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Abstract

This paper reports sequence features within nuclear genes from *Sordaria macrospora*. Eight nuclear gene sequences were analyzed for codon usage, GC content, intron regulatory sequences and translation initiation sites.

Sequence characteristics within nuclear genes from Sordaria macrospora

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This paper reports sequence features within nuclear genes from *Sordaria macrospora*. Eight nuclear gene sequences were analyzed for codon usage, GC content, intron regulatory sequences and translation initiation sites.

The homothallic ascomycete *Sordaria macrospora* is an excellent model system to study not only meiotic pairing and recombination (Zickler 1977 Chromosoma **61**:29-316) but also fruiting body development (Esser and Straub 1958 Z. Vererbungslehre **89**:729-746). Recently, these studies have been extended to a molecular level (Walz and Kück 1995 Curr. Genet. **29**:88-95) and knowledge about sequence features would be a helpful tool in sequence analysis. Until now, sequence information from *S. macrospora* was only available from a single nuclear gene (LeChevanton and Leblon 1989 Gene **77**:39-49). Here we compile sequence data from eight recently sequenced genes to determine common features of nuclear genes from *S. macrospora*. We provide a consensus sequence for the translation initiation site (Table 1), a codon usage table (Table 2), and consensus sequence features from the well studied ascomycete *Neurospora crassa* (data taken from Brucherez *et al.* 1993 Fungal Genet. Newsl. **40**:85-95; and Edelman and Staben 1994 Exp Mycol **18**:70-81) shows that *S. macrospora* sequence characteristics are very similar to those determined for *N. crassa* genes.

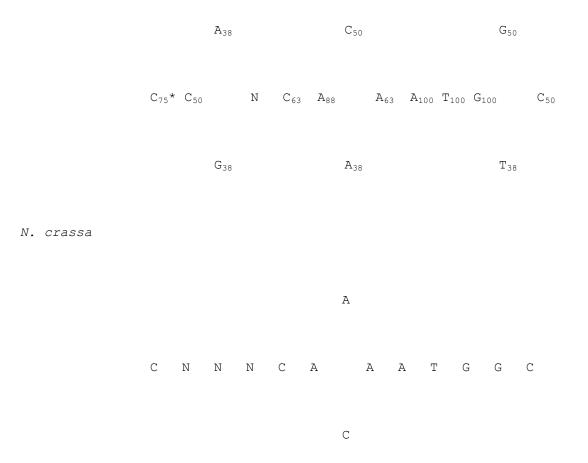
Table 1. Translation initiation context

Gene	Gene product	Translation initiation	Reference ^a
EF1-	EF1- translation elongation factor	CCGTCAAAATGGG	1
tuba	-tubulin	CATACAAAATGCG	2
ura3	orotidine phosphoribosyl transferase	CCGCCACAATGTC	3
ura5	orotidine monophosphate decarboxylase	CCAGCACAATGGC	4
SmtA-1	mating-type protein	GAAGTACGATGTC	5
SmtA-2	mating-type protein	CGACTGACATGGA	5
SmtA-3	mating-type protein	CTTTCAGCATGTC	6
Smta-1	mating-type protein	TCGAAACAATGGA	5

^a (1)Gagny, Koll and Silar, unpublished (Accession # X96615) (2) Pöggeler *et al.*, submitted (Accession # Z70290) (3) Nouwrousian, unpublished (Accession # Z70291) (4) Nouwrousian, unpublished (5) Pöggeler *et al.*, submitted (Accession # Y10616) (6) Pöggeler, unpublished

Consensus translation initiation

S. macrospora



*The subscript number indicates the percentage occurrence of the particular nucleotide.

The *S. macrospora* consensus for initiation of translation shows a high degree of identity to the *N. crassa* translation initiation consensus sequence and, as *N. crassa*, a prevalence of GC following the ATG which means that an alanine (GCN) is found at the amino terminus of most proteins studied so far.

Table 2. Codon usage analysis based upon 2497 codons

TTT-Phe 24(25.0%) ^a 3(8.6%)	TCT-Ser 22(16.4%)	TAT-Tyr	21(26.9%)	TGT-Cys
TTC-Phe 72(75.0%) 32(91.4%)	TCC-Ser 42(31.3%)	TAC-Tyr	57(73.1%)	TGC-Cys
TTA-Leu 3(1.7%) 1(14.3%)	TCA-Ser 12(9.0%)	TAA-Ter	4(57.1%)	TGA-Ter
TTG-Leu 21(11.6%)	TCG-Ser 28(20.9%)	TAG-Ter	2(28.6%)	TGG-Trp
32(100.0%) CTT-Leu 45(24.9%)	CCT-Pro 41(30.6%)	CAT-His	23(30.7%)	CGT-Arg
34(27.6%) CTC-Leu 80(44.2%) 57(46.3%)	CCC-Pro 68(50.7%)	CAC-His	52(69.3%)	CGC-Arg
J/(10.J0)				

CTA-Leu 3(1.7%) 7(5.7%)	CCA-Pro	13(9.7%)	CAA-Gln 24(23.8%)	CGA-Arg
CTG-Leu 29(16.0%) 6(4.9%)	CCG-Pro	12(9.0%)	CAG-Gln 77(76.2%)	CGG-Arg
ATT-Ile 47(32.6%) 5(3.7%)	ACT-Thr	25(19.2%)	AAT-Asn 17(17.3%)	AGT-Ser
ATC-Ile 95(66.0%) 25(18.7%)	ACC-Thr	71(54.6%)	AAC-Asn 81(82.7%)	AGC-Ser
ATA-Ile 2(1.4%) 7(5.7%)	ACA-Thr	15(11.5%)	AAA-Lys 12(7.2%)	AGA-Arg
ATG-Met 63(100.0%) 12(9.8%)	ACG-Thr	19(14.6%)	AAG-Lys 154(92.8%)	AGG-Arg
GTT-Val 40(24.2%) 61(32.3%)	GCT-Ala	70(30.7%)	GAT-Asp 61(42.4%)	GGT-Gly
GTC-Val 101(61.2%) 94(49.7%)	GCC-Ala 1	15(50.4%)	GAC-Asp 83(57.6%)	GGC-Gly
GTA-Val 5(3.0%) 24(12.7%)	GCA-Ala	21(9.2%)	GAA-Glu 33(19.0%)	GGA-Gly
GTG-Val 19(11.5%) 10(5.3%)	GCG-Ala	22(9.6%)	GAG-Glu 141(81.0%)	GGG-Gly

^aThe percent shown by each codon represents the percent of the time that the amino acid is encoded by that codon.

The GC content in a coding region of 7491 nucleotides is 56.7%. For comparison in *N. crassa* the GC content is 58.6% in the coding region (GC content in total DNA 54.1%). In cases where amino acids are represented by more than one codon, *S. macrospora*, as many other organisms, does not use synonym codons equally (Table 2).

In *S. macrospora*, as in *N. crassa*, codons are preferred with a C in the third position and in four codon families the codon ending in T is usually preferred to those ending in A or G. The stop codon TAA is more frequently used than TAG or TGA, respectively. The six least used codons for *S. macrospora* are ATA (Ile), TTA (Leu), CTA (Leu), TGT (Cys), GTA (Val), and AGT (Ser). All of these six codons are belonging to low-usage codons in *N. crassa* as well. As reported by Zhang *et al.* (1991 Gene **105**:61-67) in many organisms, low-usage codons are clearly avoided in abundant proteins and therefore may affect translation rates.

Table 3. Intron regulatory sequences and intron length

Intron	5' Intron	Branch	Distance to	3' Intron	Intron
	Donor	Site	3' Splice-Site/nt ^b	Acceptor	Length / bp
SmtA-1/1 ^a	T^GTAAGT	ACTGATT	-19-	TTCAG ²	58
$SmtA-1/2^{a}$	G^GTTAGT	ACTCGTG	-21-	GGCAG	60
SmtA-2/1 ^a	G^GTAACA	ACTGATG	-14-	GCCAG	57
SmtA-2/2 ^a	G^GTGAGT	ACTGACA	-12-	GATAG ²	71
SmtA-2/3 ^a	T^GTAAGA	ACTAATA	-12-	GACAG ²	47
SmtA-2/4 ^a	G^GTTTGC	GCTAACA	-16-	GACAG ²	55

SmtA-3/1 ^a	C^GTGAGT	ACTGACT	-12-	GTTAG^	54
Smta-1/1 ^a	A^GTAAGT	ACTGACC	-15-	TTTAG^	53
$Smta-1/2^{a}$	T^GTAGGT	ACTAACC	-12-	CTTAG^	57
tuba/1	G^GTACGT	GCTAACG	-22-	TCTAG^	256
tuba/2	G^GTAGGT	GCTAACC	-15-	ATTAG^	149
tuba/3	G^GTAAGC	GCTAACC	-17-	TACAG^	80
tuba/4	G^GTACAT	GCTTACA	-18-	CACAG^	60
tuba/5	G^GTATGT	ACTAACT	-16-	CTTAG^	64
tuba/6	T^GTAAGT	GCTAACT	-14-	CCTAG^	57
ef1/1	G^GTAATG	GCTAACG	-14-	AACAG^	100
ef1/2	G^GTTAGT	ACTGACT	-15-	AACAG^	243
ef1/3	G^GTATGT	GCTAACT	-17-	AAAAG^	60

^a positions of intron splice sites inferred from cDNA sequences

^b distance between the C of the intron branch point and the G of the 3' intron acceptor

Consensus 5' Intron-Donor

S.	macrospora	G ₆₇ ^	G ₁₀₀	T_{100}	A ₇₂	A ₆₁	G ₈₃	T_{72}
N.	crassa	G^	G	Т	A	A	G	Т

Consensus Intron Branch Site

S. macrospora	A ₅₆ G ₄₄	C ₁₀₀	A T ₁₀₀ G ₃₃	•56 A ₉₄ 3	C ₇₈	Ν
N. crassa	A G	С	A T G	A	C A	С

Consensus 3' Intron-Acceptor

S	macrospora	G ₃₃	A ₃₉	C ₅₆	A ₁₀₀	G100
b. macrospera	A_{27}	Τ ₃₉	\mathbb{T}_{44}	1100	0100	
77	ar2442	A	A	Т	7	G
10.	crassa	Т	Т	С	A	G

In *S. macrospora* genes the intron length lies between 47 bp and 256 bp, the average length is 88 bp and the median length is 60 bp. Intron length in *N. crassa* ranges from 46 to 856 bp with a tendency toward 60 to 70 bp. Among the eight genes analyzed so far, two genes, *ura3* and *ura5*, do not contain introns. In *S. macrospora* introns the distance from the C of the splice branch site to the G of the 3' splice site is between 12 nt and 22 nt. This distance varies in *N. crassa* from 14 to 30 nucleotides. The *S. macrospora* intron signals (5' donor site, intron branch site and 3' intron acceptor site) are very similar to the *N. crassa* intron consensus sequences.

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