

Table S1. Larval cuticles: Numerical analysis of RNAi phenotypes**A. Summary. Pooled results originate from separate injection experiments using different dsRNA-concentrations (in red: concentrations used)**

	<i>Fz1/2</i> 300 ng/μl / 1500 ng/μl	<i>Fz1</i> <1000 ng/μl / 1500 ng/μl	<i>Fz1/4</i>	<i>Fz4</i>	<i>arrow</i>	water
Total <i>n</i>	372 / 389	383 / 69	943	396	3290	130
Percentage of specific phenotypes (<i>n</i>)	72.5% (270) 43% (167)	29% (112) 29% (20)	57% (538)	2.5% (10)	10.5% (347)	– *
Percentage of empty eggs (<i>n</i>)	17.5% (65) 54.2% (211)	12% (45) 45% (31)	13% (121)	8.5% (34)	74% (2431)	6% (8)
Percentage of wild type (<i>n</i>)	10% (37) 2.8% (11)	59% (226) 26% (18)	30% (284)	89% (352)	15.5% (512)	94% (122)
Number of phenotypes =100%	270 / 167	112 / 20	538	10	347	–
Percentage of phenotypes classed as medium strength (<i>n</i>)	97% (262) 7.8% (13)	100% (112) 100% 20	98% (529)	50% (5)	43% (150)	–
Percentage of phenotypes classed as strong (spheres) (<i>n</i>)	3% (8) 92.2% (154)	–	2% (9)	50% (5)	57% (197)	–
Number of app.–/gut–phenotypes (<i>n</i>) =100%	–	112 / 20	529 [†]	5	–	–
Percentage of app.– only phenotypes (<i>n</i>)	–	97% (109) 100% (20)	13% (67)	100% (5)	–	–
Percentage of app. and gut phenotypes (<i>n</i>)	–	3% (3) –	87% (462)	–	–	–

app., appendages.

*3% (*n*=4). Unspecific phenotype: flagellum of the antenna missing, but as this can be caused during the embedding procedure it is counted as wild type.[†]For the comparison of fz1-fz1/4-fz4 RNAi, double stranded RNA of the same concentration was used in the single and the double RNAi experiments.

BRNAi experiments: injection series sorted by different dsRNA concentrations used**B1. Tc-fz1-RNA**

	<i>Fz1</i>	<i>Fz1</i>	<i>Fz1</i>	<i>Fz1</i>
Concentration of dsRNA	300 ng/μl	650 ng/μl	1000 ng/μl	1500 ng/μl
Total <i>n</i> analysed cuticles	177	51	155	69
Percentage of specific phenotypes (<i>n</i>)	13.6% (24)	62.7% (51)	45.1% (70)	29% (20)
Percentage of empty eggs (<i>n</i>)	1.1% (2)	37.3% (19)	15.5% (24)	45% (31)
Percentage of wild type (<i>n</i>)	85.3% (151)	–	39.4% (61)	26% (18)
Number of phenotypes =100%	24	51	70	20
Percentage of phenotypes classed as medium strength (<i>n</i>)	100% (24)	100% (51)	100% (70)	100%
Percentage of phenotypes classed as strong (spheres) (<i>n</i>)	–	–	–	–
Number of app.–/gut–phenotypes (<i>n</i>) =100%	24	51	70	20
Percentage of app.–only phenotypes (<i>n</i>)	100% (24)	96% (49)	98.5% (69)	100% (20)
Percentage of app. and gut phenotypes (<i>n</i>)	–	4% (2)	1.5% (1)	–

B2. Tc-fz2 RNA

	<i>fz2</i>	<i>fz2</i>
Concentration of dsRNA	300 ng/μl	1500 ng/μl
Total <i>n</i> analysed cuticles	26	56
Percentage of empty eggs (<i>n</i>)	50% (13)	19.6% (11)
Percentage of wild type (<i>n</i>)	50% (13)	80.4% (45)

B3. Tc-fz1+Tc-fz2 double RNAi

	<i>Fz1/2</i> (first injection series [†])	<i>Fz1/2</i>	<i>Fz1/2</i>
Concentration of dsRNA	300 ng/μl	300 ng/μl	1500 ng/μl
Total <i>n</i> analysed cuticles	62	310	389
Percentage of specific phenotypes (<i>n</i>)	45.2% (28)	78% (242)	43% (167)
Percentage of empty eggs (<i>n</i>)	4.8% (3)	20% (62)	54.2% (211)
Percentage of wild type (<i>n</i>)	50% (31)	2% (6)	2.8% (11)
Number of phenotypes =100%	28	242	167
Percentage of phenotypes classed as medium strength (<i>n</i>)	93% (26)	97% (236)	7.8% (13)
Percentage of phenotypes classed as strong (spheres) (<i>n</i>)	7% (2)	3% (6)	92.2% (154)

[†]This series was the first *fz1/2* RNAi experiment performed by R.P.

B4. Tc-fz4-RNAi

	<i>Fz4</i>	<i>Fz4</i>	<i>Fz4</i>
Concentration of dsRNA	300 ng/μl	600 ng/μl	1000 ng/μl
Total <i>n</i> analysed cuticles	115	125	156
Percentage of specific phenotypes (<i>n</i>)	0.9% (1)	3.2% (4)	3.2% (5)
Percentage of empty eggs (<i>n</i>)	0.9% (1)	17.6% (22)	7% (11)
Percentage of wild type (<i>n</i>)	98.2% (113)	79.2% (99)	89.8% (140)
Number of phenotypes =100%	1	4	5
Percentage of phenotypes classed as medium strength (<i>n</i>)	100% (1)	25% (4)	60% (3)
Percentage of phenotypes classed as strong (spheres) (<i>n</i>)	–	75% (3)	40% (2)
Number of app.–/gut–phenotypes (<i>n</i>) =100%	1	1	3
Percentage of app.– only phenotypes (<i>n</i>)	100% (1)	100% (1)	100% (3)

B5. Fz1/4-double RNAi

	<i>fz1/4</i>	<i>fz1/4</i>	<i>fz1/4</i>	<i>fz1/4</i>
Concentration of dsRNA	300 ng/μl	650 ng/μl	650 ng/μl	1000 ng/μl
Total <i>n</i> analysed cuticles	180	245	257	261
Percentage of specific phenotypes (<i>n</i>)	66.1% (119)	78% (191)	82% (210)	7% (18)
Percentage of empty eggs (<i>n</i>)	0.6% (1)	21.6% (53)	14% (37)	11.5% (30)
Percentage of wild type (<i>n</i>)	33.3% (60)	0.4% (1)	4% (10)	81.5% (213)
Number of phenotypes =100%	119	191	210	18
Percentage of phenotypes classed as medium strength (<i>n</i>)	100% (119)	97% (185)	98.5% (207)	100% (18)
Percentage of phenotypes classed as strong (spheres) (<i>n</i>)	–	3% (6)	1.5% (3)	–
Number of app.–/gut–phenotypes (<i>n</i>) =100%	119	185	207	18
Percentage of app.– only phenotypes (<i>n</i>)	56.3% (67)	–	–	–
Percentage of app. and gut phenotypes (<i>n</i>)	43.7% (52)	100% (185)	100% (207)	100% (18)

B6. Tc-arrow RNAi

	10 ng/μl	50 ng/μl	100 ng/μl	300 ng/μl	600 ng/μl	600 ng/μl	1700 ng/μl
Concentration of dsRNA	10 ng/μl	50 ng/μl	100 ng/μl	300 ng/μl	600 ng/μl	600 ng/μl	1700 ng/μl
Total <i>n</i>	796	720	601	565	337	164	107
Percentage of specific phenotypes (<i>n</i>)	9.8% (78)	17.8% (128)	7.3% (44)	7.5% (43)	11.6% (39)	9.1% (15)	–
Percentage of empty eggs (<i>n</i>)	43.2% (344)	80% (576)	78.9% (474)	86.9% (491)	86.6% (292)	90.8% (149)	98.1% (105)
Percentage of wild type (<i>n</i>)	47% (374)	2.2% (16)	13.8% (83)	5.5% (31)	1.8% (6)	–	1.9% (2)
Number of phenotypes =100%	78	128	44	43	39	15	–
Percentage of phenotypes classed as medium strength (<i>n</i>)	53.8% (42)	37.5% (48)	36.4% (16)	30% (13)	30.7% (12)	26.6% (4)	–
Percentage of phenotypes classed as strong (spheres) (<i>n</i>)	46.2% (36)	62.5% (80)	63.6% (28)	70% (30)	69.3% (27)	73.3% (11)	–