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Short Sequence-Paper

Cloning and sequencing of the *tuf* genes of *Streptomyces coelicolor* A3(2)

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Abstract

Two *tuf* genes are present in *Streptomyces coelicolor* A3(2), which have been cloned and sequenced. These genes show a high degree of nucleotide sequence identity to the *tuf1* and *tuf3* genes of *Streptomyces ramocissimus*: the *tuf1* genes are 94% identical, the *tuf3* genes 87%. *S. coelicolor tuf1* encodes a protein of 396 amino acids, while *tuf3* encodes a protein of 391 amino acids.

Keywords: Elongation factor Tu; Sequence comparison; Gene cloning; (*Streptomyces*)

One of the most abundant proteins in the bacterial cytoplasm is the *tuf*-encoded polypeptide chain elongation factor Tu (EF-Tu). Two *tuf* genes occur in *Escherichia coli*, encoding elongation factors EF-TuA and EF-TuB which differ only in their C-terminal amino acid [1,2]. The proteins occur in the bacterial cell in a 1:1 ratio [3], and are functionally indistinguishable.

Recently, we showed that in the kirromycin producer *Streptomyces ramocissimus* three *tuf* genes occur, which have been cloned and sequenced [4]. Analysis of these genes revealed that they are unexpectedly heterogeneous: *tuf1* and *tuf2* show 85% nucleotide sequence identity, whereas *tuf3* shows only 70% identity to *tuf1* and *tuf2*, which is in striking contrast to the much higher similarities found among *tuf* genes in other microorganisms [5,6]. EF-Tu1 has been shown to be a genuine elongation factor, but no physiological function could be revealed for the other two putative elongation factors. Hybridization analysis of other streptomycetes has shown that *Streptomyces coelicolor* and *Streptomyces lividans*, genetically the best-characterised streptomycetes, have only two *tuf* genes.

Cloning of the S. coelicolor tuf1 and tuf3 genes. *S. coelicolor* presumably has two *tuf* genes, designated *tuf1* and *tuf3* by analogy to their homologues in *S. ramocissimus*. From Southern hybridization data it was concluded that *tuf1* could be cloned as an approx. 4.5 kb *Bam*HI fragment and *tuf3* as an approx. 10 kb *Bam*HI fragment [7]. *S. coelicolor* M145 total DNA was digested with *Bam*HI and separated electrophoretically on a 0.7% agarose gel in TAE buffer, whereupon fragments of the appropriate size were isolated from the gel.

For *tuf1*, these fragments were subcloned into *Bam*HI-digested pAT153 [8] and screened by hybridization of the *E. coli* colonies with the 244 bp *Sma*I fragment internal to the *S. ramocissimus tuf1* gene, which encodes most of the GTP-binding region. One positive signal was obtained, which appeared to contain a 4.3 kb insert, corresponding to the size expected on the basis of Southern hybridization data. This clone, which was designated pASCT1-1, contained the gene homologous to *S. ramocissimus tuf1*, as shown below.

For cloning of *tuf3*, fragments were cloned into pBR329 [9] and plasmid DNA was isolated from 600 colonies in pools of 24. The DNA was digested with

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*Bam*HI, subjected to agarose gel electrophoresis and blotted onto a Hybond-N nylon filter. Pools with the correct insert were identified on the basis of hybridization signals obtained with the 600 bp *Sal*I fragment from *S. ramocissimus tuf3*. After repeated colony purification and screening, one DNA preparation yielding an unambiguous positive signal was obtained. *Bam*HI digestion proved the DNA to contain a 10.3 kb insert. The clone was designated pBSCT3–1. It was shown to contain the *tuf3* gene, as is demonstrated below.

Sequences of the inserts of pASCT1–1 and pBSCT3–1: nucleotide and amino acid comparisons. Sequencing of

```
   1  CTCGAGCCGATGATGGCCGTCGAGGTCACCACGCCCGAGGACTACATGGGCGACGTCATCGGCGACATCAACTCC   75
      L  E  P  M  M  A  V  E  V  T  T  P  E  D  Y  M  G  D  V  I  G  D  I  N  S

  76  CGCCGTGGCCAGATCCAGGCCATGGAGGAGCGGATGGGTGCCCGCGTCGTGAAGGGCCTCGTGCCGCTGTCGGAG   150
      R  R  G  Q  I  Q  A  M  E  E  R  M  G  A  R  V  V  K  G  L  V  P  L  S  E

 151  ATGTTCGGCTACGTCGGAGACCTCCGCAGCAAGACGTCGGGTCGCGCAAGCTACTCGATGCAGTTCGACTCCTAC   225
      M  F  G  Y  V  G  D  L  R  S  K  T  S  G  R  A  S  Y  S  M  Q  F  D  S  Y

 226  GCCGAGGTTCCCCGGAACGTCGCCGAGGAGATCATCGCGAAGGCCAAGGGCGAGTAACGGGCTACTCCGTTTAAC   300
      A  E  V  P  R  N  V  A  E  E  I  I  A  K  A  K  G  E  *

 301  GGACCCCGTTCTCACGCTTTAGGCTTGACCCCGGAGCCTGCATGGGGCATTCCGCCGTGAACCCGGTGGAATGCC   375

 376  CCCGGCACCCGGGCTTTCCAGCAAAGATCACCTGGCGCCGATGAGTAAGGCGTACAGAACCACTCCACAGGAGGA   450

 451  CCCCAGTGGCGAAGGCGAAGTTCGAGCGGACTAAGCCGCACGTCAACATCGGCACCATCGGTCACATCGACCACG   525
           V  A  K  A  K  F  E  R  T  K  P  H  V  N  I  G  T  I  G  H  I  D  H  G

 526  GTAAGACGACCCTCACGGCCGCCATTACCAAGGTGCTGCACGACGCGTACCCGGACATCAACGAGGCGTCGGCGT   600
        K  T  T  L  T  A  A  I  T  K  V  L  H  D  A  Y  P  D  I  N  E  A  S  A  F

 601  TCGACCAGATCGACAAGGCTCCCGAAGAGCGCCAGCGCGGTATCACCATCTCGATCGCGCACGTCGAGTACCAGA   675
        D  Q  I  D  K  A  P  E  E  R  Q  R  G  I  T  I  S  I  A  H  V  E  Y  Q  T

 676  CCGAGGCGCGTCACTACGCCCACGTCGACTGCCCCGGTCACGCCGACTACATCAAGAACATGATCACGGGTGCCG   750
        E  A  R  H  Y  A  H  V  D  C  P  G  H  A  D  Y  I  K  N  M  I  T  G  A  A

 751  CGCAGATGGACGGCGCCATCCTCGTGGTCGCCGCCACCGACGGCCCGATGCCGCAGACCAAGGAGCACGTGCTCC   825
        Q  M  D  G  A  I  L  V  V  A  A  T  D  G  P  M  P  Q  T  K  E  H  V  L  L

 826  TGGCCCGCCAGGTCGGCGTTCCGTACATCGTGGTCGCCCTGAACAAGGCCGACATGGTGGACGACGAGGAGATCC   900
        A  R  Q  V  G  V  P  Y  I  V  V  A  L  N  K  A  D  M  V  D  D  E  E  I  L

 901  TGGAGCTCGTCGAGCTCGAGGTGCGTGAGCTCCTCTCCGAGTACGAGTTCCCGGGCGACGACGTTCCCGTCGTCA   975
        E  L  V  E  L  E  V  R  E  L  L  S  E  Y  E  F  P  G  D  D  V  P  V  V  K

 976  AGGTCTCCGCTCTGAAGGCCCTCGAGGGCGACAAGGAGTGGGGCAACTCGGTCCTCGAGCTCATGAAGGCCGTGG   1050
        V  S  A  L  K  A  L  E  G  D  K  E  W  G  N  S  V  L  E  L  M  K  A  V  D

1051  ACGAGGCCATCCCGGAGCCCGAGCGCGACGTCGACAAGCCGTTCCTGATGCCGATCGAGGACGTCTTCACCATCA   1125
        E  A  I  P  E  P  E  R  D  V  D  K  P  F  L  M  P  I  E  D  V  F  T  I  T

1126  CCGGTCGCGGTACGGTCGTCACCGGCCGCATCGAGCGTGGTGTCCTCAAGGTCAACGAGACCGTCGACATCATCG   1200
        G  R  G  T  V  V  T  G  R  I  E  R  G  V  L  K  V  N  E  T  V  D  I  I  G

1201  GCATCAAGACCGAGAAGACCACCACCACGGTCACCGGCATCGAGATGTTCCGCAAGCTCCTCGACGAGGGCCAGG   1275
        I  K  T  E  K  T  T  T  T  V  T  G  I  E  M  F  R  K  L  L  D  E  G  Q  A

1276  CCGGTGAGAACGTCGGTCTGCTGCTTCGCGGCATCAAGCGCGAGGACGTCGAGCGCGGCCAGGTCATCATCAAGC   1350
        G  E  N  V  G  L  L  L  R  G  I  K  R  E  D  V  E  R  G  Q  V  I  I  K  P

1351  CGGGCTCGGTCACCCCGCACACCGAGTTCGAGGCCCAGGCCTACATCCTGTCGAAGGACGAGGGTGGCCGTCACA   1425
        G  S  V  T  P  H  T  E  F  E  A  Q  A  Y  I  L  S  K  D  E  G  G  R  H  T

1426  CCCCCTTCTTCAACAACTACCGTCCGCAGTTCTACTTCCGTACGACGGACGTGACCGGCGTCGTGACCCTCCCCG   1500
        P  F  F  N  N  Y  R  P  Q  F  Y  F  R  T  T  D  V  T  G  V  V  T  L  P  E

1501  AGGGCACCGAGATGGTCATGCCGGGTGACAACACCGAGATGAAGGTGGAGCTCATCCAGCCCGTCGCCATGGAAG   1575
        G  T  E  M  V  M  P  G  D  N  T  E  M  K  V  E  L  I  Q  P  V  A  M  E  E

1576  AGGGCCTGAAGTTCGCCATCCGCGAGGGTGGCCGGACCGTGGGCGCCGGCCAGGTCACCAAGATCAACAAGTAAC   1650
        G  L  K  F  A  I  R  E  G  G  R  T  V  G  A  G  Q  V  T  K  I  N  K  *

1651  TCCGCTTGCTTGTCGGTCGACCGACCTGACATGGGCTGATGCCTGAAGGGCCGTACGACTTCGGTCGTACGGGTC   1725

1726  CTTTCGCCATGTGCGGTCGGGTCCAGGCCGCTGAGGAAGTCGCCCTGCCAGAGCGCCGCGGCCGTGCGCAGCCGG   1800

1801  GCCACCGCCTCTTCCCGGCGGTCCTCGTGGCCGAGCCGTCTGGCCCCGGCCACGAGGGCGGTGAAGAGTACGGCG   1875
```

Fig. 1. Nucleotide sequence of *tuf1* and flanking regions, which include the end of the *fus* gene. The deduced amino acid sequences of both *fus* (nt positions 1–282; encoding EF-G) and *tuf1* (nt positions 456–1649; encoding EF-Tu1) are given below the nucleotide sequence.

pASCT1–1 resulted in the identification of an ORF of 1194 bp, putatively encoding a 396 amino acid protein (Fig. 1). Sequencing of pBSCT3–1 identified an ORF of 1179 bp, corresponding to a protein of 391 amino acids (Fig. 2). The ORF located on pASCT1–1 is very similar to *S. ramocissimus tuf1*, and a second ORF is located upstream of it that very much resembles the *S. ramocissimus fus* gene, indicating its probable location in the so-called S12 operon, a location typical for the major *tuf* gene in all microorganisms [10]. Therefore, the gene was designated *tuf1*. Again on the basis of the very high similarity to its homologue in *S. ramocissimus*, the ORF identified on pBSCT3–1 was designated *tuf3*.

An amino acid alignment of the *tuf*-gene products of *S. coelicolor*, *S. ramocissimus* and *E. coli* is shown in Fig. 3. From this alignment it follows that the *tuf1* and *tuf3* gene products contain the consensus sequences for GTP binding proteins [11] and show a perfect fit with the D-loop motif [12]. From alignments of both the *tuf*-gene nucleotide sequences and the amino acid sequences deduced thereof, identities were calculated (Table 1). The *tuf1* genes of *S. coelicolor* and *S. ramocissimus* are 94% identical, and the deduced amino acid sequences of their gene products (designated EF-Tu1) are 96% identical. The *tuf3* genes of these organisms are 87% identical, their gene products (designated EF-Tu3) sharing 91% identical amino acids. The low similarity of EF-Tu3 to EF-Tu1, the major EF-Tu in *Streptomyces*, is underlined by the fact that EF-Tu3

```
   1  GGTACCGCTCTCGAACGGCCGTTCCATAAAAAACCATTCGACGTGCGACGAAGCGTCGGCGATGATCTGTCTCAT   75

  76  GTTCCGGTACGCCTTCCACCTCGCAGCATCCGCGGTCGCGGATGCCCCGAAGGCTGCCGTCGCCCACTTCACCGC  150

 151  CGCAGTCGACGGCGCCCGAAGCTGACCCTCCCCGGATCGTCCGGCGGACCCCGCAGGGGGAGGGTCGGCCGGCCC  225

 226  CAGGGGTCCCCACCACCGCCCACGAGGCTTCGAGGTACCGCCATGTCCAAGACGGCGTACGTCCGCACCAAACCG  300
                                              M  S  K  T  A  Y  V  R  T  K  P

 301  CATCTGAACATCGGCACGATGGGCCATGTCGACCACGGCAAGACCACCCTGACCGCCGCCATCACCAAGGTCCTC  375
      H  L  N  I  G  T  M  G  H  V  D  H  G  K  T  T  L  T  A  A  I  T  K  V  L

 376  GCCGAGCGCGGGGCCGGCAGCACCACCCAGTACGTTTCGTTCGACCGCATCGACCGCGCCCCGGAGGAGGCGGCG  450
      A  E  R  G  A  G  S  T  T  Q  Y  V  S  F  D  R  I  D  R  A  P  E  E  A  A

 451  CGCGGCATCACCATCAACATCGCGCACGTCGAGTACGAAACCGACACCCGGCACTACGCCCACGTCGACATGCCC  525
      R  G  I  T  I  N  I  A  H  V  E  Y  E  T  D  T  R  H  Y  A  H  V  D  M  P

 526  GGCCACGCCGACTACGTCAAGAACATGGTCACCGGCGCCGCCCAGCTCGACGGGGCGATCCTCGTCGTATCCGCG  600
      G  H  A  D  Y  V  K  N  M  V  T  G  A  A  Q  L  D  G  A  I  L  V  V  S  A

 601  CTGGACGGGATCATGCCGCAGACCGCCGAACACGTGCTGCTCGCCCGGCAGGTGGGCGTCGACCACATCGTGGTC  675
      L  D  G  I  M  P  Q  T  A  E  H  V  L  L  A  R  Q  V  G  V  D  H  I  V  V

 676  GCGCTCAACAAGGCCGACGCGGGTGACGAGGAGCTGACCGACCTGGTCGAGCTGGAGGTGCGCGAACTGCTCACC  750
      A  L  N  K  A  D  A  G  D  E  E  L  T  D  L  V  E  L  E  V  R  E  L  L  T

 751  GCGCACGGCTACGGCGGCGACGCCGTGCCCGTCGTACGGGTCTCGGGGCTGAAGGCCCTGGAGGGCGACCCGCGG  825
      A  H  G  Y  G  G  D  A  V  P  V  V  R  V  S  G  L  K  A  L  E  G  D  P  R

 826  TGGACGGCCTCCGTCGAGGCGCTGCTCGACGCCGTGGACACGTACGTGCCCATGCCCGAGCGGTACCTGGACGCG  900
      W  T  A  S  V  E  A  L  L  D  A  V  D  T  Y  V  P  M  P  E  R  Y  L  D  A

 901  CCGTTCCTGCTGCCGGTGGAGAACGTGCTCACCATCACCGGCCGCGGCACCGTCGTCACCGGCGCCGTCGAGCCG  975
      P  F  L  L  P  V  E  N  V  L  T  I  T  G  R  G  T  V  V  T  G  A  V  E  P

 976  GGCACCGTGCGCGTCGGCGACCGGGTCGAGGTGCTCGGGGCGTCCGTCGAGACGGTCGTCACCGGCCTGGAGACC 1050
      G  T  V  R  V  G  D  R  V  E  V  L  G  A  S  V  E  T  V  V  T  G  L  E  T

1051  TTCGGCAAGCCGATGGAGGAGGCGCAGGCCGGGGACAACGTGGCGCTGCTGCTGCGCGGGGTCGCCCGCGACACG 1125
      F  G  K  P  M  E  E  A  Q  A  G  D  N  V  A  L  L  L  R  G  V  A  R  D  T

1126  GTGCGCCGCGGGCAGGTGGTCGCCGCACCCGGCAGCGTCGTCCCCGCGCGGCGTTTCCGGGCCCGGGTGTACGTG 1200
      V  R  R  G  Q  V  V  A  A  P  G  S  V  V  P  A  R  R  F  R  A  R  V  Y  V

1201  CTCTCGGCGCGCGAGGGCGGGCGCTCGACACCGCTCACCACCGGATACCGGCCGCAGTTCTACATCCGCACCGCC 1275
      L  S  A  R  E  G  G  R  S  T  P  L  T  T  G  Y  R  P  Q  F  Y  I  R  T  A

1276  GACGTGGTCGGCGACGTGGACCTCGGCGAGGAGGCCGTCGCCCGGCCCGGGGACACCGTCACCATGACGGTCGAG 1350
      D  V  V  G  D  V  D  L  G  E  E  A  V  A  R  P  G  D  T  V  T  M  T  V  E

1351  CTGGGACGGGACGTGCCGCTGGAGACGGGGCTCGGCTTCGCGATCCGCGAGGGCGGTCGCACCGTGGGGGCGGGG 1425
      L  G  R  D  V  P  L  E  T  G  L  G  F  A  I  R  E  G  G  R  T  V  G  A  G

1426  ACCGTGACCGCGGTGGAGTGAGCGCGCCGGGGTGAGTGGGCCGGGCTGAGGGC 1478
      T  V  T  A  V  E  *
```

Fig. 2. Nucleotide sequence of *tuf3* and flanking sequences. The deduced amino acids sequence of *tuf3* is given below the nucleotide sequence.

```
Sc EF-Tu1     VAKAKFERTK PHVNIGTIGH IDHGKTTLTA AITKVLHDAY PD.INEASAF   49
Sr EF-Tu1                                                .L   TP    49
Sc EF-Tu3     MS TAYV      L    M   V               AERG AGSTTQYVS   50
Sr EF-Tu3     MS TAYV      L    M   V               AERG SGT...FVP   47
Sr EF-Tu2           Q                                RF   .L PFTP   49
Ec EF-TuA      S E          V      V             T  AKT  GG...A R    47
Consensus     V-K-KF-RTK PHVNIGTIGH -DHGKTTLTA AITKVL---- ---------F

Sc EF-Tu1     DQIDKAPEER QRGITISIAH VEYQTEARHY AHVDCPGHAD YIKNMITGAA   99
Sr EF-Tu1      N                                                      99
Sc EF-Tu3      R  R    A A     N       E DT        M       V   V     100
Sr EF-Tu3      R  R    A A     N       E DT        M       V   V      97
Sr EF-Tu2                                                             99
Ec EF-TuA         N    K A     NTS     D PT                V          97
Consensus     D-ID-APEE- -RGITI-IAH VEY-T--RHY AHVDCPGHAD Y-KNMITGAA

Sc EF-Tu1     QMDGAILVVA ATDGPMPQTK EHVLLARQVG VPYIVVALNK ADMVDDEEIL  149
Sr EF-Tu1                                                       M     149
Sc EF-Tu3      L       S  L  I    A             DH          .AG   LT  149
Sr EF-Tu3      L       S  L  I    A             DH          .AG   LT  146
Sr EF-Tu2                                                  T          149
Ec EF-TuA                        R   I  G         I F     C       L   147
Consensus     QMDGAILVVA ATDGPMPQT- EHVLLARQVG VPYIVVALNK ADMVDDEE--

Sc EF-Tu1     ELVELEVREL LSEYEFPGDD VPVVKVSALK ALEGDKEWGN SVLELMKAVD  199
Sr EF-Tu1                          L   R           AQ TQ    D        199
Sc EF-Tu3     D           TAHGYG  A     R  G        PR TA   EA LD    199
Sr EF-Tu3     D       D     HGYG  G A   R  G        PK TA  IEA LD    196
Sr EF-Tu2               T                    R      PR TR      LD    199
Ec EF-TuA         M         Q D      T I RG          A  EA KI   AGFL  197
Consensus     ELVELEVREL LSEY-FPGDD -PVVRVSALK ALEGD--WT- SVL-L--AVD

Sc EF-Tu1     EAIPEPERDV DKPFLMPIED VFTITGRGTV VTGRIERGVL KVNETVDIIG  249
Sr EF-Tu1      S                                                      249
Sc EF-Tu3     TYV M   YL  A   L V N  L           AV P TV R GDR EVL    249
Sr EF-Tu3     TYV M   Y   A   L V N  L           AV   TV R GNR EVL    246
Sr EF-Tu2      FV   V     R                          T  N  TE E      249
Ec EF-TuA     SY      AI      L       S S         V   II   G E E V   247
Consensus     ---PEPER-V D-PFL-PIED VFTITGRGTV VTGR-ERG-- -V---VEI-G

Sc EF-Tu1     IKTEKTTTTV TGIEMFRKLL DEGQAGENVG LLLRGIKRED VERGQVIIKP  299
Sr EF-Tu1                                                             299
Sc EF-Tu3     ASV ... V    L T G PM E A   D  A      VA DT  R    VAA   296
Sr EF-Tu3     AGL ... V    L T G PM   A   D  A      VP DA  R  H VAA   293
Sr EF-Tu2      HEQR R                  R           V   Q       V R    299
Ec EF-TuA        . TQKS C   V          R       V       E I     LA     296
Consensus     I--E---TTV TG-EMFRKLL DEGQAGENVG LLLRG-KRE- VERGQV---P

Sc EF-Tu1     GSVTPHTEFE AQAYILSKDE GGRHTPFFNN YRPQFYFRTT DVTGVVTLPE  349
Sr EF-Tu1                                                       H     349
Sc EF-Tu3        V ARR R  RV V  AR     S  LTTG       I  A   V D D G   346
Sr EF-Tu3        V RSR S   V V  AR     T  VTSG       I  A   V D D G   343
Sr EF-Tu2           Q                     E                       K   349
Ec EF-TuA      TIK   K    SEV             KG                  TIE     346
Consensus     GSV-PHT-FE AQ-YILSKDE GGRHTPFF-- YRPQFYFRTT DVTG-V-LPE

Sc EF-Tu1     GTEMVMPGDN TEMKVELIQP VAMEEGLKFA IREGGRTVGA GQVTKINK    397
Sr EF-Tu1                R                                    V      397
Sc EF-Tu3     E.AVAR   T VT T   GRD  PL T  G                T  AVE.   392
Sr EF-Tu3     V.GVAR  ET VS I   GRE  PL P  G                T  ALV.   389
Sr EF-Tu2                A H Q      I                          R V     397
Ec EF-TuA      V         IK V T  H  I  DD  R                V A VLG   394
Consensus     G-EMVMPGDN --M-VELI-P VAME-GL-FA IREGGRTVGA G-VT----
```

Table 1
Nucleotide sequence identities between the *tuf* genes of *S. coelicolor*, *S. ramocissimus* and *E. coli* (above the diagonal) and amino acid identities between their deduced gene products (below the diagonal)

	Sc 1	Sc 3	Sr 1	Sr 2	Sr 3	Ec A
Sc *tuf1*, EF-Tu1		69	94	84	70	71
Sc *tuf3*, EF-Tu3	63		69	70	87	61
Sr *tuf1*, EF-Tu1	96	63		85	70	71
Sr *tuf2*, EF-Tu2	89	64	88		70	69
Sr *tuf3*, EF-Tu3	64	91	65	64		63
Ec *tufA*, EF-TuA	75	60	74	72	60	

All values are given in percentages. Abbreviations: Sc, *S. coelicolor*; Sr, *S. ramocissimus*; and Ec, *E. coli*. Alignments were done with the program 'Gap' [13].

shows almost as much amino acid identity with *E. coli* EF-Tu as with *S. coelicolor* EF-Tu1.

A surprising observation is that the nucleotide sequences of *tuf1* and *tuf3* are more homologous to each other than the deduced amino acid sequences (69% versus 63%), which is also observed for the *S. ramocissimus tuf1* and *tuf3* genes and their gene products (Table 1). This is due to nucleotide substitutions in the *tuf3* gene at the first and second codon position rather than the third ('wobble') position, so that many nucleotide substitutions lead to amino acid substitutions. Still, amino acids in the GTP-binding domains are conserved, suggesting that the *tuf3* gene product also belongs to the family of GTP-binding proteins.

Since it has been demonstrated for *S. ramocissimus* EF-Tu1 that it functions as an elongation factor [4] we assume on the basis of the very high similarity of the proteins that such is also the case for *S. coelicolor* EF-Tu1 and no further investigation to address this point has been performed. The role of EF-Tu3 is unclear and is presently being investigated in our laboratory.

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Fig. 3. Amino acid alignment of *Streptomyces* EF-Tus and *E. coli* EF-TuA. Abbreviations: *Ec*, *E. coli*; *Sc*, *S. coelicolor*; *Sr*, *S. ramocissimus*. Numbers at the right of the figure refer to the amino acid positions. A consensus (grey shaded) is given when the amino acid alignment shows more than three identical amino acids. The amino acids (of Sc EF-Tu1) that constitute the GTP binding consensus sequence are shown in bold face.