

Supplementary table 1. Sequence identity (%) at the amino acid level between different RaCIs

	RaCI2	RaCI3	RaCI6	RaCI7	RaCI4	RaCI1	RaCI5
RaCI2	100	41	38	42	40	42	38
RaCI3		100	71	73	44	35	36
RaCI6			100	76	47	36	38
RaCI7				100	45	35	37
RaCI4					100	38	35
RaCI1						100	40
RaCI5							100

Supplementary Table 2 Plasmids, primers and synthetic genes used in this study

Plasmid	Description	Restriction sites	Primers	Expression
pEpreS2-2	From Expres2ion Biotechnologies, Denmark			<i>Drosophila melanogaster</i> S2
pMJ41	Modified pExpres2-2 vector; Kozak-BIP-His6-NgoMIV	Acc65I/EcoRI	MJ41 + MJ42 (annealed and inserted)	
pMJ80	pMJ41 His6-RaCI1	NgoMIV/NotI		
pMJ85	pExpres2-2 RaCI2 (native signal peptide)	EcoRI/NotI		
pMJ81	pExpres2-2 RaCI3 (native signal peptide)	EcoRI/NotI		
pMJ82	pExpres2-2 RaCI4 (native signal peptide)	EcoRI/NotI		
pMJ84	pExpres2-2 RaCI5 (native signal peptide)	EcoRI/NotI		
pMJ83	pExpres2-2 RaCI7 (native signal peptide)	EcoRI/NotI		
pMJ100	pETM14 RaCI1 (no signal peptide)	NcoI/NotI		<i>Escherichia coli</i>
pMJ99	pETM14 RaCI2 (no signal peptide)	NcoI/NotI		
pMJ97	pETM14 RaCI3 (no signal peptide)	BspHI/NotI		
pMJ98	pETM14 RaCI4 (no signal peptide)	NcoI/NotI		
pMJ111	pETM14 - RaCI2 ΔC10 (for NMR experiments)		MJ127 + MJ128 (template = pMJ99)	
pMJ103	pETM14 - RaCI2 ΔN10		MJ87 + MJ88 (template = pMJ99)	
pMJ104	pETM14 - RaCI2 ΔC18		MJ149 + MJ150 (template pMJ99)	
pMJ15	pKLAC2-His10-OmCI wildtype ¹		MJ6 + MJ8	<i>Kluyveromyces lactis</i>
pMJ19	pKLAC2-His10-OmCI N78Q		MJ20 + MJ21 (template = pMJ15)	
pMJ27	pKLAC2-His10-OmCI N78Q/N102Q		MJ22 + MJ23 (template = pMJ19)	

Primer	Sequence	Description
MJ6	GCGCGGCGGCCGCCTAGCAGTCCTTGAGATGGGGAT	OmCI + stopcodon + NotI (rv)
MJ8	GGCCCCCTCGAGAAAAGAATGGCTAGCCATCACCATCACCATCACCATCACCATCACTCCGGAGACAGCGAAAGCGACTGCACTG	XhoI + His10 + OmCI (fw)
MJ20	CGGTGATGATGACGTTTAAGCAAGGCACAGACTGGGCTTCAACCG	OmCI - N78Q (fw)
MJ21	CGGTTGAAGCCCAGTCTGTGCCCTTGCTTAAACGTATCATCACCG	OmCI - N78Q (rv)
MJ22	GGTAACGGCAACCCTTGGTCAACTAACCCTAAATAGGGAAGTGG	OmCI - N102Q (fw)
MJ23	CCACTTCCCTATTTGGGTTAGTTGACCAAGGGTTGCCGTTACC	OmCI - N102Q (rv)
MJ41	GTACCGCCACCATGAAGCTGTGCATCCTGCTGGCCGTGGTGGCCTTCGTGGGACTGAGCCTGGGACACCACCACCATCACCACGCCGGCG	Acc65I-Kozak-BIP-His6-NgoMIV-EcoRI
MJ42	AATTGCGCGGCGTGGTGATGGTGGTGGTGTCCCAGGCTCAGTCCACGAAGGCCACCACGGCCAGCAGGATGCACAGCTTCATGGTGGCG	Acc65I-Kozak-BIP-His6-NgoMIV-EcoRI

MJ87	CTGTTCCAGGGGCCCATGAAAGATCAGTGTGCAAATG	RaCl2 ΔN10 (fw)
MJ88	CATTTGCACACTGATCTTTTCATGGGCCCTGGAACAG	RaCl2 ΔN10 (rv)
MJ127	CACCGAAAAGCACCAACACCT TA ACCGAACAGAGCTTTAATATG	RaCl2 ΔC10 (fw)
MJ128	CATATTAAAGCTCTGTTCCGGT TTAG GTGGTGGTGTCTTTTCGGTG	RaCl2 ΔC10 (rv)
MJ149	CACCTGTTATCTGCTGGC ATAA ACCGGAAAAGCACCAAC	RaCl2 ΔC18 (fw)
MJ150	GGTGGTGTCTTTTCGGT TTAT GCCAGCAGATAACAGGTG	RaCl2 ΔC18 (rv)

Synthetic gene ²	Sequence
RaCl1 (pMJ80)	CGAATTGGCGGAAGGCCGTCAAGGCCACGTGTCTTGTCCAGAGCTCGCCGGCGAGGAAGTGAAGACCACCCCATCCCCAACCCACAGTGCGTGAACGCCACGTGCGAGCGCAAGCTGGATGCCCTGGGCAATGCCGTGATCACCAAGTGCCACAGGGCTGCCTGTGCGTGTGCGCGGAGCCAGCAATATCGTGCCCGCAACGGAACTGCTTCCAGCTGGCCACCACCAAGCCACCAATGGCCCCAGGCCATAACAAGGACAACAAGGAGGAGGTCCAAGTGA <u>CGCGCCGCGGT</u> ACCTGGAGCACAAGACTGGCCTCATGGGCCCTCCGCTCACTGC
RaCl2 (pMJ85)	CGAATTGAAGGAAGGCCGTCAAGGCCACGTGTCTTGTCCAGAGCTCGAATTCGCCACCATGAACGCCGTGACCGTGTGGCCTTACCGCCTTCGCCCTGATTGTGCACGATTGTACAGCGAGGAGGCCAACACCACCCCATCAGCGTGAAGGATCAGTGCGCCAAACGTGACCTGCCGCGCACCGTGGATAATCGCGGGCAAGCGCCACATCGATGGCTGCCACCAGGATGCCTGTGCGTGTGAAGGGCCCCGATAGCAAGGATAACCTGGATGGCACGTGTACCTGTGGCCACCACCCCAAAGAGCACCACCACCACTACCGAGCAGAGCTTCAACATGGAGGAGTAGCGGCGCGGTACCTGGAGCACAAGACTGGCCTCATGGGCCCTCCTTTCACTGC
RaCl3 (pMJ81)	CGAATTGGCGGAAGGCCGTCAAGGCCACGTGTCTTGTCCAGAGCTCGAATTCGCCACCATGGCCGCCCTGAATGGCCTGGTGTGCTGCTGCTGACCATCAGCGCCATGTTTCATCAGCGAGTGCTACAGCAGCGGCGAGAGCCAGAGCATTCAGCGCAAGGGACAGTGCGAGGAAGTGATCTGCCACCGCAAGCTGAACCACTGGGAGAGCGCGTGACCAAGCGGATGCCAACCGGATGCCTGTGCGTGATCCGCGAGCCCGATAACGTGGACAACGCCAACGGAACTGCTACGCCCTGATGAGCAGCACCACACGACCACGACGACCCAGATGGAAACCACCACTCCGAAGAGGAGGAGTAGCGGCGCGGTACCTGGAGCACAAGACTGGCCTCATGGGCCCTCCGCTCACTGC
RaCl4 (pMJ82)	CGAATTGGCGGAAGGCCGTCAAGGCCACGTGTCTTGTCCAGAGCTCGAATTCGCCACCATGAGCGCCTTCAACATCTTCGCCCTGGTGTGGTGTGTCGCCCTGATGATCAACGAGTGCTGCACCAGCCAGGAGCCACCACCCCACTGAAGGCCGCCAGTCAGTGACGCAACGTGAAGTGCCGCCGCCCTTCGATCACCTGGGCAATAGTGTGACCGAGGGGTGCCAAAGCGGATGCCTGTGCGTGATACAGGCCACCGGTACAACCAGGAGGCCAACGGAACTGCTACGAGCTGATGAAGACCTCGACCACCACGACGACCGAGGGAAACCCAGCCCGATAGCGGCGCGGTACCTGGAGCACAAGACTGGCCTCATGGGCCCTCCGCTCACTGC
RaCl5 (pMJ84)	CGAATTGGCGGAAGGCCGTCAAGGCCACGTGTCTTGTCCAGAGCTCGAATTCGCCACCATGAACGCCGTGATCGTGTGCGTGACCATCAGCGCCGTGCTGATCAACCAAGTGCTACAGTACCGCCAGCCACCTGAGCATCAACGGCGGCCGATATGTCATCGAGAAGACCTGCAACCGCTCCATCGATGCCGCCGGAAGAAAGTATCGCCGGATGCCAGGCGGCTGCCTGTGCGTGTTCACGTGTCCGATGTGACCTACCCCGCAACGGCACGTGTATCAGCTGGCCACCACCACGACCAATCGCCAGGCGCCGTGATGGAACCGAGCGCTAAGCGGCCGCGGTACCTGGAGCACAAGACTGGCCTCATGGGCCCTCCGCTCACTGC
RaCl7 (pMJ83)	CGAATTGGCGGAAGGCCGTCAAGGCCACGTGTCTTGTCCAGAGCTCGAATTCGCCACCATGGCCGCCCTGAATGGCCTGGTGTGCTGCTGCTGACCATCAGCGCCATGTTTCATCAGCGAGTGCTACAGCAGCGGCGAGAGCCAGAGCATTCAGCGCAAGGGACAGTGCGAGGAAGTGACCTGCCACCGCACCTGAATCACCTGGGAGTGGCCGTGACCAAGCGGATGCCAAAGTGATGCCTGTGCGTGATCAGCGCCCGATAGCGCCGTGAACGTGAACGGCACGTGTACCACTGATGGGCTCCACCAGTACCACCACGACGACCCCCAAGCAGCGAGGATCAGGAGTAGCGGCGCGGTACCTGGAGCACAAGACTGGCCTCATGGGCCCTCCGCTCACTGC
RaCl1 (pMJ100)	CGAATTGAAGGAAGGCCGTCAAGGCCACGTGTCTTGTCCAGAGCTCGCATGGAAGAAGTTAAACCACCCGATTCCGAATCATCATGTGTGTTAATGCAACCTGTGAACGTAAACTGGATGCACCTGGGTAAATGCAGTTATTACCAATGTCCGCAGGGTGTCTGTGTGTTGTTCTGGTGGTCAAGCAATATTGTTCCGGCAATGGCACCTGTTTTCAGCTGGCAACCACCAACCGCCTATGGCACCGGGTGATAATAAGATAACAAAGAAGAAGAAAGCAATTAAGCGGCCGCGGTACCTGGAGCACAAGACTGGCCTCATGGGCCCTCCTTTCACTGC
RaCl2 (pMJ99)	GGCCCCCATGGAGGAAGCAAAATACCACCCGATTAGCGTTAAAGATCAGTGTGCAAAATGTTACCTGTGCTGCTGATGATAATCGTGGTAAACGTCATATTGATGGTGTCCGCTGGTGTGCTGTGTGTTCTGAAAGGTCCGGATAGCAAAAGATAATCTGGATGGCACCTGTTATCTGCTGGCAACCACACCGAAAAGCACCACCACAGTACCGAACAGAGCTTTAATATGGAAGAGTAAGCGGCCGCGCGC
RaCl3 (pMJ97)	GGCCCTCATGAGCGGTGAAAGCCAGAGCATTACGCGTAAAGGTCAGTGTGAAGAAGTTATCTGTCATCGTAAACTGAATCATCTGGTGAACGTGTACCAAGCGTTGTCCGACCGGTTGTCTGTGTGTTATTCTGTGAACCGGATAATGTGGATAATGCAATGGCACCTGTTATGCACTGATGAGCAGCACCACCAACCACACGACACCGGATGGTACAACCACCTCTGAAGAAGAAGATAAGCGGCCGCGCGC
RaCl4 (pMJ98)	GGCCCCCATGGCACAAGAACCAGCACACCGCTGAAAGCAGCAAGCCAGTGTAGCAATGTTAAATGTGCTGCTGCTTTTGATCATCTGGGTAAATAGCGTTACCGAAGGTTGTCCGAGCGGTTGTCTGTGTGTTTATCAGGCAACCGGTTATAATCAAGAAGCAACGGCACCTGTTATGAGCTGATGAAACCAGTACCACCACCAACCAGAAAGGTACACCGGCACAGTAAGCGGCCGCGCGC

¹ Amplified from pMETαC-OmCl (Nunn, M.A. et al., *J. Immunol* **174**, 2084-2091, 2005).

² Synthetic fragments were subcloned into constructs that are indicated between brackets. Restriction sites are underlined.

Mutations introduced with primers are in bold.
Fw = forward primer, rv = reverse primer.