

# Supplemental Materials

*Molecular Biology of the Cell*

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## Supplemental Information

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Gene	Allele	Molecular lesion	Recessivity in suppressing ALM-PN	Strain name [with <i>mec-7(u278)</i> ]
<i>mec-7</i>	<i>u1151</i>	C12Y	recessive	TU6475
	<i>u1152</i>	G13S	recessive	TU6476
	<i>u1153</i>	S25F	semi-dominant	TU6477
	<i>u1154</i>	E69K	dominant	TU6478
	<i>u1155</i>	G71E	recessive	TU6479
	<i>u1040</i>	G98E	recessive	TU5152
	<i>u1156</i>	R121K	recessive	TU6480
	<i>u1157</i>	G141E	recessive	TU6481
	<i>u1169</i>	G146E	recessive	TU6537
	<i>u1158</i>	T149I	recessive	TU6482
	<i>u1159</i>	Q191*	recessive	TU6483
	<i>u1160</i>	D203V	recessive	TU6484
	<i>unk22</i>	A254T	recessive	CGZ46
	<i>u1161</i>	A302V	recessive	TU6485
	<i>u1170</i>	P358S	semi-dominant	TU6538
	<i>u1162</i>	Q384*	recessive	TU6486
	<i>u1171</i>	R391H	recessive	TU6539
	<i>u1164</i>	G402R	recessive	TU6488
<i>u1163</i>	80bp deletion	recessive	TU6487	
<i>mec-12</i>	<i>u1172</i>	G13E	semi-dominant	TU6540
	<i>u1173</i>	Q133*	recessive	TU6541
	<i>u1174</i>	G134E	recessive	TU6542
	<i>unk46</i>	G246E	semi-dominant	TU5153
	<i>u1175</i>	T257K	semi-dominant	TU6543
	<i>unk23</i>	G350E	semi-dominant	CGZ47
	<i>u1165</i>	G354E	semi-dominant	TU6489
	<i>u1176</i>	E415K	recessive	TU6544
	<i>u1177</i>	G416E	recessive	TU6545
<i>mec-15</i>	<i>u1042</i>	R26*	recessive	TU5183
<i>mbl-1</i>	<i>u1178</i>	C86Y	recessive	TU6546

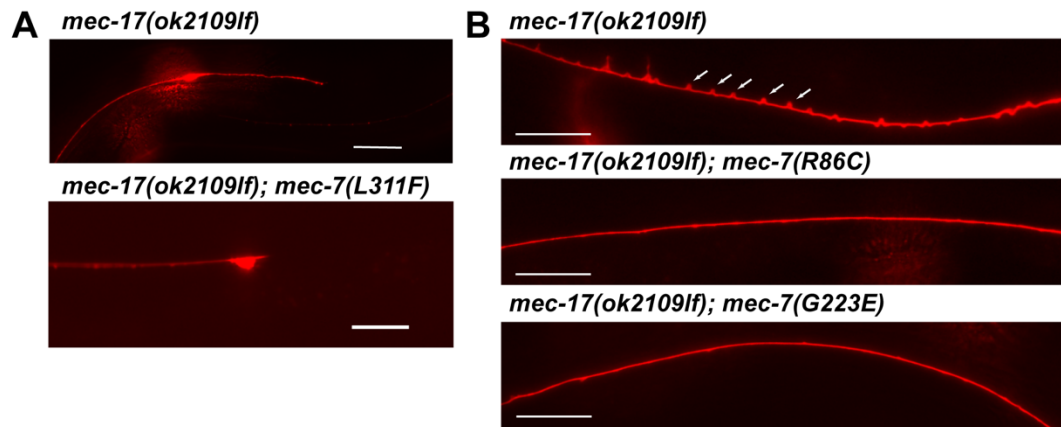
**Table S1. A list of *mec-7(u278 neo)* suppressors isolated from the screen.**

Gene	Allele	Molecular lesion	Recessivity in suppressing ALM-PN	Strain name [with <i>mec-17(ok2109)</i> ]
<i>mec-7</i>	<i>u1123</i>	E3K	Recessive	TU6122
	<i>u1111</i>	Q8*	Recessive	TU6110
	<i>u1118</i>	S25F	Semi-dominant	TU6117
	<i>u1120</i>	S25F	Semi-dominant	TU6119
	<i>u1124</i>	G34S	Recessive	TU6123
	<i>u1115</i>	R86C	Recessive	TU6114
	<i>u1122</i>	S145F	Recessive	TU6121
	<i>u1119</i>	P171L	Dominant	TU6118
	<i>u1114</i>	G223E	Recessive	TU6113
	<i>u1110</i>	L225F	Semi-dominant	TU6109
	<i>u1117</i>	D249N	Dominant	TU6116
	<i>u1121</i>	Q280*	Recessive	TU6120
	<i>u1125</i>	Intron 4 splicing variant	Recessive	TU6124
	<i>u1147</i>	L311F	Recessive	TU6407
	<i>u1116</i>	P357S, Q424L	Recessive	TU6115
<i>u1113</i>	G369E	Recessive	TU6112	
<i>mec-12</i>	<i>u1130</i>	M1I; start loss	Recessive	TU6129
	<i>u1129</i>	A19V	Semi-dominant	TU6128
	<i>u1131</i>	P63S	Recessive	TU6130
	<i>u1132</i>	E71K	Semi-dominant	TU6131
	<i>u1128</i>	S178F	Dominant	TU6127
	<i>u1126</i>	S236N	Dominant	TU6125
	<i>u1134</i>	A240T	Recessive	TU6133
	<i>u1127</i>	S241F	Semi-dominant	TU6126
	<i>u1135</i>	G246E	Semi-dominant	TU6134
<i>u1133</i>	R320C	Semi-dominant	TU6132	

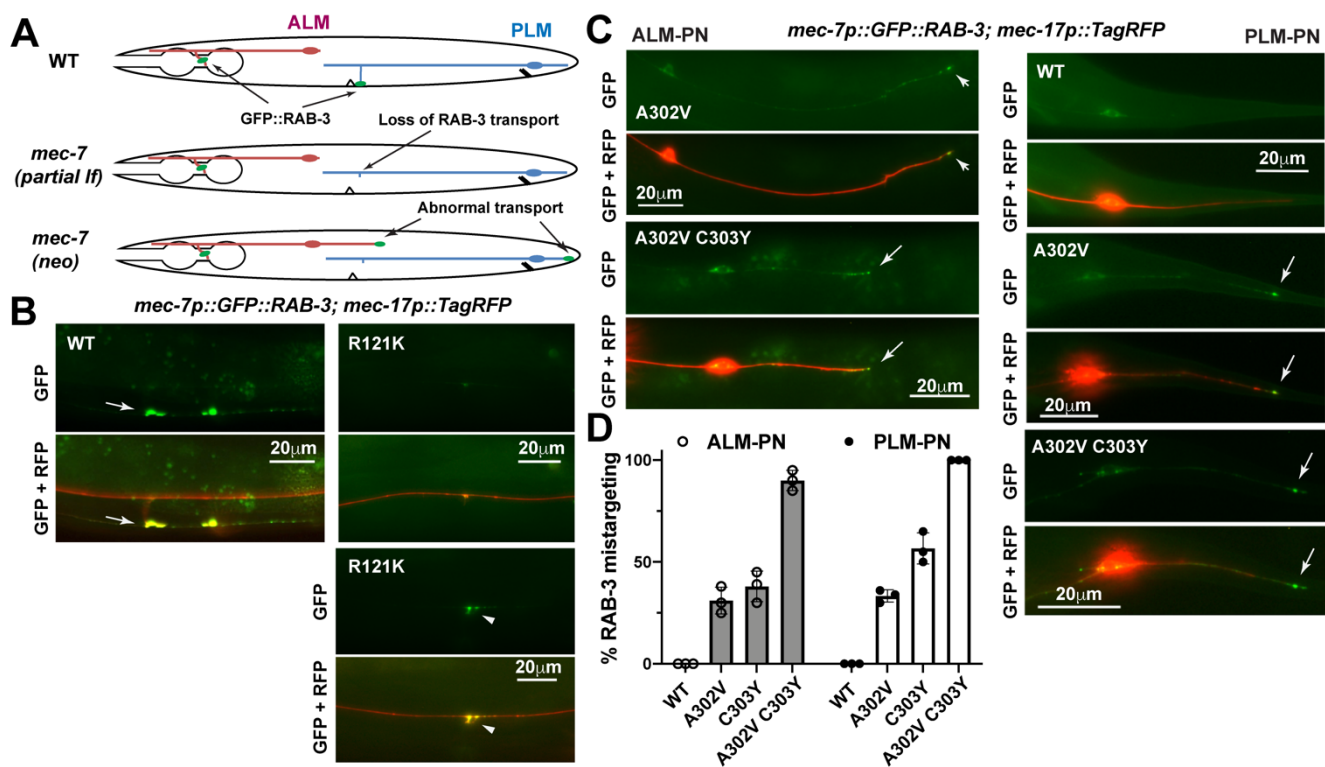
**Table S2. *mec-7* and *mec-12* alleles isolated from the *mec-17(lf)* suppressor screen.**

Allele	Mutation	CRISPR/Cas9 target	Repair template ssDNA
<i>unk64</i>	R121K	5'- GAGTAAGTTGAAATCCTTGA -3'	5'- AGCAGAACTTGTAGACAA TGTTCTTGACGTTGTCAAG AAGGAGGCTGAGAGTACT GACTGTCTTCAAGGATTTC AACTTACTCACTCACTTGG AGG-3'
<i>unk62</i>	A254T	5'- CAATGCGGATCTACGAAAGT -3'	5'- ACTTGCCTCCGCTTCCCTG GTCAACTCAATGCGGACCT CCGCAAATTAACAGTGAA CATGGTTCCATTCCCACGT CTTCACTTC-3'
<i>unk60</i>	A302V	5'- GCTGCATGCGATCCAAGACA -3'	5'- ACCCAACAATGTTTCGACG CAAAGAACATGATGGCCG TTTGCGACCCTAGGCATGG ACGTTATCTCACCGCTGCT GCCATTTCCG-3'
<i>unk63</i>	R391H	5'- GCCGCAAAGCTTTCCTTCAT- 3'	5'- atTTtgactaatttcataTTTTTccagCTAT GTTTCGGCATAAGGCATTG CTGCATTGGTACACTGGCG AGGGAATGGACGAGATGG A-3'

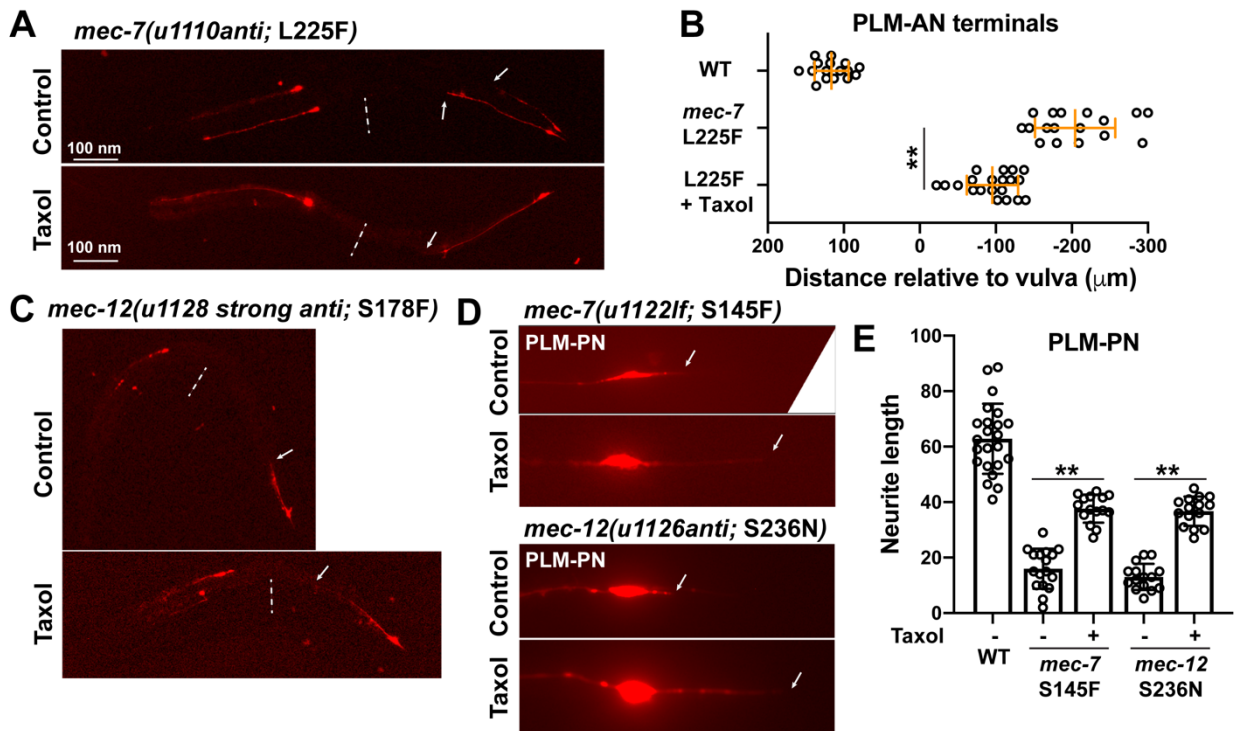
**Table S3. CRISPR/Cas9 target and repair template sequences used to recreate four missense mutations.**



**Figure S1. Suppression of *mec-17(-)* phenotypes by mutations in *mec-7*.** (A) The ectopic ALM-PN in young adults of *mec-17(ok2109)* deletion allele was suppressed by *mec-7* partial *lf* allele carrying L311F mutation. (B) The swelling and looping of ALM-AN in *mec-17(ok2109)* animals were suppressed by *mec-7* weak *neo* mutants.

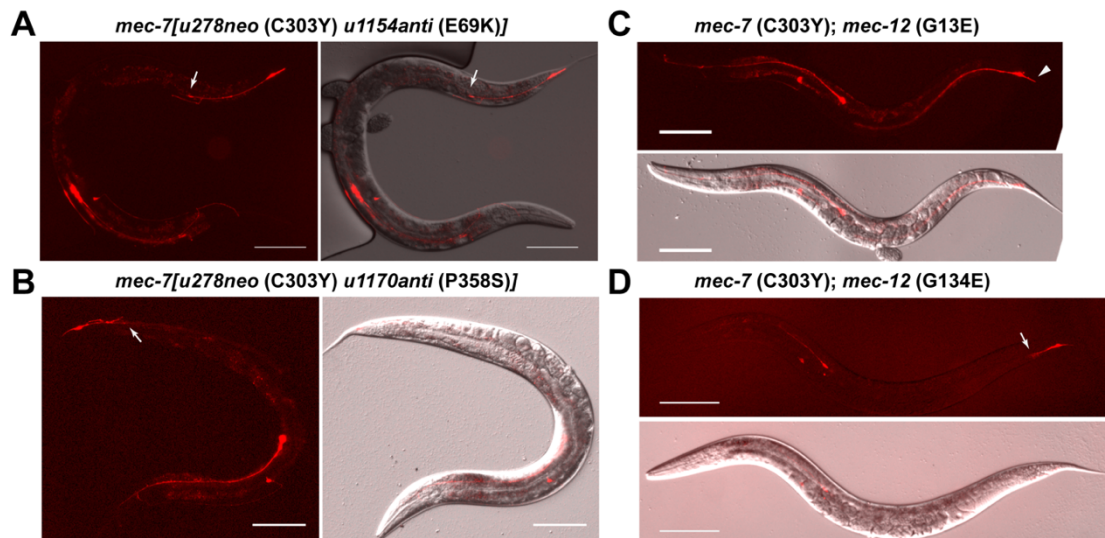


**Figure S2. *mec-7* mutations disrupted the transport of synaptic vesicle proteins.** (A) Schematic diagram for the localization of GFP-fused synaptic vesicle protein RAB-3 in the TRNs in wild-type and *mec-7* mutant animals. (B) In wild-type animals, GFP::RAB-3 in PLM neurons was located at the place where the synaptic branch of PLM-AN contacted the ventral nerve cord (arrows). In *mec-7*(R121K) partial *lf* mutants, the synaptic branch was not made and no GFP::RAB-3 signal was found on the PLM-AN. In some *mec-7*(R121K) animals, PLM-AN grew a very small branch, where weak GFP::RAB-3 signal was observed (arrow heads). (C) In *mec-7*(A302V *neo*) and *mec-7*(A302V C303Y) mutants, GFP::RAB-3 was mistargeted to the terminals (arrows) of the ALM-PN (left panel) and PLM-PN (right panel). (D) The percentages of ALM and PLM neurons that had the mistargeting of GFP::RAB-3 in *mec-7*(A302V *neo*), *mec-7*(C303Y *neo*), and *mec-7*(A302V C303Y) mutants.



**Figure S3. Taxol treatment partially rescued neurite growth defects in tubulin *lf* and *anti* mutants.** (A) PLM-AN morphology in *mec-7(u1110anti)* mutants treated with 1  $\mu$ M paclitaxel (taxol). Dashed line indicates the position of the vulva and the arrows indicate the PLM-AN terminals. (B) The distance between the vulva and the PLM-AN terminals in *mec-7(u1110anti)* mutants treated with taxol. (C) PLM-AN in *mec-12(u1128 strong anti)* mutants treated 1  $\mu$ M taxol. (D) PLM-PN in *mec-7(u1122lf)* and *mec-12(u1126anti)* mutants treated with 1  $\mu$ M Taxol. Arrows indicated where PLM-PN ends. (E) The length of PLM-PN in the treated *mec-7(u1122lf)* and *mec-12(u1126anti)* mutants. Double asterisks indicate statistically significance difference ( $p < 0.01$ ).





**Figure S4. *mec-7* and *mec-12 anti* alleles are epistatic to *mec-7 neo* alleles.** (A) *mec-7(neo)* and *mec-7(anti)* double mutants showed the shortening of PLM-AN similar to the *mec-7(anti)* single mutants. Arrows point to the premature termination of PLM-AN. (B) *mec-7(neo)* and *mec-12(anti)* showed similar phenotype as the *mec-12(anti)* alleles alone. In top panel, arrow head points to the shortening of PLM-PN seen before in *mec-12(anti)* alleles. In bottom panel, arrow points to the shortening of PLM-AN seen in the newly isolated strong *anti* alleles of *mec-12*. G13E and G134E are representative *anti* and strong *anti* alleles, respectively.