Cloning and sequencing of bacteriophage T4 genes between map positions 128.3 – 130.3

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The bacteriophage T4 region located between map position 128.3 and 130.3 is of special interest since it contains the early gene 31 encoding a gene product (gp) essential for proper capsid assembly (1–3). In the absence of gp31 the major capsid protein gp23 assembles into ‘lumps’ representing some sort of inclusion bodies as they are often revealed when trying to express cloned eukaryotic genes in E. coli cells. In the upstream region of the 1906-bp EcoRI-PstI fragment containing gene 31 that was cloned and sequenced earlier (4), we found two short open reading frames which we called orf A (78 amino acids long) and orf B (102 amino acids long). Downstream of gene 31 we also found two unknown reading frames, orf C (78 amino acids long) and orf D (97 amino acids long). We subsequently subcloned all reading frames of this fragment into plasmids under the control of the phage T7 promoter. While reading frames A, B and gene 31 gave rise to high-level expression in E. coli strain BL21 (DE3) containing the T7 RNA polymerase gene on its genome (5), the products of reading frames C and D appeared to be very toxic to the same cells.

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REFERENCES


1 GM!T(^GTGMTAAT(MmCGmGGTAAMCTCACAG(^m^^

121 AGTAnATAAGACAMTCGGGTTOTArcMTAGACCATATAGTTGGAMTCTGAm^^

241 TAATAGGTAAATAAATGAAACT(!ACAACTGAGCAGAMGTAGCMnCGTGAMOT

361 GTACTATGAAAGGTACTCGTGATGCAGAtmATGCCMCCATGCAAATO

481 CGTGGCGAGGItTTTCTAATTGAAATTTGAAATGAAACTGAGCAGAMGTAGCMnCGTGAMOT

601 TTAACAAAMGGAAAAACCGTCTGACTCTGCTGTAATGTATTATTATTCTGAACTCTCAACGCGGGTACGAAACGCTACGAATTAC

721 AOTATTATCGGTAAACGTGmAAGGTGAAGTCCCTimGTGTGTAGrc^^

841 ?CMAnCGAJUTGnCCGCATCCTmCTAGCTCTGGGTCnMGCAGCCAAAAGAAATTAAACAAMATTCGmCCTCTCACTATIJWGm

961 ATAATAATATGAAnGGGTGTCGISMTAATAAGnMCCGMCMTO^ 

1081 AATCTCTAACGAGAATmTAAATGAnAAACUnA(^CACGCTCTTGMCTGCMCGAAACGCATGGMTMTGGTCACGM

1201 TCTCTACTCGCTGAAACCGTCTGACTCTGCTGTAATGTATTATTATTCTGAACTCTCAACGCGGGTACGAAACGCTACGAATTAC

1321 GCTCGCTACTCGCTGAAACCGTCTGACTCTGCTGTAATGTATTATTATTCTGAACTCTCAACGCGGGTACGAAACGCTACGAATTAC

1441 ACCCCGACAGTmACGGTGTACCTOTGAATGTGMTGATGACGGGmATGGmT^^

1561 AGTCCTGCATffiCAmAAAAGAIMmATMTGGCAAAACMGCTAMGC^ 

1681 CGTATCAATCAAACTGn(y(^GATCATGCGCCGAGCACGTGCGGTT(Tr(^GATGATGOTCT(^TTTTGGTMG(aGAMGCA

1906 Het (orf I)

216x111 Bet (orf D)

249x111 Ter (orf B)