

Figure S1

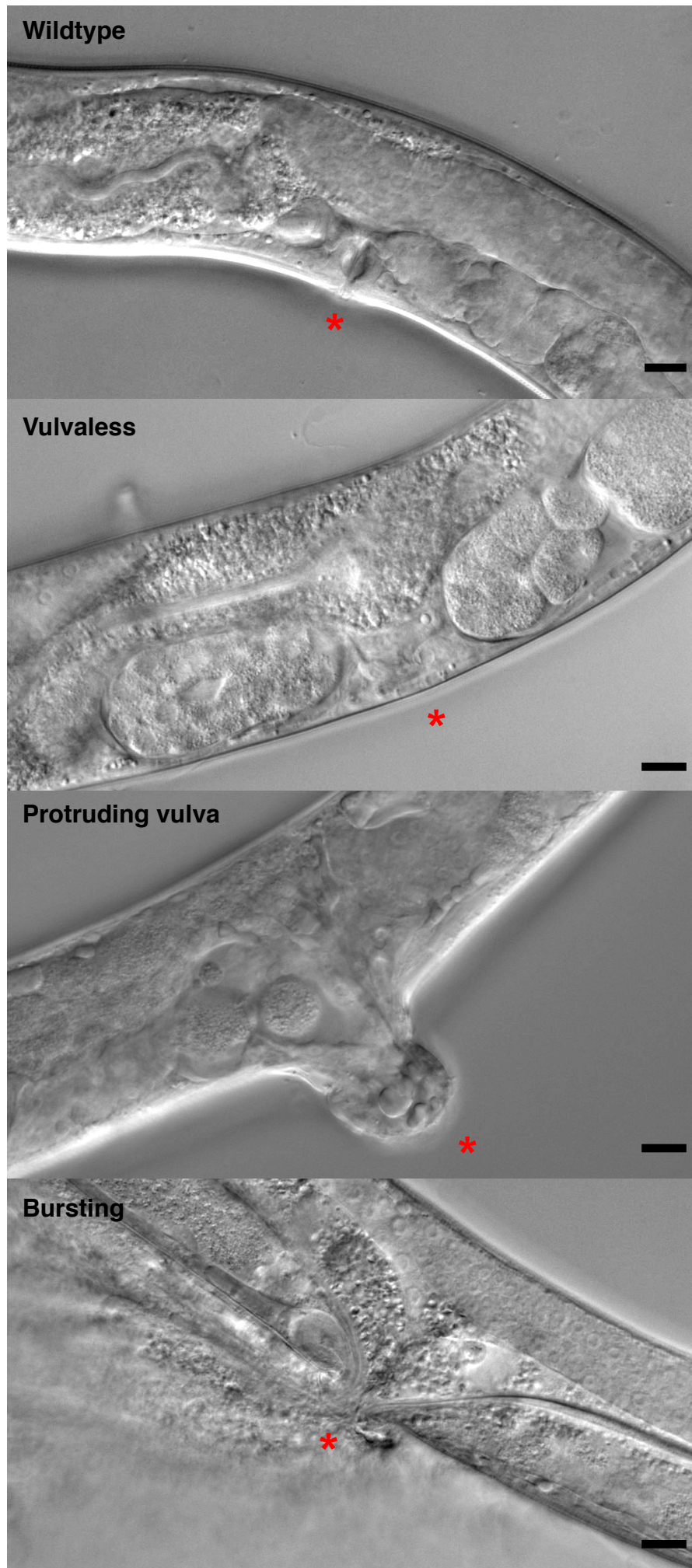


Figure S2Ai

lin4_m1							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#208.2.2.1	1.9	40.6	36.8	20.8	106	79.2	
#208.3.1.0	0.0	8.1	19.5	72.4	123	27.6	
#208.3.2.1	7.0	43.4	27.1	22.5	129	77.5	
#208.3.2.2	39.6	34.7	16.0	9.7	144	90.3	
#209a.1.1.1	8.2	43.9	26.5	21.4	98	78.6	
#209a.1.1.2	33.3	40.7	19.5	6.5	123	93.5	
#209a.3.1.1	38.1	42.9	16.2	2.9	105	97.1	
Average	18.3	36.3	23.1	22.3	-	77.7	
Error of mean	6.7	4.8	2.8	8.9	-	8.9	

lin4_m2							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#210.1.1.0	0.9	25.5	22.6	50.9	106	49.1	
#210.1.2.0	0.0	25.7	39.6	34.7	101	65.3	
#210.1.2.1	0.0	14.0	7.5	78.5	107	21.5	
#211.1.1.1	0.0	10.2	28.1	61.7	128	38.3	
#211.2.1.1	4.3	35.0	39.3	21.4	117	78.6	
Average	1.0	22.1	27.4	49.4	-	50.6	
Error of mean	0.8	4.5	6.0	10.0	-	10.0	

lin4_m3							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#212a.1.1.0	0.0	3.7	18.3	78.0	109	22.0	
#212a.3.1.0	0.9	2.6	0.0	96.6	116	3.4	
#212a.4.1.0	0.0	1.7	3.4	95.0	119	5.0	
#213.1.1.1	1.1	2.2	0.0	96.7	91	3.3	
#213.1.2.1	0.0	0.8	0.0	99.2	121	0.8	
#213.1.3.1	0.0	1.0	1.0	98.0	100	2.0	
Average	0.3	2.0	3.8	93.9	-	6.1	
Error of mean	0.2	0.4	3.0	3.2	-	3.2	

lin4_m4							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#199.3.1.1	0.0	1.1	2.1	96.8	94	3.2	
#199.3.2.1	0.0	1.9	1.9	96.2	209	3.8	
Average	0.0	1.5	2.0	96.5	-	3.5	
Error of mean	0.0	0.4	0.1	0.3	-	0.3	

lin4_m5							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#215.1.1.1	0.0	0.8	5.6	93.7	126	6.3	
#215.2.1.1	0.0	0.0	0.9	99.1	110	0.9	
#214.3.1.0	0.8	8.3	11.6	79.3	121	20.7	
#214.3.1.1	0.0	6.8	10.2	83.0	88	17.0	
Average	0.2	4.0	7.1	88.8	-	11.2	
Error of mean	0.2	2.1	2.4	4.6	-	4.6	

lin4_m6							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#237a.3.1.0	0.0	0.0	0.0	100.0	128	0	

lin4_m7							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#216.1.1.0	0.0	9.9	16.6	73.5	151	26.5	
#216.1.1.1	0.0	5.1	7.6	87.3	118	12.7	
#216.1.1.2	0.0	3.5	2.6	93.9	114	6.1	
#216.1.2.1	0.0	2.9	5.9	91.2	102	8.8	
#216.1.3.1	0.0	2.5	13.1	84.4	122	15.6	
#217.1.1.1	0.0	22.2	15.9	61.9	126	38.1	
#217.3.1.0	1.6	18.0	17.2	63.3	128	36.7	
#217.3.2.0	0.0	20.5	11.4	68.2	88	31.8	
Average	0.2	10.6	11.9	78.0	-	22.0	
Error of mean	0.2	3.0	1.3	4.5	-	4.5	

lin4_m8							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#241.2.1.0	0.0	5.8	9.9	84.3	121	15.7	
#240A.1.2.0	0.0	6.0	2.4	91.7	84	8.3	
#240A.1.3.0	0.0	0.0	0.0	100.0	79	0.0	
Average	0.0	3.9	4.1	92.0	-	8.0	
Error of mean	0.0	2.0	3.0	4.5	-	4.5	

lin4_m9							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#243.2.1.1	51.8	27.7	13.3	7.2	83	92.8	
#242A.1.1.0	16.9	49.4	20.5	13.3	83	86.7	
#242A.1.2.0	27.7	36.2	25.5	10.5	94	89.4	
#242A.1.3.0	17.7	29.1	39.2	13.9	79	88.1	
Average	28.5	35.6	24.6	11.3	-	86.7	
Error of mean	8.1	5.0	5.5	1.5	-	1.5	

lin4_m10							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#235.2.1.1	0.0	17.0	10.4	72.6	106	27.4	
#234B.1.3.0	0.0	5.8	14.0	80.2	86	19.8	
Average	0.0	11.4	12.2	76.4	-	23.6	
Error of mean	0.0	5.6	1.8	3.8	-	3.8	

lin4_m11							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#238.4.1.0	0.0	2.0	5.0	93.1	101	6.9	
#239A.1.1.1	0.0	0.0	1.2	98.8	84	1.2	
Average	0.0	1.0	3.1	95.9	-	4.1	
Error of mean	0.0	1.0	1.9	2.9	-	2.9	

lin4_m12							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
250.1.1.0	10.0	36.7	31.7	21.7	120	78.3	
250.2.2.0	60.6	26.5	8.3	4.5	132	95.5	
250.2.1.0	34.8	41.1	17.7	6.4	141	93.6	
Average	35.1	34.8	19.2	10.9	-	89.1	
Error of mean	14.6	4.3	6.8	5.4	-	5.4	

lin4_m13							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
251.1.1.1	0.0	21.3	14.6	64.0	89	36.0	
251.1.1.3	1.4	40.5	18.9	39.2	148	60.8	
251.1.1.4	29.8	37.7	21.9	10.5	114	89.5	
251.1.2.0	19.8	34.9	19.0	26.2	126	73.8	
251.1.3.0	32.7	40.7	10.7	16.0	150	84.0	
251.1.4.1	7.3	40.9	27.3	24.5	110	75.5	
Average	15.2	36.0	18.7	30.1	-	69.9	
Error of mean	5.8	3.1	2.3	7.9	-	7.9	

lin4_m14							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
252.1.1.0	2.6	39.7	34.5	23.3	116	76.7	
252.2.1.1	29.2	40.6	25.5	4.7	106	95.3	
Average	15.9	40.1	30.0	14.0	-	86.0	
Error of mean	13.3	0.5	4.5	9.3	-	9.3	

WT lin-4							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
#173.1.1.1	4.2	38.3	30.8	26.7	120	73.3	
#173.3.1.1	2.0	26.0	19.0	53.0	100	47.0	
#173.3.2.1	2.8	27.4	20.8	49.1	106	50.9	
#173.3.3.1	11.1	28.7	52.8	7.4	108	92.6	
#173.3.4.1	0.0	16.1	33.9	50.0	118	50.0	
#172.1.1.1	0.0	6.6	12.1	81.3	91	18.7	
#172.1.2.1	0.0	12.2	9.2	78.6	131	21.4	
#172.1.3.1	0.0	10.5	7.0	82.5	114	17.5	
#172.1.4.1	0.8	27.2	30.4	41.6	125	58.4	
Average	2.3	21.4	24.0	52.2	-	47.8	
Error of mean	1.2	3.5	4.9	8.5	-	8.5	

Figure S2Aii

lin4_cm2							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
PD1088	89.1	3.0	6.0	2.0	101	98.0	

lin4_cm6							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
PD1093	0.0	0.0	14.8	85.2	54	14.8	

lin4_cm7							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
PD1095	0.0	3.2	90.3	6.5	62	93.5	
PD1263	0.0	0	100.0	0.0	11	100.0	
PD1264	0.0	9.1	90.9	0.0	11	100.0	
PD1265	0.0	0	90.0	10.0	10	90.0	
Average	0.0	3.1	92.8	4.1	-	95.9	
Error of mean	0.0	2.1	2.4	2.5	-	2.93	

lin4_cm11							
Line	% Egg-laying	% Pvl	% Bursting	% Vulvaless	Total	% Vulval rescue	
PD1086	1.3	0	1.3	97.4	78	2.6	

Figure S2B

Strains		# lin-4 reads	% microRNAs	total reads
N2		18,703	0.97	3,423,171
lin-4(e912)		0	0.00	4,133,073
lin-4(ma161)		1,076	0.08	2,435,830
WT lin-4	Line 1448	5,386	1.12	1,796,199
	Line 1469	8,922	2.07	2,284,580
lin4_m1	Line 208.3.2.2	13,656	1.66	752,404
	Line 209a.1.1.2	9,780	2.40	452,985
lin4_m2	Line 210.1.2.0	1,303	0.93	135,796
	Line 211.2.1.1	3,099	0.55	854,643
lin4_m3	Line 212a.1.1.0	2,216	0.48	1,232,863
	Line 213.1.2.1	3,174	1.97	1,235,272
lin4_m4	Line 199.3.2.1	1,145	0.48	464,624
	Line 214.3.1.0	6,369	1.25	501,990
lin4_m5	Line 215.1.1.1	47,833	6.91	619,924
	Line 216.1.1.0	8,112	1.41	519,230
lin4_m7	Line 217.1.1.1	4,554	0.69	666,453

Figure S2C

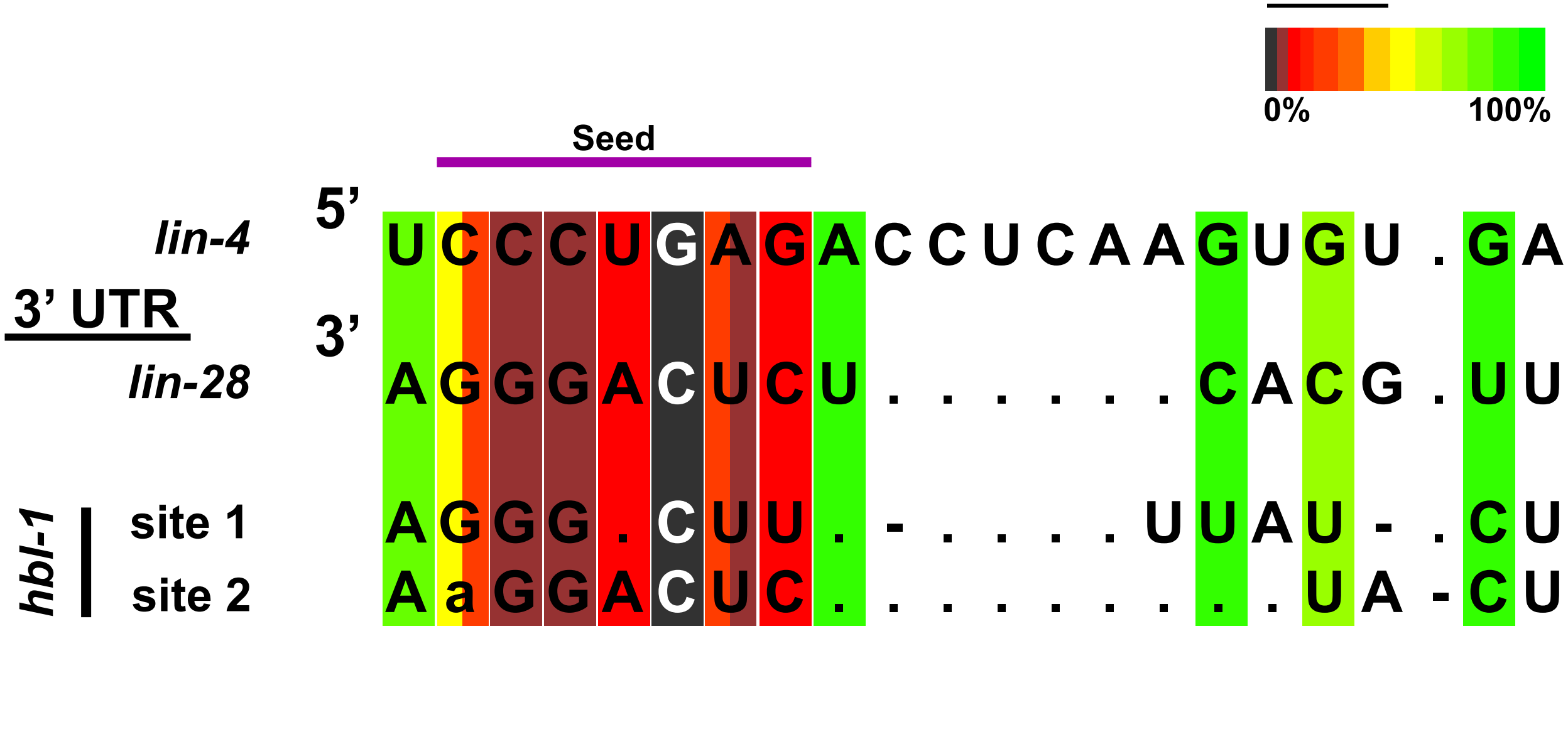


Figure S2Di

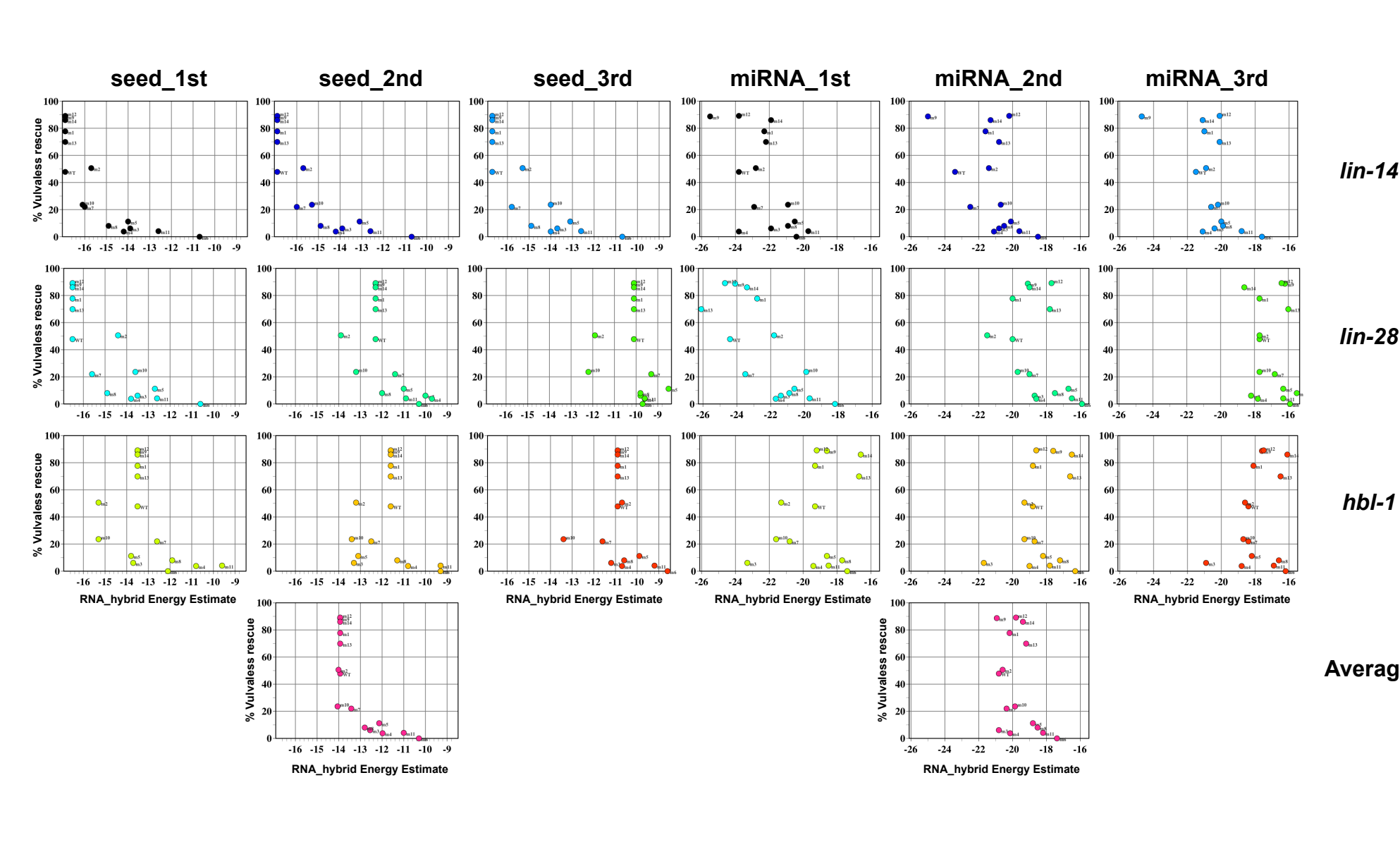


Figure S2Dii

Mutation	Minimum free energy (lin-4 mature microRNA)									% Vulval rescue	
	lin-14			lin-28			hbl-1				Average
	1st	2nd	3rd	1st	2nd	3rd	1st	2nd	3rd		
WT	-23.8	-23.4	-21.5	-24.4	-20.0	-17.7	-19.3	-18.8	-18.4	-20.8	47.8
m1	-22.3	-21.6	-21.0	-22.8	-20.0	-17.7	-19.3	-18.8	-18.1	-20.2	77.7
m2	-22.8	-21.4	-20.9	-21.8	-21.5	-17.7	-21.3	-19.3	-18.6	-20.6	50.6
m3	-21.9	-20.8	-20.4	-21.4	-18.7	-18.2	-23.3	-21.7	-20.9	-20.8	6.1
m4	-23.8	-20.1	-21.1	-21.7	-18.6	-17.8	-19.4	-19.0	-18.8	-20.1	3.8
m5	-20.5	-20.1	-20.0	-20.6	-16.7	-16.3	-18.6	-18.2	-18.2	-18.8	11.2
m6	-20.4	-18.5	-17.6	-18.2	-15.9	-15.9	-17.4	-16.3	-16.2	-17.4	0.0
m7	-22.9	-22.5	-20.6	-23.5	-19.0	-16.8	-20.8	-18.7	-18.4	-20.4	22.0
m8	-20.9	-20.5	-19.9	-20.9	-17.5	-15.5	-17.7	-17.2	-16.6	-18.5	8.0
m9	-25.5	-25.0	-24.7	-24.1	-19.1	-16.2	-18.6	-17.6	-17.6	-20.9	88.7
m10	-20.9	-20.7	-20.2	-19.9	-19.7	-17.7	-21.6	-19.3	-18.7	-19.9	23.6
m11	-19.7	-19.6	-18.8	-19.7	-16.5	-16.3	-18.5	-17.8	-16.9	-18.2	4.1
m12	-23.8	-20.2	-20.1	-24.7	-17.7	-16.4	-19.2	-18.6	-17.5	-19.8	89.1

Figure S3A

let7_m1	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	92.8	7.2	0.0	97	100.0
#203z.2.1.1	91.1	8.9	0.0	101	100.0
Average	91.9	8.1	0.0	-	100.0
Error of mean	0.8	0.8	0.0	-	0.0

let7_m2	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	51.5	29.9	18.6	97	81.4
#205z.1.1.0	51.5	29.9	18.6	97	81.4
Average	51.5	29.9	18.6	-	81.4
Error of mean	0.0	0.0	0.0	-	0.0

let7_m3	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	24.5	64.2	11.3	106	88.7
#225.3.1.0	5.0	51.0	44.0	100	56.0
Average	14.8	57.6	27.7	-	72.4
Error of mean	9.8	6.6	16.3	-	16.3

let7_m4	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	27.6	51.0	21.4	98	78.6
#220.3.2.1	26.1	49.6	24.3	115	75.7
#221.2.1.1	18.0	60.0	22.0	100	78.0
#221.2.2.1	15.3	52.3	32.4	111	67.6
Average	21.7	53.2	25.1	-	74.9
Error of mean	3.0	2.3	2.5	-	2.5

let7_m5	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	57.1	42.1	0.0	133	99.2
#260.2.2.1	57.1	42.9	0.0	98	100.0
Average	57.1	42.5	0.0	-	99.6
Error of mean	0.0	0.4	0.4	-	0.4

let7_m6	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	33.7	65.2	1.1	89	98.9
#248a.1.1.0	33.7	65.2	1.1	89	98.9
Average	33.7	65.2	1.1	-	98.9
Error of mean	0.0	0.0	0.0	-	0.0

let7_m7	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	82.5	17.5	0.0	120	100.0
#227.1.1.0	82.5	17.5	0.0	120	100.0
Average	82.5	17.5	0.0	-	100.0
Error of mean	0.0	0.0	0.0	-	0.0

let7_m8	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	66.4	32.8	0.9	116	99.1
#249.1.1.1	43.6	46.4	10.0	140	90.0
#259.1.1.1	42.6	48.5	8.9	101	91.1
#259.1.2.1	49.6	47.1	3.3	121	96.7
Average	50.5	43.7	5.8	-	94.2
Error of mean	5.5	3.7	2.2	-	2.2

let7_m9	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	57.9	38.9	3.2	95	96.8
#247.1.1.1	57.9	38.9	3.2	95	96.8
Average	57.9	38.9	3.2	-	96.8
Error of mean	0.0	0.0	0.0	-	0.0

let7_m10	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	66.1	33.1	0.8	118	99.2
#258.1.1.1	59.5	36.9	3.6	111	96.4
Average	62.8	35.0	2.2	-	97.8
Error of mean	3.3	1.9	1.4	-	1.4

let7_m11	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	4.2	68.4	27.4	95	72.6
#256.1.1.1	4.2	68.4	27.4	95	72.6
Average	4.2	68.4	27.4	-	72.6
Error of mean	0.0	0.0	0.0	-	0.0

let7_m12	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	43.4	45.6	11.0	136	89.0
#245.2.1.0	42.7	43.6	13.6	110	86.4
#245.2.1.2	21.4	60.0	18.9	90	81.1
#245.2.2.0	45.2	49.0	5.8	104	94.2
#254.1.1.0	35.1	35.1	29.8	114	70.2
Average	37.5	46.7	15.8	-	84.2
Error of mean	4.4	4.0	4.1	-	4.1

let7_m13	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	9.9	29.5	60.7	61	39.3
#253.1.1.1	13.1	51.2	35.7	84	64.3
#253.2.1.2	31.3	88.6	10.1	99	89.9
#253.2.2.0	34.6	62.8	2.6	78	97.4
#253.2.4.1	32.0	54.0	14.0	100	86.0
#253.2.5.0	30.4	53.0	16.5	115	83.5
#244.2.4.0	38.4	52.7	17.0	112	83.0
#244.2.1.1	5.7	14.8	79.5	88	20.5
Average	23.4	47.1	29.5	-	70.5
Error of mean	5.3	8.4	13.7	-	13.7

let7_seedmut	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	0.0	4.0	96.0	80	4.0
#230.1.3.0	0.0	0.0	100.0	95	0.0
#230.1.3.1	0.0	0.8	99.2	130	0.8
Average	0.0	1.6	98.4	-	1.6
Error of mean	0.0	1.2	1.2	-	1.2

let7_strandswap	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	74.2	15.9	8.8	132	93.1
#232.1.1.0	74.1	23.1	2.8	108	97.2
#233.1.2.0	85.7	12.5	1.8	112	98.2
Average	78.0	18.2	3.8	-	96.2
Error of mean	3.9	3.1	1.5	-	1.6

WT let-7	% Egg-laying	% Pvl	% Bursting	Total	% Rescue (bursting)
Line	72.7	25.1	1.1	118	94.9
C29.2.1.0	58.5	36.4	5.1	77	97.4
C29.2.2.0	54.5	42.9	2.6	77	97.4
Average	61.9	35.1	2.9	-	97.0
Error of mean	5.5	4.9	1.2	-	1.2

Figure S3B

let7_m1	% with adult alae	Total
Line	100.0	48
#203z.4.1.1	100.0	48
Average	100.0	-
Error of mean	0.0	-

let7_m2	% with adult alae	Total
Line	100.0	51
#205z.1.1.0	100.0	51
Average	100.0	-
Error of mean	0.0	-

let7_m3	% with adult alae	Total
Line	2.1	48
#225.3.1.0	0.0	48
Average	1.0	-
Error of mean	1.0	-

let7_m4	% with adult alae	Total
Line	55.6	54
#221.2.1.0	79.2	48
#221.2.2.1	59.0	48
#220.3.2.1	50.0	48
Average	58.7	-
Error of mean	7.0	-

let7_m5	% with adult alae	Total
Line	82.0	50
#260.2.2.1	78.0	50
Average	80.0	-
Error of mean	2.0	-

let7_m6	% with adult alae	Total
Line	50.8	61
#248a.1.1.0	50.8	61
Average	50.8	-
Error of mean	0.0	-

let7_m7	% with adult alae	Total
Line	83.3	54
#227.1.1.0	83.3	54
Average	83.3	-
Error of mean	0.0	-

let7_m8	% with adult alae	Total
Line	47.2	53
#249.1.1.1	35.3	51
#259.1.1.1	28.9	52
#259.1.2.1	33.3	48
Average	35.7	-
Error of mean	4.2	-

let7_m9	% with adult alae	Total
Line	93.9	49
#247.1.1.1	93.9	49
Average	93.9	-
Error of mean	0.0	-

let7_m10	% with adult alae	Total
Line	100.0	41
#258.1.1.1	85.1	47
Average	92.6	-
Error of mean	7.5	-

let7_m11	% with adult alae	Total
Line	5.7	53
#256.1.1.1	5.7	53
Average	5.7	-
Error of mean	0.0	-

let7_m12	% with adult alae	Total
Line	35.2	54
#254.1.1.0	20.8	53
#245.2.1.1	14.6	48
#245.2.1.2	30.6	62
#245.2.2.0	17.9	56
Average	23.8	-
Error of mean	3.9	-

let7_m13	% with adult alae	Total
Line	20.0	55
#253.1.1.1	10.4	48
#253.2.1.2	7.3	55
#253.2.2.0	6.0	50
#253.2.4.1	10.4	48
#253.2.5.0	11.1	54
#244.2.1.1	18.0	50
#244.2.4.0	28.1	57
Average	13.9	-
Error of mean	2.7	-

let7_strandswap	% with adult alae	Total
Line	87.0	54
#232.1.1.0	100.0	48
#233.1.2.0	93.8	48
Average	93.8	-
Error of mean	3.8	-

WT let-7	% with adult alae	Total
Line	90.7	54
C29.2.1.0	100.0	46
C29.2.1.1	90.7	49
C29.2.2.0	96.2	49
Average	96.2	-
Error of mean	2.8	-

Figure S3C

Strains	# let-7 reads	% microRNAs	total reads
N2	98,447	6.82	3,423,171
let7_m1	20,858	4.52	1,014,126
let7_m2	5,335	1.10	206,735
let7_m3	398	0.82	260,682
let7_m4	17	1.86	7,428
let7_m7	863	0.82	369,456
let7_m7	591	1.16	262,801
let7_strandswap	61	0.53	118,634
let7_strandswap	114	0.53	204,384

Figure S3Di

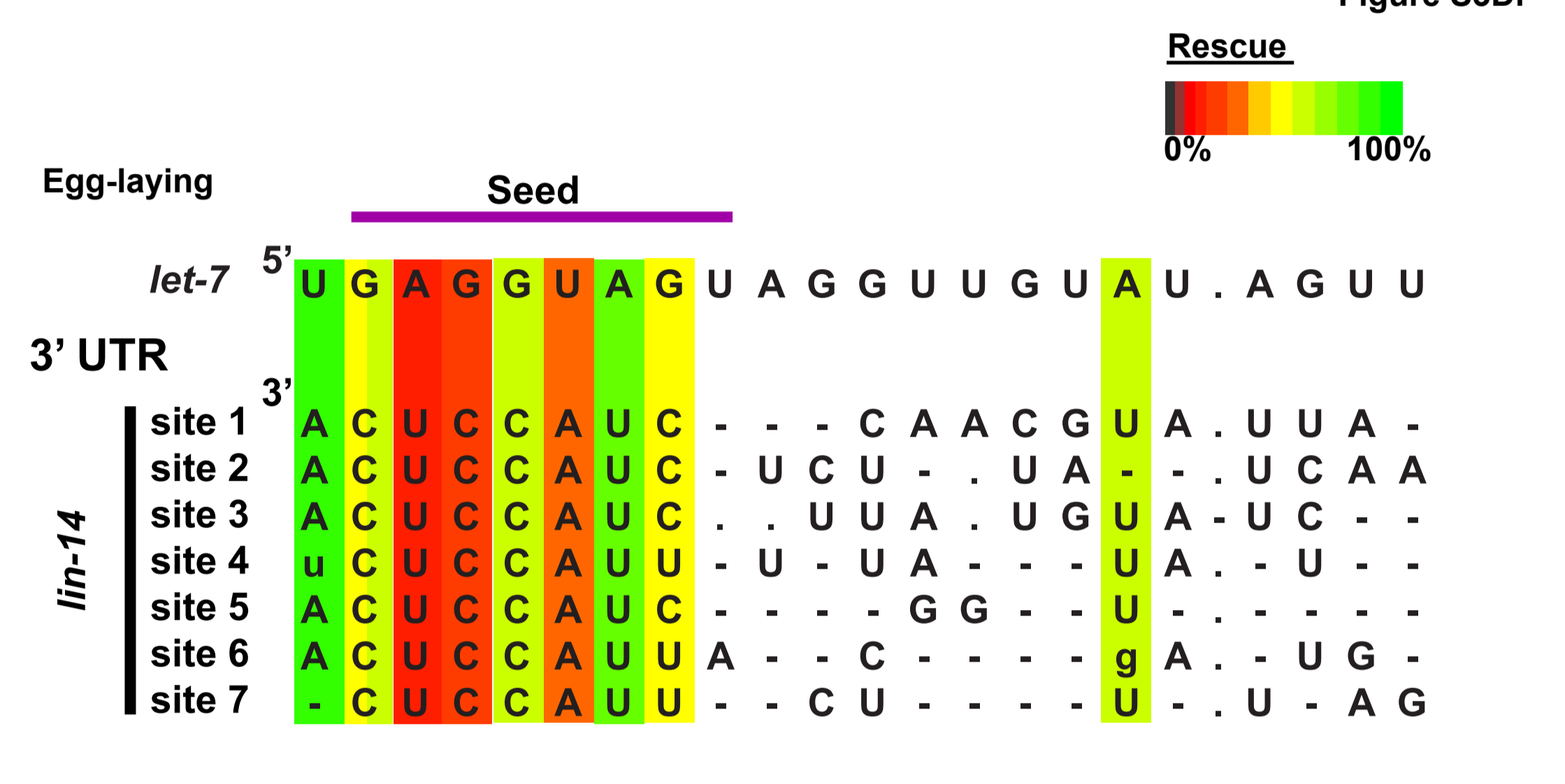


Figure S3Dii

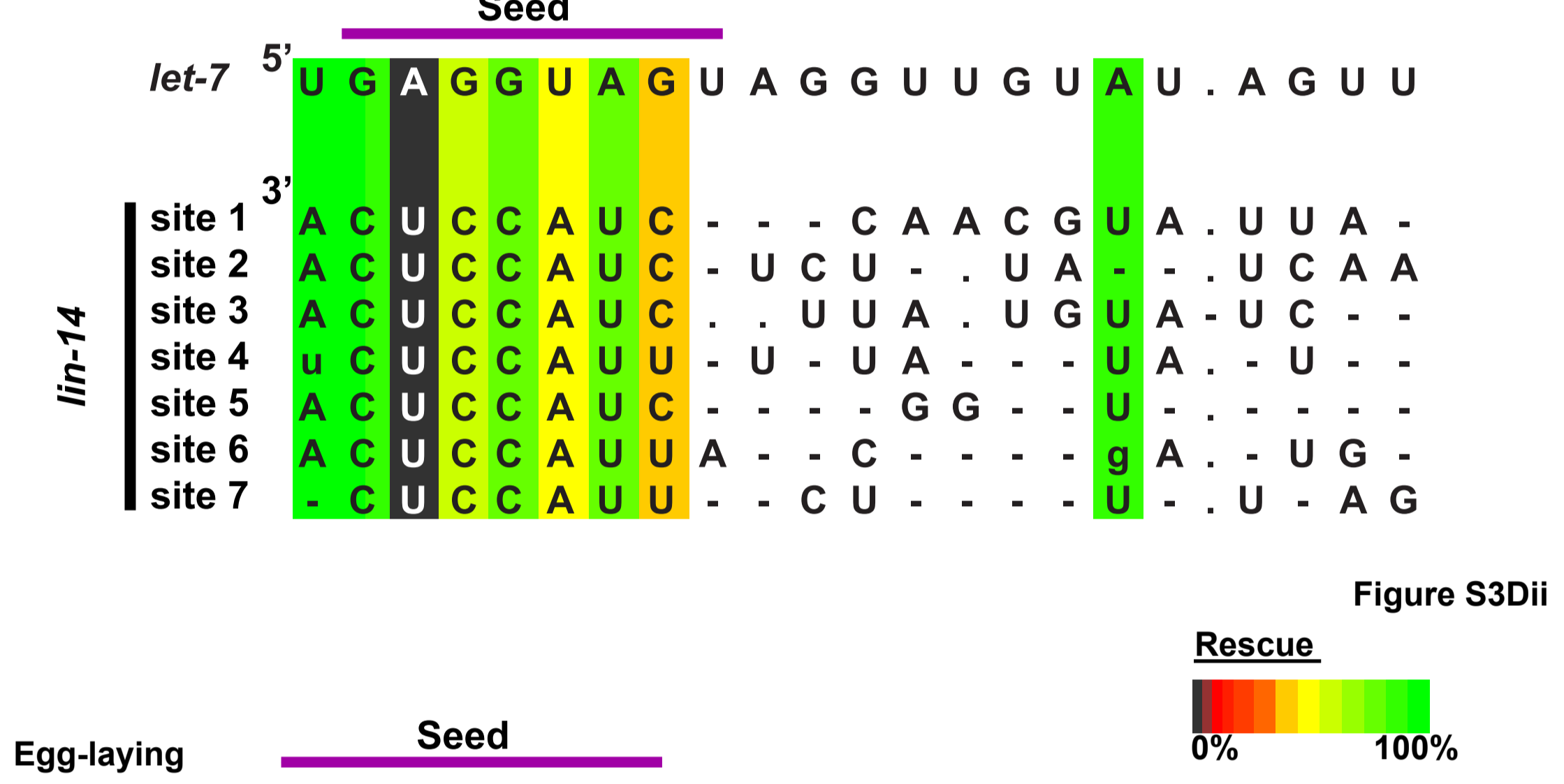


Figure S3Diii

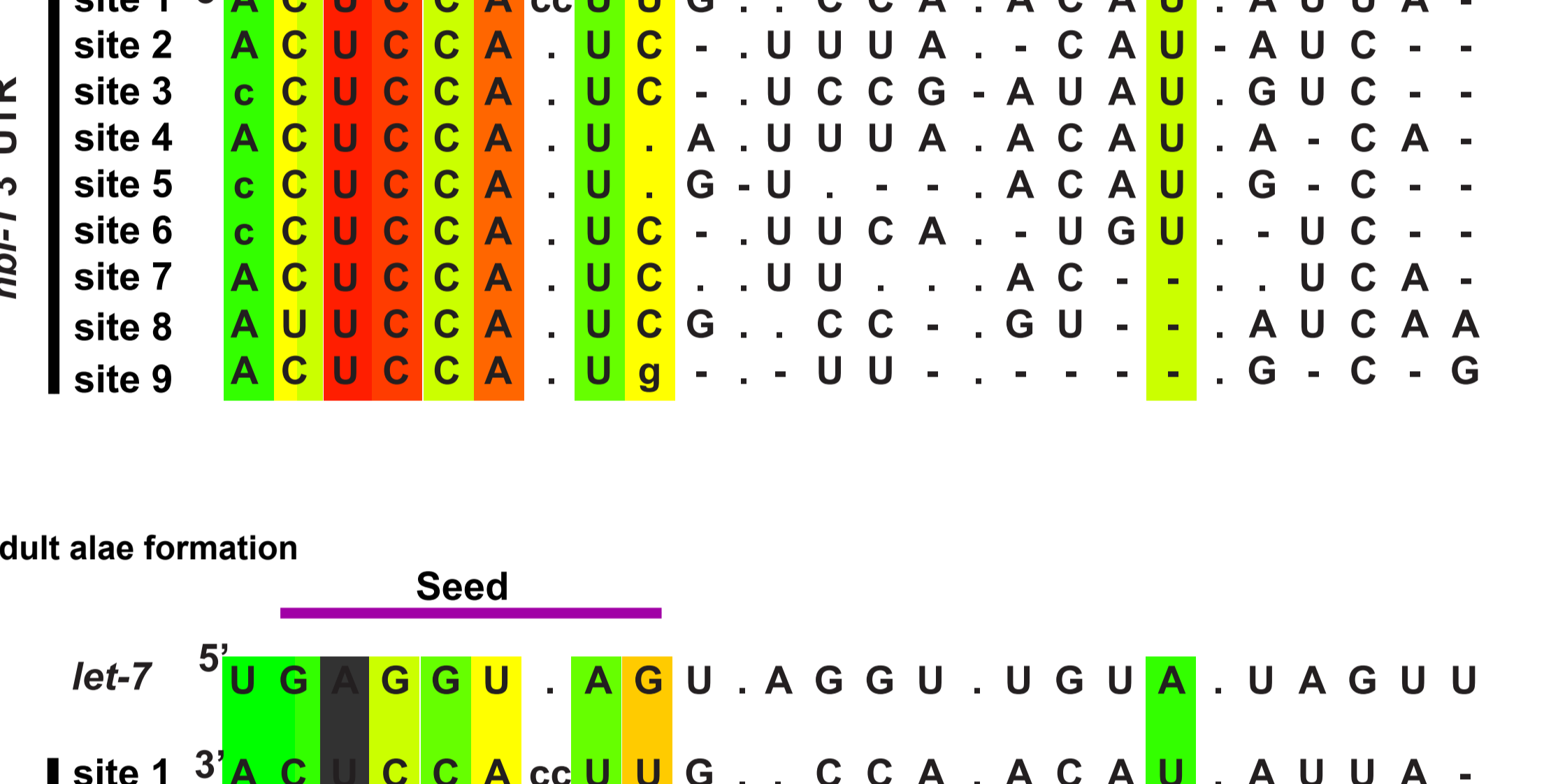


Figure S3Ei

Figure S3Eii

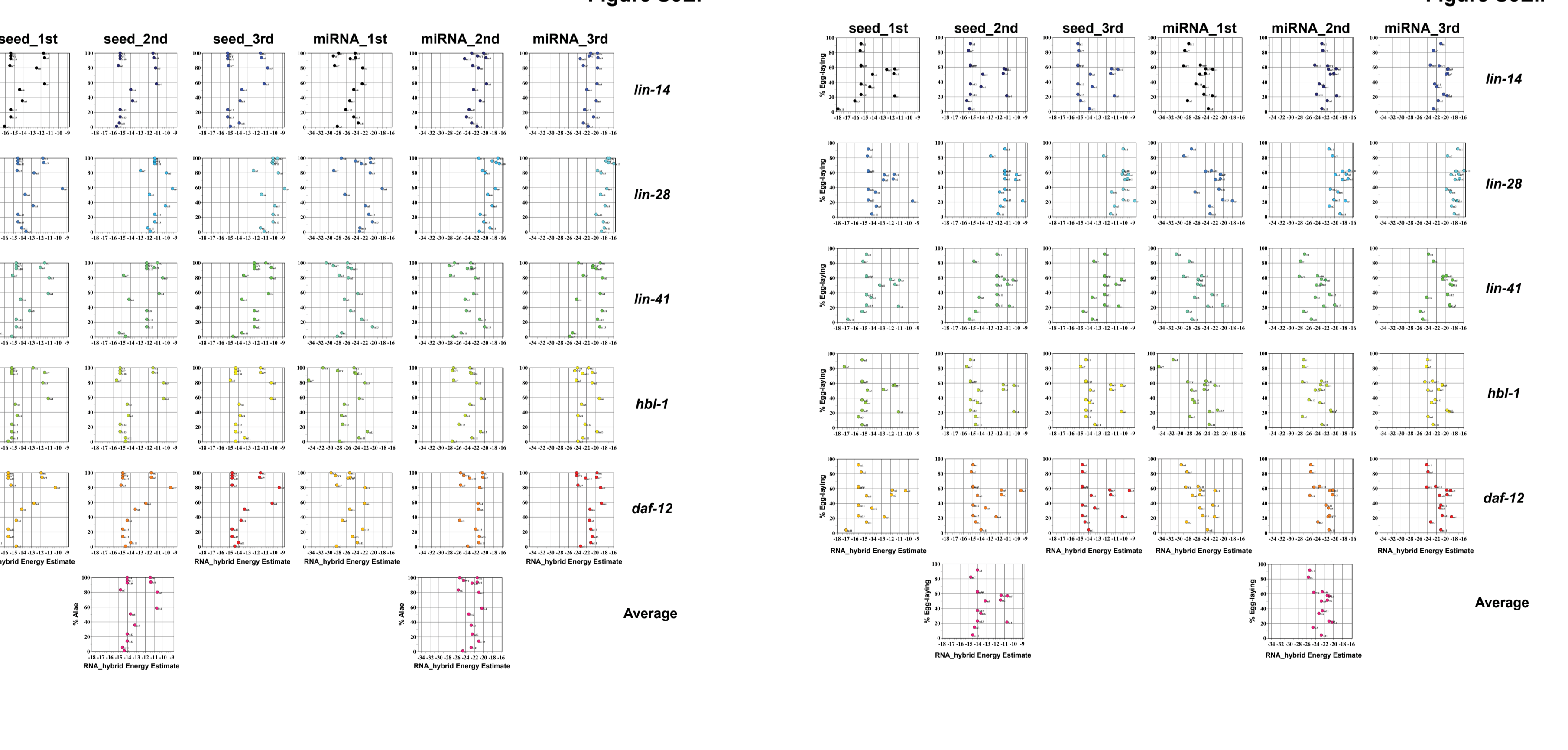


Figure S3Eiii

Mutation	Minimum free energy (let-7 mature microRNA)															Rescue		
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