Supplementary Material for

Reoccuring bovine anthrax in Germany on the same pasture after 12 years

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Supplementary Table S1: Genome sequences accession numbers of newly sequenced and additional B. anthracis strains from publically available databases.

| Strain designation | Country    | Year | SNP-group | Accession number (chromosome) |
|---------------------|------------|------|-----------|-------------------------------|
| Ames Ancestor       | U.S.A.     | 1981 | A.Br.Ames | NC_007530                     |
| BF-5                | Germany    | 2021 | B.Br.CNEVA | SRR16572036                  |
| BF-1                | Germany    | 2009 | B.Br.CNEVA | CP047131                      |
| Tyrol 6282          | Austria    | 1979 | B.Br.CNEVA | SRR10743038                  |
| Tyrol 3520          | Austria    | 1980 | B.Br.CNEVA | SRR10743039                  |
| Tyrol 3520          | Austria    | 1980 | B.Br.CNEVA | SRR10743039                  |
| Tyrol 4675          | Austria    | 1988 | B.Br.CNEVA | CP018903                      |
| A024                | Slovakia   | 1972 | B.Br.CNEVA | QAEO00000000                  |
| A016                | Switzerland| *   | B.Br.CNEVA | QAEM00000000                  |
| A046                | Germany    | *   | B.Br.CNEVA | QAEN00000000                  |
| BA0188              | Italy      | 2007 | B.Br.CNEVA | SRR12435826                  |
| **IMB 3011**        | Italy      | 2005 | B.Br.CNEVA | SRR16573065                  |
| 17OD930             | Switzerland| 2017 | B.Br.CNEVA | SRR7100210                   |
| CNEVA-9066          | France     | 1992 | B.Br.CNEVA | NZ_AAEN01000000               |
| ANSES 97-105        | France     | 1997 | B.Br.CNEVA | ERR1596585                    |
| ANSES 11-11_11      | France     | 2011 | B.Br.CNEVA | ERR1596595                    |

**bold:** sequenced in this study; *information missing (Sahl et al., 2016**).

**Sahl JW, Pearson T, Okinaka R, Schupp JM, Gillece JD, Heaton H, et al. (2016) A Bacillus anthracis genome sequence from the Sverdlovsk 1979 autopsy specimens. MBio 7.**
Supplementary Table S2: Primers used for SNP typing of additional isolates of *B. anthracis* BF-5 retrieved from contaminated soil at carcass site.

| Primer Name (position) | Forward (5’-3’) | Reverse (5’-3’) |
|------------------------|-----------------|-----------------|
| SNP 1                  | GCCACACTGGGACTGAGAC | GCCACCTACGTATTACCGCG |
| SNP 2                  | GAGGAAAGAGAGATGTATTGTAGAAG | TACTCCTGGAACTCCTGAAGTAAC |
| SNP 3                  | TTGAGCCATTGTAATAATCATCCCTTC | GTTTCTGTATCATAATAATAATAGTTGGGATG |
| SNP 4                  | ATTTCTTCTTTTCAAGTACATAATAAAGCAG | CTAAAGGAGTTGACGTATTTAAATGGAAG |

Supplementary Table S3: Ct values of 16S rRNA SNP (RT)-PCR of total nucleic acids from blood samples.

| Sample                              | Ct value 16S rRNA SNP-PCR | Ct value 16S rRNA SNP RT-PCR |
|-------------------------------------|---------------------------|----------------------------|
| Inactivated blood 1:10              | 24.9                      | not done                   |
| Inactivated blood 1:100             | 28.1                      | not done                   |
| Inactivated blood 1:1000            | 31.7                      | not done                   |
| Nucleic acid extraction from blood 1:10 | 13.9                  | 9.7                        |
| Nucleic acid extraction from blood 1:100 | 18.1                 | 12.8                       |
| Nucleic acid extraction from blood 1:1000 | 21.5                | 17.8                       |
| H₂O (negative control)              | -                         | -                          |

* total nucleic acids were extracted from inactivated cow blood samples.