

Nucleotide sequence of the penicillin-binding protein 2B gene of Streptococcus pneumoniae strain R6

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Previously only part of the penicillin-binding protein 2B gene (penA) of Streptococcus pneumoniae had been cloned and sequenced (1). The whole of penA was isolated on a 3.2-kb EcoRV fragment and cloned into pBGS9 (2). E.coli TG1 containing the resulting plasmid (pCGD3) expressed large amounts of a novel penicillin-binding protein of 74kD. A fragment of the insert, containing the previously unsequenced amino-terminal coding region of penA, was subcloned into bacteriophage M13 and sequenced on both strands using a series of oligonucleotide primers.

GATATCTGGTTTGTCCATAGGGAAGCTCCTTTACATGAGTCTACTGGAAAGACTAGATCAGGGAATAGTCACACTTCTATTATAACACATAATATAAGGATAGATAAATAAAAAAGCAGTC 120
M R K
TCTGTTTTAAAAAGCAAAAAATCGAAAAAGCTCTCTCTTTTCCATAAATTTTCTACTCAAATTTGGTACAATTAAGAGTAAGATTTTAAAGTTAGAANAAGAGACTGATTTGTATGAGAAA 240
F N S M S I P I R L L H L L F S I Y I L L L F M H T I G R L L L M Q V F N K D P Y E
ATTTAACAGCCATTCTGATTCCGATTCGGCTTAATTTATTGTTTTCAATCGCATTTTACTCTTTATGACCATTATTTGGTCGTTTGTGTATATGACGGTTTTCAACAAAGGATTTTTCGA 360
K K L A S A S Q T K I T S S S A R G E I Y D A S G K P L V E N T L K Q V V S F T
AAAAAGCTAGCTCAGCTAGTCAGACCAAGATTACAGACAGTTTCAGCCCGGGGAAATTTATGATGCTAGTGGAACCTTTGGTAGAAAACCTTTGGTAGAAAACCTGTAAGACAGGTTTCTCTTAC 480
R S N K M T A T D L K E T A K K L L T Y V S I S S P N L T E R A G D Y Y L A D P
GGTAGCAATAAATGACGGCTACAGACTTAAAAAGAACAGCTAAAAAGTTACTGACTTATGTGAGCATCAGTCTCCAAATTTGACAGACAGCAGCTGGGGATTACTTTGGCTGATCC 600
E I Y K K I V E A L P S E K R L D S D G N R L S E S E L Y N N A V D S V Q T S Q
TGAATCTATAAAAAATAGTGAAGCTCTCCCAAGCGAGAAACGCTTGGATTTCAGATGGCAATCGTCTATCCGAATCAGAAGCTATAACAATAGCGGTCGATAGTGTACAACAGAGTCA 720
L N Y T E D E R K E I Y L F S Q L N A V G H F A T G T I A T D P L N D S Q V A V
ACTAATACAGAGGATGACAAAGAAAGAAATCTACTCTTTTATGTCAGTTAAATGCTGTGGAAACCTTGGCACAGGGACCATGCTACAGATCCCTCAAATGATTTCTCAGGTGGCTGT 840
I A S I S K E M P G I S I S T S W D R K V L E T S L S S I V G S V S S E K A G L
TATTGCCCTATTTCAAAGGAGATGCTGGCATTAGTATTTCTACTCTTGGGATAGAAAAGGTTTGGAAACCTTCCCTTCTCTATAGTTGGGAGTGTCCAGTGA AAAAGCTGGTCT 960
P A E E A E A Y L K K G Y S L N D R V G T S Y L E K Q Y E E T L Q G K R S V K E
CCACGGGAAGAGCAGAAGCTATCTTAAAAAGGCTATTCTCTAAATGACCGTGTAGGAACCTCTATTTGGAAAAGCAATATGAAGACCTTACAAGAAAACCGCTGGTAAAAAGA 1080
I H L D K Y G N M E S V D T I E E S K G N N I K L T I D L A F Q D S V D A L L
AATCCATCTGGATAAATATGGCAATATGGAAGCGGTGGATAAATGGAAGGAGTAGTAAGGGAACAAATATCAAATGACCATGATTTGGCTTTCCAAAGATAGCGTGTGATCTTACT 1200
K S Y F N S E L E N G G A K Y S E G V Y A V A L N P R T G A V L S M S G I K H D
GAAAAGTTATTCAAATCTGAGCTAGAAAATGGTGGAGCAAGTATTCGAAAGGTGTCTATGACAGTCGCCTTAAACCAAAAACAGGTGGCGTTTGTCTATGTACAGGGATTAACACTGA 1320
L K T G E L T P D S L G T V T N V F V P G S V V K A A T I S S G W E N G V L S G
CTTGAACCGGAGAGTATGAGCTGATTCCTTGGGAACGGTAACCAATGCTTTGTTCCAGGTTTCGGTTGCAAGCGCGGACCATCAGCTCAGGTTGGGAAAATGGAGTCTTGTACAGG 1440
N Q T L T D Q S I V F Q G S A P I N S M W Y T Q A Y G S F P I T A V Q A L E Y S S
AAACAGACTTGAACAGAGCTCAATGCTCTCCAAAGGTTTCAGCTCCCATCAATCTTGGATACTCAGGCTTACGGTTTATTCCATCACAGCGGTCGAAAGCTGCGATTCATC 1560
N T Y M Y Q T A L G L M G Q T Y Q P H M F V G T S N L E S A H E K L R S T F G E
AAATACCTATGGTCCAAACAGCTTAGGCTCTATGGGGCAACCTATCAACCCCAATATGTTGTGGCCAGCAGCAATCTAGAGCTGTCTAGGAGAACTGGCTTCACTTTGGCA 1680
Y G L G T A T G I D L P D E S T G F V P K E Y S F A N Y I T N A F G Q F D N Y T
ATATGGCTTGGTACTGCGACAGGAANTTGACCTACAGATGAATCTACTGGATTTGTTCCAAAGAGTATAGCTTTGCTTAATTAATTACTAATGCTTTGGGAGTTTGTATAACTATAC 1800
P M O L A Q Y V A T I A N N G V R V A P R I V E G I Y G N N D K G G L G D L I Q
GCGATGCAAGTGGCTCAGTATGAGCAACTATTGCAAAATAGGTTGCTGTTGGCTCTCGTATTTGTAAGGCATTTATGGTAAATGATAAGGGAGGACTGGGTGACTTGATTCA 1920
Q L Q P T E M N K V N I S D S D M S I L H Q G F Y Q V A H G T S S G L T T C R A F
GCAATCGAACCGACAGAGATGAATAGSTCAATATCTCGACTCCGATATGAGCATCTGACCAAGGTTTTTATCAGGTTGCCATGGTACTAGTGATTGACAATCGGACGTGCTT 2040
S N G A L S V S I S G K T G T A E S Y V A D G G Q A T N T N A V A Y A P S D N P Q
TTCAAATGGTCTTGGTATCCATTAGCGAAAACAGGTCACGGCAAGACTATGTGGCAGATGGTCAGCAGCAACCAATCAACATGCGGCTGATGCCCTGATGCAATCCCA 2160
I A V A V V F P H N T N L T H G V G P S I A R D I I N L Y Q K Y H P H *
AATCGCTGCGCAGTGGCTCTTCCCTCATAATACCAACTCTAAACAAATGGGTAGGACCTCCATTGGCGGTGACATTCACTCTCTATAAAAAATACCATCAATGACTAGAAAAGGAAA 2280

Fig.1. The sequence shown is that of the penA gene of S.pneumoniae strain R6. The active site serine is marked with an asterisk.

REFERENCES (1) Dowson C.G. et al. (1989) Mol. Microbiol. 3, 95-102; (2) Spratt B.G. et al. (1986) Gene 41, 337-342.