Isolates previously submitted to pubMLST under a separate isolate ID are identified by that ID within the Alternate pubMLST ID and Notes column. Isolates with an Unknown ST have their missing allele(s) identified in this column, in addition to the five isolates that originate from the USA.

| Sequence Type | Alternate pubMLST ID and Notes | ID      | Year | Feedlot | Anatomical Location | Health Status | Host ID |
|---------------|--------------------------------|---------|------|---------|---------------------|---------------|--------|
| 2             | MJ259                          | MPLM0042| 2016 | N       | joint              | dead          | 16     |
| 2             | MPLM0632                       | 2007    | K    | nasopharynx | dead          | 83     |
| 2             | MPLM0644                       | 2007    | Q    | nasopharynx | diseased      | 42     |
| 14            | USA Isolate                    | MPLM0833| 2017 | E       | nasopharynx       | healthy       | 31     |
| 21            | MJ260                          | MPLM0054| 2017 | O       | lung               | dead          | 70     |
| 21            | MJ287                          | MPLM0154| 2017 | N       | lung               | dead          | 67     |
| 21            | MPLM0645                       | 2007    | Q    | nasopharynx | diseased      | 56     |
| 21            | MPLM0648                       | 2006    | J    | nasopharynx | diseased      | 73     |
| 21            | MPLM0662                       | 2007    | Q    | nasopharynx | diseased      | 51     |
| 21            | MPLM0698                       | 2007    | Q    | nasopharynx | healthy       | 59     |
| 21            | MPLM0706                       | 2007    | Q    | nasopharynx | healthy       | 55     |
| 24            | MPLM0638                       | 2015    | I    | joint    | dead            | 88     |
| 24            | MPLM0647                       | 2007    | Q    | nasopharynx | diseased      | 45     |
| 24            | MPLM0657                       | 2007    | Q    | nasopharynx | diseased      | 46     |
| 24            | MPLM0660                       | 2007    | Q    | nasopharynx | diseased      | 40     |
| 24            | MPLM0661                       | 2007    | Q    | nasopharynx | diseased      | 44     |
| 24            | MPLM0664                       | 2007    | Q    | nasopharynx | diseased      | 39     |
| 24            | MPLM0666                       | 2007    | Q    | nasopharynx | diseased      | 57     |
| 24            | MPLM0669                       | 2007    | Q    | nasopharynx | dead          | 43     |
| 24            | MPLM0684                       | 2007    | Q    | nasopharynx | healthy       | 49     |
| 24            | MPLM0692                       | 2007    | Q    | nasopharynx | healthy       | 58     |
| 24            | MPLM0700                       | 2007    | Q    | nasopharynx | healthy       | 52     |
| 24            | MPLM0714                       | 2007    | Q    | nasopharynx | healthy       | 48     |
| 24            | MPLM0715                       | 2007    | Q    | nasopharynx | healthy       | 54     |
| 27            | MPLM0541                       | 2018    | N    | lung     | dead           | 90     |
| 27            | MPLM0555                       | 2018    | N    | lung     | dead           | 64     |
| 27            | MPLM0556                       | 2018    | N    | joint    | dead           | 64     |
| 40            | MPLM0652                       | 2007    | Q    | nasopharynx | diseased      | 50     |
| ID  | Code         | Year | Sex | Body Part | Condition | Age |
|-----|--------------|------|-----|-----------|-----------|-----|
| 40  | MPLM0665     | 2007 | Q   | nasopharynx | diseased  | 41  |
| 40  | MPLM0667     | 2007 | R   | nasopharynx | healthy   | 78  |
| 40  | MPLM0668     | 2007 | R   | nasopharynx | healthy   | 79  |
| 40  | MPLM0713     | 2007 | Q   | nasopharynx | healthy   | 47  |
| 42  | MPLM0634     | 2015 | M   | lung      | dead      | 65  |
| 42  | MJ292        | 2014 | S   | lung      | dead      | 97  |
| 42  | MPLM0640     | 2006 | G   | nasopharynx | healthy   | 77  |
| 42  | MPLM0642     | 2006 | U   | nasopharynx | diseased  | 71  |
| 42  | MPLM0649     | 2006 | P   | nasopharynx | diseased  | 74  |
| 42  | MPLM0670     | 2007 | R   | nasopharynx | healthy   | 75  |
| 42  | MPLM0671     | 2007 | R   | nasopharynx | healthy   | 76  |
| 43  | MPLM0825     | 2018 | C   | nasopharynx | healthy   | 26  |
| 44  | MPLM0061     | 2017 | O   | joint    | dead      | 69  |
| 44  | MJ267        | 2017 | N   | lung      | dead      | 17  |
| 44  | MJ268        | 2017 | N   | joint    | dead      | 17  |
| 44  | MJ272        | 2017 | N   | joint    | dead      | 9   |
| 44  | MJ273        | 2017 | N   | lung      | dead      | 35  |
| 44  | MJ274        | 2017 | N   | joint    | dead      | 35  |
| 44  | MJ278        | 2017 | N   | lung      | dead      | 8   |
| 44  | MJ286        | 2017 | N   | joint    | dead      | 68  |
| 44  | MJ289        | 2017 | N   | lung      | dead      | 12  |
| 45  | MJ280        | 2017 | S   | lung      | dead      | 4   |
| 45  | MPLM0136     | 2017 | A   | nasopharynx | healthy   | 19  |
| 45  | MPLM0815     | 2017 | D   | nasopharynx | healthy   | 27  |
| 45  | MPLM0827     | 2018 | D   | nasopharynx | healthy   | 28  |
| 45  | MPLM0832     | 2017 | E   | nasopharynx | healthy   | 30  |
| 48  | MPLM0646     | 2007 | R   | nasopharynx | healthy   | 80  |
| 52  | MJ246        | 2016 | T   | joint    | dead      | 89  |
| 52  | MJ255        | 2016 | T   | lung      | dead      | 60  |
| 52  | MJ257        | 2016 | T   | lung      | dead      | 98  |
| 52  | MPLM0020     | 2016 | L   | joint    | dead      | 82  |
| 60  | MJ237        | 2016 | T   | lung      | dead      | 10  |
| 60  | MJ238        | 2016 | T   | joint    | dead      | 10  |
| 60  | MJ240        | 2016 | T   | joint    | dead      | 95  |
| 60  | MJ241        | 2016 | T   | lung      | dead      | 11  |
| 60  | MJ242        | 2016 | T   | joint    | dead      | 11  |
| 60  | MPLM0016     | 2016 | T   | joint    | dead      | 62  |
| 60  | MJ244        | 2016 | T   | lung      | dead      | 61  |
| 60  | MJ245        | 2016 | T   | joint    | dead      | 61  |
| 60  | MJ249        | 2016 | T   | lung      | dead      | 3   |
| 60  | MJ250        | 2016 | T   | joint    | dead      | 3   |
| 60  | MJ253        | 2016 | T   | lung      | dead      | 2   |
|   | Isolate | Species | Year | Tissue | Condition | Age |
|---|---------|---------|------|--------|-----------|-----|
| 60 | MJ258   | MPLM0035| 2016 | T      | lung      | dead 86 |
| 60 | MPLM0041| 2016    | N    | lung   | dead 16   |
| 60 | MPLM0157| 2017    | N    | lung   | dead 87   |
| 60 | MPLM0533| 2018    | N    | lung   | dead 7    |
| 60 | MPLM0534| 2018    | N    | joint  | dead 7    |
| 60 | MPLM0538| 2018    | N    | joint  | dead 14   |
| 60 | MPLM0542| 2018    | N    | joint  | dead 90   |
| 60 | MPLM0545| 2018    | N    | lung   | dead 91   |
| 60 | MPLM0546| 2018    | N    | joint  | dead 91   |
| 60 | MPLM0559| 2018    | N    | lung   | dead 93   |
| 60 | MPLM0560| 2018    | N    | joint  | dead 93   |
| 60 | MJ291   | MPLM0635| 2014 | H      | lung      | dead 96 |
| 61 | MJ243   | MPLM0015| 2016 | T      | lung      | dead 62 |
| 62 | MPLM0703| 2007    | Q    | nasopharynx | healthy | 53   |
| 65 | MJ235   | MPLM0007| 2016 | T      | lung      | dead 63 |
| 65 | MJ236   | MPLM0008| 2016 | T      | joint     | dead 63 |
| 65 | MJ239   | MPLM0011| 2016 | T      | lung      | dead 95 |
| 65 | MJ247   | MPLM0021| 2016 | T      | lung      | dead 6  |
| 65 | MJ248   | MPLM0022| 2016 | T      | joint     | dead 6  |
| 65 | MJ251   | MPLM0029| 2016 | N      | lung      | dead 1  |
| 65 | MPLM0821| 2017    | B    | nasopharynx | healthy | 22   |
| 66 | USA     | MPLM0820| 2017 | A      | nasopharynx | healthy | 21   |
| 66 | Isolate | MPLM0838| 2017 | E      | nasopharynx | healthy | 34   |
| 67 | MJ261   | MPLM0057| 2017 | O      | lung      | dead 18 |
| 67 | MJ262   | MPLM0058| 2017 | O      | joint     | dead 18 |
| 67 | MJ263   | MPLM0060| 2017 | O      | lung      | dead 69 |
| 67 | MJ269   | MPLM0093| 2017 | N      | lung      | dead 13 |
| 67 | MJ271   | MPLM0102| 2017 | N      | lung      | dead 9  |
| 67 | MJ276   | MPLM0111| 2017 | N      | lung      | dead 5  |
| 67 | MJ277   | MPLM0112| 2017 | N      | joint     | dead 5  |
| 67 | MJ282   | MPLM0145| 2017 | N      | lung      | dead 66 |
| 67 | MJ283   | MPLM0146| 2017 | N      | joint     | dead 66 |
| 67 | MPLM0155| 2017    | N    | joint  | dead 67   |
| 67 | MPLM0158| 2017    | N    | joint  | dead 87   |
| 67 | MPLM0164| 2017    | N    | joint  | dead 36   |
| 67 | MPLM0209| 2014    | I    | joint  | dead 85   |
| 67 | MPLM0219| 2014    | I    | lung    | dead 85   |
| 77 | MJ234 | MPLM0003 | 2016 | F | lung | dead | 94 |
| 77 |   | MPLM0004 | 2016 | F | joint | dead | 94 |
| 79 | MJ252 | MPLM0030 | 2016 | N | joint | dead | 1 |
| 80 |   | MPLM0132 | 2016 | S | lung | dead | 38 |
| 80 |   | MPLM0134 | 2016 | S | lung | dead | 15 |
| 80 |   | MPLM0135 | 2016 | S | joint | dead | 15 |
| 149 |   | MPLM0608 | 2018 | N | lung | dead | 92 |
| 150 | USA Isolate | MPLM0830 | 2018 | D | nasopharynx | healthy | 29 |
| 151 |   | MPLM0643 | 2006 | G | nasopharynx | healthy | 72 |
| 152 |   | MPLM0822 | 2017 | B | nasopharynx | healthy | 23 |
| 153 |   | MPLM0823 | 2018 | C | nasopharynx | healthy | 24 |
| 153 |   | MPLM0824 | 2018 | C | nasopharynx | healthy | 25 |
| 154 | USA Isolate | MPLM0834 | 2017 | E | nasopharynx | healthy | 32 |
| 154 | USA Isolate | MPLM0837 | 2017 | E | nasopharynx | healthy | 33 |
| Unknown | dnaA, gltX, gpsA, gyrB, tdk | MPLM0019 | 2016 | T | lung | dead | 89 |
| Unknown | MJ254, Missing gpsA | MPLM0036 | 2016 | T | joint | dead | 86 |
| Unknown | MJ285, Missing gyrB | MPLM0148 | 2017 | N | lung | dead | 68 |
| Unknown | Missing gpsA, Novel pta2 | MPLM0817 | 2018 | A | nasopharynx | healthy | 20 |
Figure S1 Minimum spanning tree of 126 *Mycoplasma bovis* genomes (125 field isolates plus PG45) typed by MLST. Clonal complexes 1 and 2 are surrounded by red and blue lines, respectively. Sequence types are colour coded with the size of the circle reflecting the number of isolates, with the partitioning lines within a circle delineating isolates with an identical genotype. The number of different alleles is indicated over the line connecting the sequence types. The PG45 reference genome is identified as the singleton with ST12.
**Figure S2** Minimum spanning tree of 102 *Mycoplasma bovis* isolates (101 field isolates plus PG45) created with cgMLST, based on alleles at 296 core genome loci. Clades 1 - 3 are denoted by a surrounding circle. The isolates are identified by MLST sequence type and production year (ST, Production Year). Sequence types are colour coded with the size of the circle reflecting the number of isolates, with the partitioning lines within a circle delineating isolates with an identical genotype. The number of different alleles is indicated over the line connecting the sequence types.
Figure S3 Minimum spanning tree of 102 *Mycoplasma bovis* isolates (101 field isolates plus PG45) based on 283 core genome loci, using cgSNV, based on 3,925 SNVs. The clades are
indicated by an encompassing circle. The 102 *M. bovis* isolates are identified by MLST sequence type and production year (ST, Production Year). The nodes are colour coded by the MLST sequence type. The size of the circle reflects the number of isolates represented, with the partitioning lines within a circle delineating isolates with an identical genotype. The number of different alleles is indicated over the line connecting the sequence types.
Figure S4 Maximum-likelihood tree of 130 *Mycoplasma bovis* isolates (129 field isolates plus PG45) based on 14383 SNVs in the core and accessory genomes typed by wgSNV. Clades 1 and 2 are indicated by a surrounding line in red and blue, respectively. The isolates are labelled with...
MLST sequence type and production year (ST, Production Year), and colour coded with MLST sequence type. Each individual node represents a single isolate. No two isolates had identical SNV matrices.