

Table S3. Phenotypes Scored in the *mpk-1(ga111)* Enhancement Assay, Related to Figure 2 and Figure S2

<i>daf-21</i> and <i>F54D12.5</i> RNAi enhance <i>mpk-1(ga111ts)</i> loss-of-function phenotypes								
			Phenotypes in Dissected Germlines-post RNAi (%)^b					Phenotypes in live animals post RNAi
Genotype	RNAi treatment	dpMPK-1^a accumulation	Delayed pachytene progression	Disorganized pachytene cells	Disorganized oocytes	Large Oocytes	N (from two independent trials)	Germ cell apoptosis (No. of corpses / gonad arm)
N2	<i>gfp</i>	++++	0	0	0	0	120	2-4 (n=34)
<i>mpk-1(ga111ts)</i>	<i>gfp</i>	++	3	1	2	0	160	2-5 (n=32)
<i>let-60(ga89ts)</i>	<i>gfp</i>	++++++	0	0	0	0	172	2-4 (n=35)
<i>rrf-1</i>	<i>gfp</i>	++++	0	0	0	0	130	3-5 (n=45)
<i>rrf-1;mpk-1(ga111ts)</i>	<i>gfp</i>	++	1	2	2	0	145	3-7 (n=47)
<i>rrf-1;let-60(ga89ts)</i>	<i>gfp</i>	+++++	0	0	0	0	136	2-7 (n=51)
N2	<i>daf-21</i>	++	2	8	8	0	190	5-9 (n=65)
<i>mpk-1(ga111ts)</i>	<i>daf-21</i>	+/- ^c	18	22	28	25	180 ^{d, e}	10-14 (n=57)
<i>let-60(ga89ts)</i>	<i>daf-21</i>	++	0	0	0	0	145	4-7 (n=53)
<i>rrf-1</i>	<i>daf-21</i>	++	5	7	10	0	175	7-10 (n=63)
<i>rrf-1;mpk-1(ga111ts)</i>	<i>daf-21</i>	+/- ^c	14	18	32	31	160 ^f	11-14 (n=59)
<i>rrf-1;let-60(ga89ts)</i>	<i>daf-21</i>	++	0	0	0	0	130	6-8 (n=68)
N2	<i>F54D12.5</i>	++++	0	0	0	0	190	^g
<i>mpk-1(ga111ts)</i>	<i>F54D12.5</i>	++	3	25	28	0	190	^g
<i>let-60(ga89ts)</i>	<i>F54D12.5</i>	++++++	0	0	0	0	168	^g
<i>rrf-1</i>	<i>F54D12.5</i>	++++	0	0	0	0	132	^g
<i>rrf-1;mpk-1(ga111ts)</i>	<i>F54D12.5</i>	++	7	28	32	0	190	^g
<i>rrf-1;let-60(ga89ts)</i>	<i>F54D12.5</i>	+++++	0	0	0	0	130	^g

a: dpMPK-1 strength assessed by setting the accumulation level / gain in N2 (with GFP RNAi) at ++++ as baseline
b: F1 animals analyzed. Phenotypes described in detail in Lee et al. (2007) and Arur et al. (2009).
c: No/very minimal dpMPK-1 signal visible when pictures taken at the same gain as WT and *ga111* with GFP RNAi
d: Some germlines had more than one phenotype
e: Not included are 16% of germlines that showed 'null' *mpk-1* phenotypes / very severe loss-of-function.
f: Not included are 21% of germlines that showed 'null' *mpk-1* phenotypes / very severe loss-of-function.
g: Apoptosis was also scored, but not found to change following *F54D12.5* RNAi.

To determine whether *daf-21* or F54D12.5 function in the MAPK pathway, we determined whether partial RNAi of *daf-21* or F54D12.5 could enhance the *mpk-1(ga111)* loss-of-function phenotype at the permissive temperature (20°C). Attenuated RNAi, gonad dissection, immunofluorescence and gonad analysis was performed as described previously (Arur et al., 2009). Briefly, partial RNAi of *daf-21* or F54D12.5 was performed by feeding bacteria expressing dsRNA to the worms and examining their gonads at timepoints after feeding onset when the observed gonad morphology, following either partial depletion, was comparable to controls (Figures 2E and 2F). When partial *daf-21(RNAi)* was performed in the *mpk-1(ga111)* background, either in the presence or absence of the *rrf-1* null mutation; a significantly enhanced phenotype was observed. Approximately 20% of dissected gonads exhibited the *mpk-1* null phenotype and the remaining gonads showed partial disruption of one or more MPK-1 dependent processes: pachytene progression, pachytene cellular organization, oocyte organization and differentiation, oocyte growth control, and inhibition of germ cell apoptosis (for examples see Figures 2E and 2F; Figure S2). Partial RNAi of F54D12.5 also enhanced the *mpk-1(ga111)* phenotype. In this case, two phenotypes associated with MAPK knockdowns were observed: disorganized pachytene cells and disorganized oocytes (**Figure 2F**, Figure S2).