Certificate of Analysis for NR-4694

Yersinia pestis, Strain Kimberley Derivative 12 (D12)

Catalog No. NR-4694

Product Description: *Yersinia pestis* (*Y. pestis*) is an aerobic, non-spore-forming, Gram-negative rod-shaped bacterium. *Y. pestis*, strain Kimberley(D12) is an avirulent derivative of the Kimberley strain that contains the pMT1 and pPCP1 plasmids as well as the unstable *pgm* locus, but lacks the pCD1 plasmid that is essential for virulence.

Lot[1]: 57753204 **Manufacturing Date: 20JUL2007**

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis		
Cellular morphology	Gram-negative rod	Gram-negative rod
Colony morphology[2]	Report results	Circular, convex, entire, opaque (Figure 1)
Congo red (CR) agar[3,4]	Red colonies (Crb^+)	Red colonies (Crb^+)
Biochemical Analyses		
Analytical profile index (API 20 E®)	Consistent with *Y. pestis*	Consistent with *Y. pestis*
Nitrate reduction	Positive	Positive
Fermentation of glycerol	Negative	Negative
Urease	Negative	Negative
Genotypic Analysis		
Sequencing of 16S ribosomal RNA gene (1420 bp)	Consistent with *Y. pestis*	Consistent with *Y. pestis*[5]
PCR Assay of Extracted DNA		
16S ribosomal RNA gene	~ 1500 bp amplicon	~ 1500 bp amplicon
Presence of virulence-associated plasmids		
pMT1 (pFra; 100 kb plasmid)	~ 1200 bp amplicon	~ 1200 bp amplicon
pCD1 (pYV; 70 kb plasmid)	None detected	None detected
pPCP1 (pPla; 9.5 kb plasmid)	~ 400 bp amplicon	~ 400 bp amplicon
Viability (post-freeze)[4]	Growth on agar	Growth on agar

[1]Y. pestis, strain Kimberley(D12) was deposited by Professor Robert R. Brubaker of the Department of Microbiology and Molecular Genetics at Michigan State University, East Lansing, Michigan. NR-4694 was prepared by broth (Trypticase Soy Broth; BD 211768) culture of the deposited material. Broth inoculum was added to Kolles which were grown 48 hours at 28°C and aerobic atmosphere to produce this lot.
[2]48 hours at 28°C and aerobic atmosphere on Tryptic Soy Agar (BD 236950)
[3]24 hours at 28°C and aerobic atmosphere on CR agar
[4]Hare, J. M. and K. A. McDonough. "High-Frequency RecA-Dependent and -Independent Mechanisms of Congo Red Binding Mutations in *Yersinia pestis*." J. Bacteriol. 181 (1999): 4896-4904. PubMed: 10438760.
[5]Also consistent with other *Yersinia* species

Figure 1

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