.99	1
GCA_0019768 25.1_Burkholderia_pseudomallei_RF6_BP15_Contig_ST178	1	1	1	0.99	1	1	1	0.99	1	1	1	0.99	1	1	0.99	1
GCA_0007703 95.1_Burkholderia_pseudomallei_MSHR62_Complete_Genome_ST471	1	1	0.99	1	0.99	1	0.99	0.99	0.99	0.99	1	0.99	1	0.95	0.99	1

GCA_0007731 65.1_Burkholderia_pseudomallei_MSHR5613 Contig_ST866	1	1	1	1	1	1	0.99	0.99	1	0.99	1	1	1	1	0.99	1
GCA_0019787 05.1_Burkholderia_pseudomallei_NAU20B_2 Contig_ST326	1	1	1	1	1	0.87	1	0.99	1	0.99	1	0.99	1	0.99	0.99	1
GCA_0007705 15.1_Burkholderia_pseudomallei_K42_Complete_Genome_ST267	1	1	0.99	1	1	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_0019777 35.1_Burkholderia_pseudomallei_INT2_BP12 Contig_ST58	1	1	1	1	1	0.99	1	0.99	1	1	1	1	1	1	1	1
GCA_0019810 85.1_Burkholderia_pseudomallei_3964b_Contig_ST671	1	0.73	0.99	1	1	0.99	1	1	1	0.86	1	1	1	0.95	0.99	1
GCA_0019798 15.1_Burkholderia_pseudomallei_MSHR3107 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019796 65.1_Burkholderia_pseudomallei_MSHR1679 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019775 65.1_Burkholderia_pseudomallei	1	1	0.99	1	0.99	1	1	1	1	0.99	1	0.99	1	1	0.99	1

lei_RF61_BP2 Contig_ST17																
GCA_0019762 65.1_Burkhold eria_pseudomal lei_200272178 8_Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.7	1	1	0.99	1
GCA_0004546 65.1_Burkhold eria_pseudomal lei_NCTC_133 92_morphotype _7_Contig_ST2 3	1	1	0.99	1	0.99	0.99	0.99	1	1	1	1	0.99	1	0.95	0.96	1
GCA_0001825 85.2_Burkhold eria_pseudomal lei_MSHR346_ Chromosome_ ST243	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	1	0.99	1	0.95	0.99	1
GCA_0019803 35.1_Burkhold eria_pseudomal lei_MSHR4637 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019771 85.1_Burkhold eria_pseudomal lei_Ubon_P19_ Bp57_Contig_ ST670	0.99	1	1	1	1	1	0.99	1	1	1	1	1	1	1	0.99	1
GCA_0021112 65.1_Burkhold eria_pseudomal lei_201383305 7_Complete_G enome_ST297	1	0.99	1	1	1	0.99	1	0.99	1	1	1	0.99	1	0.95	0.99	1
GCA_0019766 25.1_Burkhold eria_pseudomal	1	1	1	1	1	0.99	0.99	0.99	1	1	1	0.7	1	1	0.99	1

lei_4169d_Contig_ST507																	
GCA_001980175.1_Burkholderia_pseudomallei_MSHR3841_Contig_ST335	1	1	0.99	1	1	0.99	0.99	1	0.99	0.99	1	1	1	0.95	1	1	
GCA_000775105.1_Burkholderia_pseudomallei_TSV32_Contig_ST817	1	0.99	1	1	1	1	0.99	1	0.99	1	1	1	1	0.95	0.99	1	
GCA_001979345.1_Burkholderia_pseudomallei_MSHR0087_Contig_ST131	1	0.68	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0	0	0.12	
GCA_002115385.1_Burkholderia_pseudomallei_200872464_Contig_ST698	1	0.99	1	1	1	0.99	1	0.99	1	1	1	0.99	1	0.98	0.99	1	
GCA_001976395.1_Burkholderia_pseudomallei_200272174_1_Contig_ST84	0.99	1	1	1	1	1	0.99	0.99	1	1	1	0.71	1	1	0.99	1	
GCA_001446055.1_Burkholderia_pseudomallei_NCTC_13178_Contig_ST286	1	0.99	1	1	1	1	0.99	0.99	1	0.99	1	1	1	0.95	0.99	1	
GCA_001979245.1_Burkholderia_pseudomallei_MSHR1418_Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1	

GCA_0019792 55.1_Burkhold eria_pseudomal lei_MSHR1821 Contig_ST131	1	0.99	0.99	1	1	0.99	1	1	0.99	1	1	1	1	0.95	0.99	1
GCA_0019770 95.1_Burkhold eria_pseudomal lei_319a_Conti g_ST17	0.99	1	1	1	0.99	0.99	1	1	1	0.98	1	1	1	1	1	1
GCA_0019791 05.1_Burkhold eria_pseudomal lei_MSHR0073 Contig_ST135	1	0.99	1	1	1	1	0.99	0.99	0.99	0.99	1	0.99	1	0.95	0.99	1

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