

Table S1. List of PCR primers used in this study.

Name	Prime sequence (5'-3')	Notes
Construction of expression vector		
Ptef-F	GGCTGACTTGAAGTAATCTCTGCAGATCTCATGTACCTTGACGTCCTCCGAG	Cloning of <i>Ptef</i>
Ptef-R	TGTTTCAGGATCTCCTTGAGCTCGCCATTCTGAAGAACGAAACTGGCGACT	Cloning of <i>Ptef</i>
Spmae-F	CGCAAGTCGCCAGTTTCGTTCTTCAGAATGGGCGAGCTCAAGGAGATC	Cloning of <i>Spmae</i>
Spmae-R	TGATGATTTTCAGTAACGTTAAGTGGATCCGAATTCGATTCAGAGGCTGTCGTGGTCCGA	Cloning of <i>Spmae</i>
gpdA-F	ACTTGAAGTAATCTCTGCAGATCTTAAATTAACTCGAGTGACGGTGCTTTTCACCTCTC	Cloning of <i>PgpdA</i>
gpdA-R	TGATTTAGTAACGTTAAGTGGATCCGAATTCGATATCGTTTAAACACTAGTTTGGATTTC TGTGATGTGGG	Cloning of <i>PgpdA</i>
Rofum-F	ACTCACCTCCCACATCACAGAAATCAAACTAGTATGAACAACCTCTCCTCGTCT	Cloning of <i>Rofum</i>
Rofum-R	TAACGTTAAGTGGATCCGAATTCGATATCGTTTAAACTTAATCCTTGGCAGAGATCAT	Cloning of <i>Rofum</i>
Ecfum-F	CTCCCCACATCACAGAAATCAAACTAGTATGAATACAGTACG	Cloning of <i>Ecfum</i>
Ecfum-R	GATCCGAATTCGATATCGTTTAAACTTAACGCCCGGCTTTCATA	Cloning of <i>Ecfum</i>
Ckfum-F	CTCCCCACATCACAGAAATCAAACTAGTACTAGTATGCTGGCGGC	Cloning of <i>Ckfum</i>
Ckfum-R	GATCCGAATTCGATATCGTTTAAACGTTTAAACTCAATCCTTCGGGC	Cloning of <i>Ckfum</i>
Msfum-F	CCTCCCCACATCACAGAAATCAAACTAGTATGACAGCGTTTCGTATTGAA	Cloning of <i>Msfum</i>
Msfum-R	GTTAAGTGGATCCGAATTCGATATCGTTTAAACTTATTTCAGCTACCAACCA	Cloning of <i>Msfum</i>
Peif-F	GTTTGCAGTTGGCTGACTTGAAGTAATCTCTGCAGATCTCACAGCAGTTCGCACGCTCC	Cloning of <i>Peif</i>
Peif-R	TCGGCTTCTTGCCGCTTTTGCCAACCTTTGTGCCATCTTGTGTTGTTGTGTTG	Cloning of <i>Peif</i>
Mtsfc-F	TCACCGTTACGCTCCTCAACACAACAACAACAAGATGGCGACAAAAGTTGGCAA	Cloning of <i>Mtsfc</i>
Mtsfc-R	CAAGCTGTTTGATGATTTTCAGTAACGTTAAGTGGATCCTCACTCCTCGTACCTCCAC	Cloning of <i>Mtsfc</i>
Construction of sgRNA expressing plasmids		
U6p-F	AGGATCGGTGGAGTGAAGTTCGGAA	Cloning of U6 promotor
U6p-fr1-R	CTCTAAAACCGCTACGATGATGACGGGCGAGGAAAGAAAAGAAAGAGAGGAG	Cloning of U6 promotor
U6p-fr2-R	TAGCTCTAAAAGTCTTGTCAAGGACTGTGACGAGGAAAGAAAAGAAAAGAAG	Cloning of U6 promotor
U6p-Mtfum-R	GCTCTAAAACAGTATTTGTCGGCCGGCACCGAGGAAAGAAAAGAAA	Cloning of U6 promotor
U6p-moc-R	TCTAGCTCTAAAAGTATCCGAGTGTCTCTGACGAGGAAAGAAAAGAAAAGAAGA	Cloning of U6 promotor
U6p-Mtmae-R	TTCTAGCTCTAAAACCCACGGTAAATGCGGATCCCGAGGAAAGAAAAGAAAAGAGAG	Cloning of U6 promotor
U6p-neo-R	CTAAAACAGTCATAGCCGAATAGCCTCGAGGAAAGAAAAGAAAAGAGAGGAG	
g-fr1-F	CTTTTCTTTCTTCTCCTCGCCGTCATCATCGTAGGCGGTTTTAGAGCTAGAAATAG	<i>fr1</i> -gRNA5'
g-fr2-F	TTTCTTTCTTCTCCTCGTCACAGTCCTTGACAAGCAGTTTTAGAGCTAGAAATAG	<i>fr2</i> -gRNA5'
g-Mtfum-F	CTTTTCTTTCTTCTCCTCGGTGCCGCGACAAATACTGTTTTAGAGCT	<i>Mtfum</i> -gRNA5'
g-moc-F	CTTTTCTTTCTTCTCCTCGTCAGGAACACTCGGATACAGTTTTAGAGCTAGAAATAG	<i>moc</i> -gRNA5'
g-Mtmae-F	CTTTTCTTTCTTCTCCTCGGGATCCGATTTACCGTGGGTTTTAGAGCTAGAAATAGCA	<i>Mtmae</i> -gRNA5'
g-neo-F	CTTTCCTCGAGGCTATTCGGCTATGACTGTTTTAGAGCTAGAAATAGCAAG	<i>neo</i> -gRNA5'
gRNA-R	AAAAAGCACCGACTCGGTGCCACTT	gRNA3'
Donor DNA construction		
neo-F	CGACGTTAACTGATATTGAAGGA	Cloning of <i>PtrpC-neo</i>
neo-R	TCAGAAGAAGCTCGTCAAGAA	Cloning of <i>PtrpC-neo</i>
nat-F	CGACGTTAACTGATATTGAAGGA	Cloning of <i>PtrpC-nat</i>

nat-R	CTATTATCAGGGGCACGGCATG	Cloning of <i>PtrpC-nat</i>
Dfr1-5'-F	GATGTGGAGTGGGCGCTTACACAGTACACGAGGACTTGCTTCTTACTCGACTCAGTGA	<i>fr1</i> -upstream
Dfr1-5'-R	CCCAAAAAATGCTCCTTCAATATCAGTTAACGTCGTCACCTTGTCGGTGTGGATGCCAT	<i>fr1</i> -upstream
Dfr1-3'-F	GCATCGCCTTCTATCGCCTTCTTGACGAGTTCTTCTGAGGCAACTCGGTACCCCGCTTC	<i>fr1</i> -downstream
Dfr1-3'-R	AAGTCATGTGATTGTAATCGACCGACGGAATTGAGGATGTTGTGGATCAGCATCGCCTG	<i>fr1</i> -downstream
Dfr2-5'-F	GATGTGGAGTGGGCGCTTACACAGTACACGAGGACTTTCATATGGTCCATTGACAGTG	<i>fr2</i> -upstream
Dfr2-5'-R	CCCAAAAAATGCTCCTTCAATATCAGTTAACGTCGTCACCGGCGAGATAGATGGTGTG	<i>fr2</i> -upstream
Dfr2-3'-F	GCATCGCCTTCTATCGCCTTCTTGACGAGTTCTTCTGAAGTTCCTGGCATGGCCATCAC	<i>fr2</i> -downstream
Dfr2-3'-R	AAGTCATGTGATTGTAATCGACCGACGGAATTGAGGATGCCTGCTGGAAGAGGTAGTTG	<i>fr2</i> -downstream
DMtfum-5'-F	GTGGAGTGGGCGCTTACACAGTACACGAGGACTTGGAAGTGGCGTGGCATAATTCG	<i>Mtfum</i> -upstream
DMtfum-5'-R	AAAATGCTCCTTCAATATCAGTTAACGTCGTTAGATCTCGCCGAACGCATCGCT	<i>Mtfum</i> -upstream
DMtfum-3'-F	CGCCTTCTATCGCCTTCTTGACGAGTTCTTCTGAAGTGCATCGACCACTTCCCC	<i>Mtfum</i> -downstream
DMtfum-3'-R	CATGTGATTGTAATCGACCGACGGAATTGAGGATGGCCGTTTCATGCCACCGATC	<i>Mtfum</i> -downstream
Dmoc-5'-F	GATGTGGAGTGGGCGCTTACACAGTACACGAGGACTTTGCTTGGGTAGCTACCTATTT	<i>moc</i> -upstream
Dmoc-5'-R	CCCAAAAAATGCTCCTTCAATATCAGTTAACGTCGTCATTGCTGCGGATGATCTTCTG	<i>moc</i> -upstream
Dmoc-3'-F	AGCTCTTACATGAGCATGCCGTGCCCTGATAATAGTCTCTGGCGCCACATTCTCTG	<i>moc</i> -downstream
Dmoc-3'-R	AAGTCATGTGATTGTAATCGACCGACGGAATTGAGGATTCCTCCTCTCACGATGAGGG	<i>moc</i> -downstream
DMtmae-5'-F	GATGTGGAGTGGGCGCTTACACAGTACACGAGGACTTCGATAAGAGCGAGTTATCAGTG	<i>Mtmae</i> -upstream
DMtmae-5'-R	CCAAAAAATGCTCCTTCAATATCAGTTAACGTCGTCAGAAGTAGGGGTGGTTCAGCAG	<i>Mtmae</i> -upstream
DMtmae-3'-F	CGCATCGCCTTCTATCGCCTTCTTGACGAGTTCTTCTGACAGATGACATCGACTACATC	<i>Mtmae</i> -downstream
DMtmae-3'-R	AAGTCATGTGATTGTAATCGACCGACGGAATTGAGGATCACTTCAATCTCGACTTCGTT	<i>Mtmae</i> -downstream
PCR analysis of gene disruption		
fr1-out-F	TACCCACGGTTGCCGAATCTT	Detecting <i>fr1</i> disruption
fr1-out-R	GGTCCCCCAGGTGATGTGGAA	Detecting <i>fr1</i> disruption
fr2-out-F	AACTTCGCGGAGTGTGTCAT	Detecting <i>fr2</i> disruption
fr2-out-R	CGCGAGCCTTCTTGATGATCTT	Detecting <i>fr2</i> disruption
moc-out-F	CGACTGCTTCCAGAAGATCAT	Detecting <i>moc</i> disruption
moc-out-R	AGCAATTGCCGAACCTGGAA	Detecting <i>moc</i> disruption
Mtmae-out-F	GGATTCCCAACACGACAACAA	Detecting <i>Mtmae</i> disruption
Mtmae-out-R	ATCGTTCAGAAGTGCCTCGTT	Detecting <i>Mtmae</i> disruption
Mtfum-out-F	AGGATGGCAGCGAACACGAGAA	Detecting <i>Mtfum</i> disruption
Mtfum-out-R	CTCGATCGCGGTTGCTGAT	Detecting <i>Mtfum</i> disruption
RT-qPCR analysis of copy number		
Spmae-RT-F	GTCTACCTCTGCTGTTCCG	Detecting <i>Spmae</i>
Spmae-RT-R	AGCACCACGAGCAATGTTAATG	Detecting <i>Spmae</i>
Msfum-RT-F	CAATGCCAAGCCATGACAATG	Detecting <i>Msfum</i>
Msfum-RT-R	GCGATTAATTGAGCGGATTGC	Detecting <i>Msfum</i>
Ecfum-RT-F	AGTGAATTACTCGGCGGTGTG	Detecting <i>Ecfum</i>
Ecfum-RT-R	GCTGAGGAATGAGTTGCTTGC	Detecting <i>Ecfum</i>
Ckfum-RT-F	CGGTGTCGCTCTTCAAAATCG	Detecting <i>Ckfum</i>
Ckfum-RT-R	CTTCGTTCTGCGTCGGATTG	Detecting <i>Ckfum</i>
Rofum-RT-F	TGATGCCCTTCAAGCCAAATC	Detecting <i>Rofum</i>
Rofum-RT-R	ACACGAGCAATACCGTAAGTC	Detecting <i>Rofum</i>

Mtsfc-RT-F	CGCTCTACACCATCGTCAAGG	Detecting <i>Mtsfc</i>
Mtsfc-RT-R	TGTACTCGGGCTGCCACTG	Detecting <i>Mtsfc</i>
actin-F	TGCCATCTACGAGGGTTTC	Detecting <i>Spmae</i>
actin-R	TTGATGTCACGGACAATTTCAC	Detecting <i>Spmae</i>