Supplemental Data to

CAENORHABDITIS ELEGANS EVOLVES A NEW ARCHITECTURE FOR THE MULTI-AMINOACYL-tRNA SYNTHETASE COMPLEX.

Svitlana Havrylenko[1,2], Renaud Legouis[3], Boris Negrutskii[2], and Marc Mirande[1]

Supplemental Table 1, and Figures S1-S2

Supplemental Table 1

Transgenic strains generated in this study

	Name	Genotype
	RD125	*unc119(ed3); Is[unc119(+); pprs-1::ProRS-Ce::TAP]*
	RD126	*unc119(ed3); Is[unc119(+); pmrs-1::MetRSΔC-Ce::HA]*
	RD159	*unc119(ed3); Is[unc119(+); peft-3::MetRSΔC-Ce::HA]*
	RD160	*unc119(ed3); Is[unc119(+); peft-3::MetRSΔCC-Ce::Cp43::HA]*
	RD168	*unc119(ed3); Is[unc119(+); peft-3::MetRSΔNΔC-Hs::C-Ce]*
	RD169	*unc119(ed3); Is[unc119(+); peft-3::MetRSΔNC-Ce::Np43]*

Plasmids used in this study

Name	Vector backbone	Insert	Reference
pME18S-FL3		*mrs-1* cDNA (YK1416a08)	Y. Kohara
pDONR201			Invitrogen
pSB_GW::TAG			Polanowska et al. 2004
pSB_GW::TAP	pSB_GW::TAG	TAP cassette	This study
	pEGFP-N1	p43-Hs cDNA	Shalak et al. 2007
	pUC18	MetRS-Hs cDNA	Kaminska et al. 2001

Expression vectors

Name	Vector backbone	Insert	Reference
pSB_PRS-Ce_TAP	pSB_GW::TAP	*pprs-1::prs-1*	This study
pSB_MRSΔC-Ce	pSB_GW::TAG	*pmrs-1::mrs-1Δ(596-917)::HA*	This study
pSB_eMRSΔC-Ce	pSB_GW::TAG	*peft-3::mrs-1Δ(596-917)::HA*	This study
pSB_eMRSΔCC-Ce:Cp43	pSB_GW::TAG	*peft-3::mrs-1Δ(750-917)::p43-Hs(147-312)::HA*	This study
pSB_eMRSΔNC-Ce:Np43	pSB_GW::TAG	*peft-3::mrs-1Δ(596-917)::p43-Hs(1-144)::mrs-1(750-917)*	This study
pSB_eMRSΔNΔC-Hs:C-Ce	pSB_GW::TAG	*peft-3::MetRS-Hs(215-823)::mrs-1(596-917)*	This study

RNAi vectors

	Vector backbone	Insert	Reference
	L4440		Timmons et al. 2001
	L4440	*CAT* (WBRNAi00008897)	Kamath and Ahringer 2003
	L4440	*C* (1876-2769nt *mrs-1* mRNA)	This study
	L4440	*NC* (1876-2265 nt *mrs-1* mRNA)	This study
	L4440	*CC* (2278-2769 nt *mrs-1* mRNA)	This study

Supplemental Data

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Legends to Supplementary Figures

Figure S1. Expression of the mutant allele *mrsp-38(ok2139)*. The 1252 nt deletion encompasses the 3'-end of *mrsp-38* and the 5'-end of *dcap-1*. Total mRNA was isolated from the mutant strain and RT-PCR was performed between oligonucleotides 01 and 02 to assess that mRNA is expressed (RT-PCR product of 359 bp expected), and between oligonucleotides 01 and 03 to check whether the remaining part of exon III from *dcap-1* is spliced or not. If exon III' would be spliced, the size of the RT-PCR product obtained with oligonucleotides 01 et 03 would be of 469 bp, instead of 774 bp if this exon is present in mRNA. RT-PCR products were analyzed on an agarose gel. Exon III' is not spliced, and the first stop codon in frame with the ATG of *mrsp-38* is located near the 5'-end of this exon. M: size marker.

Figure S2. Expression of ectopic MetRS in the transgenic worms. Extracts from wild-type strain (WT) and from transgenic worms expressing MetRSΔC-Ce (RD159), MetRSΔCC-Ce::Cp43 (RD160) or MetRSΔNC-Ce::Np43 (RD169) were analyzed by immunoblotting with anti-MetRSΔC-Ce and anti-tubulin antibodies.

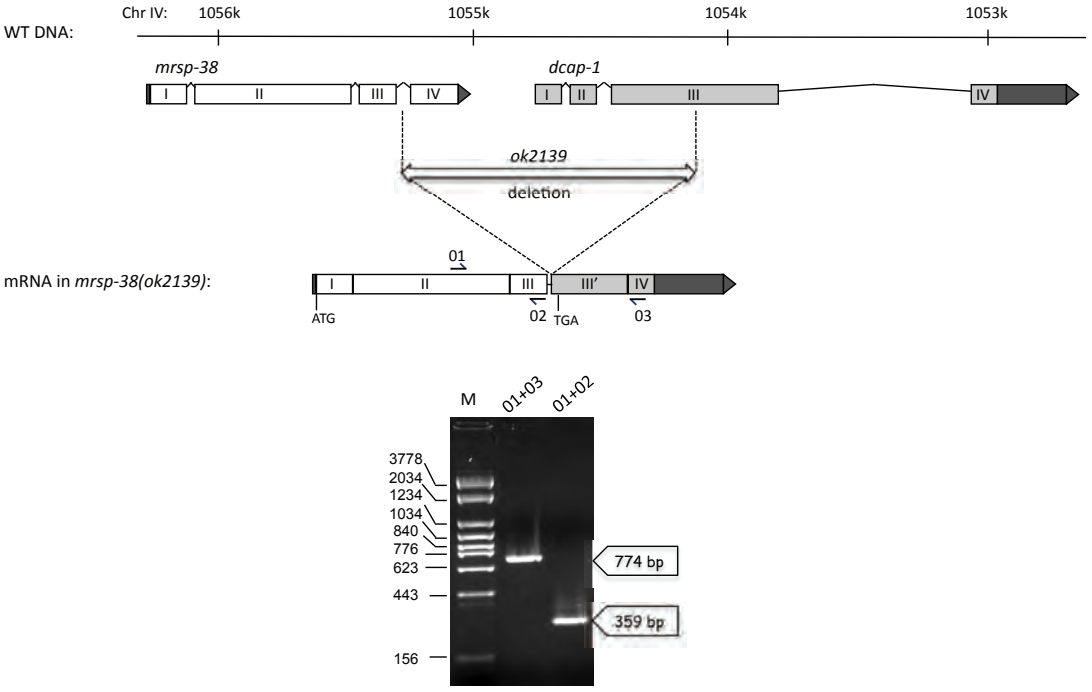
WT DNA:
Chr IV:
1056k
1055k
1054k
1053k
mrsp-38
dcap-1
ok2139
deletion
mRNA in mrsp-38(ok2139):
01
02
03
ATG
TGA
M
01+03
01+02
3778
2034
1234
1034
840
776
623
443
156
774 bp
359 bp

Fig. S1

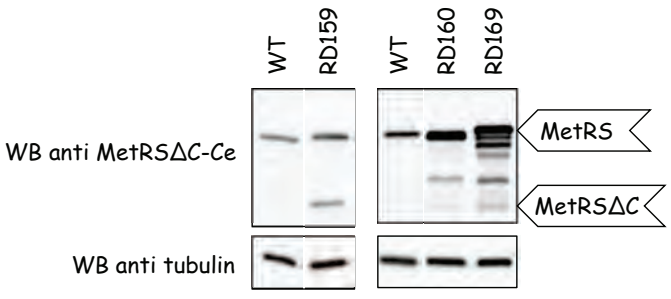
WT
RD159
WT
RD160
RD169
WB anti MetRSΔC-Ce
MetRS
MetRSΔC
WB anti tubulin

Fig. S2