



THE UNIVERSITY *of* EDINBURGH

Edinburgh Research Explorer

## Complete Genome Sequence of a Pathogenic Genotype 1 Subtype 3 Porcine Reproductive and Respiratory Syndrome Virus (Strain SU1-Bel) from Pig Primary Tissue

**Citation for published version:**

Lu, ZH, Wilson, AD, Wang, X, Frossard, J-P, Stadejek, T, Archibald, AL & Ait-Ali, T 2015, 'Complete Genome Sequence of a Pathogenic Genotype 1 Subtype 3 Porcine Reproductive and Respiratory Syndrome Virus (Strain SU1-Bel) from Pig Primary Tissue', *Genome announcements*, vol. 3, no. 3, e00340-15. <https://doi.org/10.1128/genomeA.00340-15>

**Digital Object Identifier (DOI):**

[10.1128/genomeA.00340-15](https://doi.org/10.1128/genomeA.00340-15)

**Link:**

[Link to publication record in Edinburgh Research Explorer](#)

**Document Version:**

Publisher's PDF, also known as Version of record

**Published In:**

Genome announcements

**General rights**

Copyright for the publications made accessible via the Edinburgh Research Explorer is retained by the author(s) and / or other copyright owners and it is a condition of accessing these publications that users recognise and abide by the legal requirements associated with these rights.

**Take down policy**

The University of Edinburgh has made every reasonable effort to ensure that Edinburgh Research Explorer content complies with UK legislation. If you believe that the public display of this file breaches copyright please contact [openaccess@ed.ac.uk](mailto:openaccess@ed.ac.uk) providing details, and we will remove access to the work immediately and investigate your claim.



# Complete Genome Sequence of a Pathogenic Genotype 1 Subtype 3 Porcine Reproductive and Respiratory Syndrome Virus (Strain SU1-Bel) from Pig Primary Tissue

Zen H. Lu,<sup>a</sup> Alison D. Wilson,<sup>a</sup> Xinglong Wang,<sup>a</sup> Jean-Pierre Frossard,<sup>b</sup> Tomasz Stadejek,<sup>c</sup> Alan L. Archibald,<sup>a</sup> Tahar Ait-Ali<sup>a</sup>

The Roslin Institute and Royal (Dick) School of Veterinary Studies, University of Edinburgh, Easter Bush, United Kingdom<sup>a</sup>; Virology Department, Animal and Plant Health Agency (APHA), Weybridge, Addlestone, United Kingdom<sup>b</sup>; Department of Pathology and Veterinary Diagnostics, Faculty of Veterinary Medicine, Warsaw University of Life Sciences, Warsaw, Poland<sup>c</sup>

**We report here the complete genome of the pathogenic eastern European subtype 3 porcine reproductive and respiratory syndrome virus (PRRSV) strain SU1-Bel, sequenced directly from a pig lymph node. While sharing substantial sequence similarity with other subtype 3 strains, SU1-Bel is found to harbor unique indels and contain putative novel subgenomic RNAs.**

Received 9 March 2015 Accepted 17 April 2015 Published 21 May 2015

**Citation** Lu ZH, Wilson AD, Wang X, Frossard J-P, Stadejek T, Archibald AL, Ait-Ali T. 2015. Complete genome sequence of a pathogenic genotype 1 subtype 3 porcine reproductive and respiratory syndrome virus (strain SU1-Bel) from pig primary tissue. *Genome Announc* 3(3):e00340-15. doi:10.1128/genomeA.00340-15.

**Copyright** © 2015 Lu et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 3.0 Unported license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Tahar Ait-Ali, [tahar.aitali@roslin.ed.ac.uk](mailto:tahar.aitali@roslin.ed.ac.uk).

Porcine reproductive and respiratory syndrome virus (PRRSV) is the etiologic agent of a swine disease causing immense losses to the pig industry since the late 1980s. This positive-stranded RNA arterivirus exists in two distinct genotypes, European type 1 (PRRSV-1) and North American type 2 (PRRSV-2). The emergence of highly pathogenic PRRSV-2 strains in China has resulted in the death of millions of animals and, more recently, pathogenic PRRSV-1 subtype 3 strains capable of inducing fatal infections in pigs have also been isolated in Belarus (1–4). However, increasing observations suggest that different pathogenic PRRSV-1 strains may elicit different host responses via dissimilar routes. For example, PRRSV-1 subtype 3 strain Lena showed enhanced replication *in vivo* (1–3, 5, 6), whereas SU1-Bel, another subtype 3 strain, caused inflammatory response without a marked increase in replication (1). A whole-genome comparison of these strains is therefore warranted.

We were able to assemble the complete genome of SU1-Bel without any prior knowledge of the sequence from the primary tissue of a pig experimentally infected with the virus (1). Total RNA was isolated from tracheobronchial lymph nodes 3 days postinfection (p.i.), reversed transcribed, and a 101-bp paired-end RNA-sequencing (RNA-seq) library was constructed, as described previously (7). Sequencing was performed on the Illumina HiSeq 2500 platform at Edinburgh Genomics (United Kingdom) (<http://genomics.ed.ac.uk>). Contaminating pig and adapter-containing sequencing reads were discarded, and low-quality (Phred score, <20) reads were trimmed before the remaining reads were assembled *de novo* using Velvet (version 1.2.09) (8) at multiple *k*-mers. The full-length genome sequence was then obtained by merging the Velvet contigs using the Lasergene SeqMan Pro (version 10.1). This genomic sequence was confirmed by remapping the reads back to the consensus sequence using BWA-MEM (version 0.7.10) (9).

The 14,958-nucleotide (nt) SU1-Bel genome shares a substantial sequence similarity with other PRRSV-1 strains, with 79% and

88% similarity with the prototypical strain Lelystad (LV) and strain Lena, respectively. However, not only is the SU1-Bel genome the shortest among the three, it also harbors unique indels. While the highest variability clusters on the *Nsp2* gene, small insertions, compared with that of strain Lena, are present in both *Nsp1* and the 3' untranslated region (UTR). The significance of these variations remains to be investigated.

Consistent with other reported PRRSV strains, an analysis of the genome shows that SU1-Bel contains 10 open reading frames (ORFs), which can be translated into all the known PRRSV (poly)proteins. Interestingly, in addition to the full-length genomic RNA transcripts, contigs from the assembled reads strongly suggest the transcription of multiple novel subgenomic RNAs by SU1-Bel infecting the lymph nodes. Some of them are predicted to encode novel proteins not previously described in other PRRSV-1 strains.

To our knowledge, this is the first complete genome sequence of a subtype 3 PRRSV-1 determined directly from primary tissue. Together with that of strain Lena, the SU1-Bel sequence will enable us to gain further understanding into the genetic diversity, epidemiology, and pathogenesis of PRRSV-1 subtype 3 strains.

**Nucleotide sequence accession number.** The complete genome sequence of SU1-Bel was deposited in GenBank with the accession no. [KP889243](https://www.ncbi.nlm.nih.gov/nuclot/KP889243).

## ACKNOWLEDGMENTS

T.A.-A., A.L.A., and Z.H.L. were supported by a United Kingdom Biotechnology and Biological Sciences Research Council (BBSRC) Institute Strategic Programme grant (BB/J004235/1). A.D.W., J.-P.F., and T.S. were supported by an EC FP7 PoRRSCon grant (agreement no. 245141). Edinburgh Genomics is funded by a BBSRC National Capacity grant (BB/J004243/1). X.W. was funded by the Chinese Scholarship Council.

We thank the COST Action FA0902 and the British Pig Executive (BPEx) for their support.

## REFERENCES

1. Morgan SB, Graham SP, Salguero FJ, Sánchez Cordón PJ, Mokhtar H, Rebel JM, Weesendorp E, Bodman-Smith KB, Steinbach F, Frossard JP. 2013. Increased pathogenicity of European porcine reproductive and respiratory syndrome virus is associated with enhanced adaptive responses and viral clearance. *Vet Microbiol* 163:13–22. <http://dx.doi.org/10.1016/j.vetmic.2012.11.024>.
2. Weesendorp E, Morgan S, Stockhofe-Zurwieden N, Popma-De Graaf DJ, Graham SP, Rebel JM. 2013. Comparative analysis of immune responses following experimental infection of pigs with European porcine reproductive and respiratory syndrome virus strains of differing virulence. *Vet Microbiol* 163:1–12. <http://dx.doi.org/10.1016/j.vetmic.2012.09.013>.
3. Karniychuk UU, Geldhof M, Vanhee M, Van Doorselaere J, Saveleva TA, Nauwynck HJ. 2010. Pathogenesis and antigenic characterization of a new East European subtype 3 porcine reproductive and respiratory syndrome virus isolate. *BMC Vet Res* 6:30. <http://dx.doi.org/10.1186/1746-6148-6-30>.
4. Stadejek T, Oleksiewicz MB, Scherbakov AV, Timina AM, Krabbe JS, Chabros K, Potapchuk D. 2008. Definition of subtypes in the European genotype of porcine reproductive and respiratory syndrome virus: nucleocapsid characteristics and geographical distribution in Europe. *Arch Virol* 153:1479–1488. <http://dx.doi.org/10.1007/s00705-008-0146-2>.
5. Morgan SB, Frossard JP, Pallares FJ, Gough J, Stadejek T, Graham SP, Steinbach F, Drew TW, Salguero FJ. 10 November 2014. Pathology and virus distribution in the lung and lymphoid tissues of pigs experimentally inoculated with three distinct type 1 PRRS virus isolates of varying pathogenicity. *Transbound Emerg Dis*. <http://dx.doi.org/10.1111/tbed.12272>.
6. Weesendorp E, Rebel JM, Popma-De Graaf DJ, Fijten HP, Stockhofe-Zurwieden N. 2014. Lung pathogenicity of European genotype 3 strain porcine reproductive and respiratory syndrome virus (PRRSV) differs from that of subtype 1 strains. *Vet Microbiol* 174:127–138. <http://dx.doi.org/10.1016/j.vetmic.2014.09.010>.
7. Lu ZH, Brown A, Wilson AD, Calvert JG, Balasch M, Fuentes-Utrilla P, Loecherbach J, Turner F, Talbot R, Archibald AL, Ait-Ali T. 2014. Genomic variation in macrophage-cultured European porcine reproductive and respiratory syndrome virus Olot/91 revealed using ultra-deep next generation sequencing. *Virology* 461:11–22. <http://dx.doi.org/10.1016/j.virus.2014.09.010>.
8. Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. *Genome Res* 18:821–829. <http://dx.doi.org/10.1101/gr.074492.107>.
9. Li H. 2013. Aligning sequence reads, clone sequences and assembly contigs with BWA-MEM. arXiv 1303.3997. <http://arxiv.org/abs/1303.3997v2>.