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# *Yersinia pestis* Plasminogen Activator Gene Homolog in Rat Tissue

## Technical Appendix

## Primers Used in *Yersinia pestis* Plasminogen Activator Gene Homolog in Rat Tissues

Technical Appendix Table 1. Primers for amplification and sequencing of the plasminogen activator gene*

| Forward primer | Sequence (5′→3′) | Reverse primer | Sequence (5′→3′) | Position† |
|---|---|---|---|---|
| *pla*sq-f | ATGAAAATCAATCTGAGTGGACAGATCAC | *pla*sq-r | CAGAAGCGATATTGCAGAC | 6990–7602 |
| *pla*sq2-f | TAACTATTCTGTCCGGGAGT | *pla*sq2-r | ATTATCATGTGCCCGAAC | 6696–7339 |

**pla*, plasminogen activator/coagulase.
†Position in entry AF053945 from *Y. pestis* strain KIM5.

Technical Appendix Table 2. Primers for amplification and detection of conserved regions on plasmid pPCP1*

| Forward primer | Sequence (5'→3') | Reverse primer | Sequence (5'→3') | Position† | Present in |
|---|---|---|---|---|---|
| pPratTr21-f | TCTCGGTTCCTCAGGAG | pPratTr21-r | TGCACGTCATACTCTTTTTTCT | 397–1021 | Ype + Yps |
| pPratPst-f | GAGATGGAGAAAGACAGTGA | pPratPst-r | ATTTTAACAATCCACTATCGATA | 4817–5699 | Ype |
| pPratR1-f | AGGCCATGAACGACT | pPratR1-r | GATGGGAAATACAACTACGAAAATTA | 2100–2826 | Ype |

*Ype, *Yersinia pestis*; Yps, *Yersinia pseudotuberculosis.*
†Position in entry AF053945 from *Y. pestis* strain KIM5.

Technical Appendix Table 3. Primers for genewalking

| Primer | Sequence (5'→3') | Function |
|---|---|---|
| usPuTSP1 | ATTTGGTATTAACTGAGATGAT | Upstream *Pla* TSP1 primer |
| us*Pla*TSP2 | CTGCATTAGCACTCCCGGA | Upstream *Pla* TSP2 primer |
| us*Pla*TSP3 | CCCGGACAGAATAGTTATAATGG | Upstream *Pla* TSP3 primer |
| ds*Pla*TSP1 | CCATTGATAAGAATAGTGGAGA | Downstream *Pla* TSP1 primer |
| ds*Pla*TSP2 | TCTGTCTCTATTGGCGGAGA | Downstream *Pla* TSP2 primer |
| ds*Pla*TSP3 | GCTGCCGGTATTTCCAATAA | Downstream *Pla* TSP3 primer |

*Following the instructions of the DNA Walking SpeedUp Premix Kit (Seegene Inc.), gene-specific nested primers targeting the flanking regions of the pla gene were designed. These were used in combination with supplied primers for 3 consecutive rounds of PCR amplification. *Pla*, plasminogen activator/coagulase.

Technical Appendix Table 4. Primers for amplification of the concatenated *pla-rep* sequence*

| Forward primer | Sequence (5'→3') | Reverse primer | Sequence (5'→3') | PCR product |
|---|---|---|---|---|
| us*Pla*TSP1 | ATTTGGTATTAACTGAGATGAT | *Pla*Rep1 | GGTAAATTTTCGTCGAAGTAT | Used for sequencing *pla*-rep |
| us*Pla*TSP1 | ATTTGGTATTAACTGAGATGAT | *Pla*Rep2 | TCATCCGTGTGATAGGGAC | Not formed |
| us*Pla*TSP1 | ATTTGGTATTAACTGAGATGAT | *Pla*Rep3 | GTCGCTCCAGTCAATCG | Not formed |
| us*Pla*TSP1 | ATTTGGTATTAACTGAGATGAT | *Pla*Rep4 | GGCGCGGTTCTCACGG | Not formed |
| us*Pla*TSP1 | ATTTGGTATTAACTGAGATGAT | *Pla*Rep5 | ATCGCCCTGTAAGCCCATTT | Not formed |
| us*Pla*TSP1 | ATTTGGTATTAACTGAGATGAT | *Pla*Rep6 | TCYTGCCAATAGCCC | Not formed |

*Primers *Pla*Rep 2–6 were based on an alignment of conserved domains in enterobacterial *rep* genes. *Pla*, plasminogen activator/coagulase; rep, replicon.