

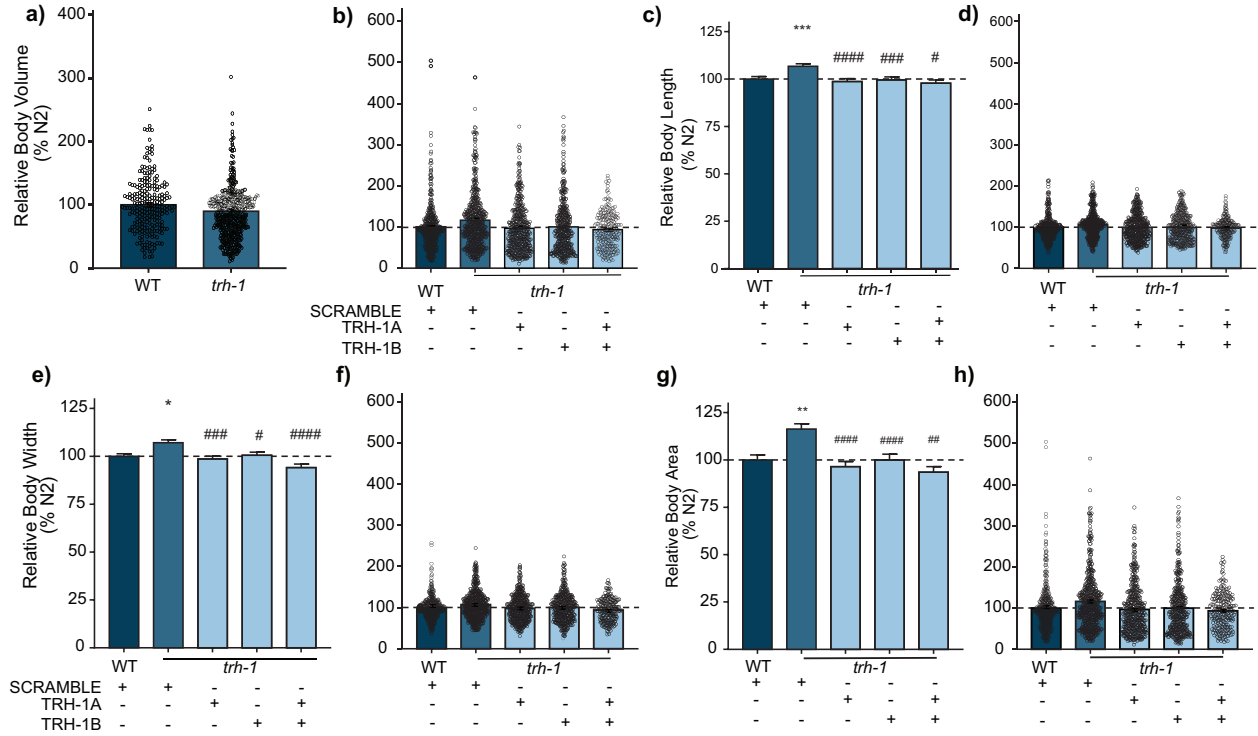
a)

Plasmid	Forward Sequence	Reverse Sequence
SCRAMBLE	GGGGACAAGTTTGTACAAAAAAGCAGGC TGGATGCGCTTTGGAAAACGTAATTCGA GCTCCACCGCGGTGGCGGCCGCTCTAGA ACTAGTGGATCCACCGGTTCCATGGCTA GCCACGCGCGTGGATCCCCGGGCTGCA GGAAAACGTAAATAACACCCAGCTTTCTT GTACAAAGTGGTCCCC	GGGGACCACTTTGTACAAGAAAGCTGG GTGTTATTTACGTTTCCTGCAGCCCGGG GGATCCACGCGCGTGGCTAGCCATGGAA CCGGTGGATCCACTAGTTCTAGAGCGGC CGCCACCGCGGTGGAGCTCGAATTACGT TTTCCAAAGCGCATCCAGCCTGCTTTTTT GTACAAACTTGTCCCC
TRH-1A	GGGGACAAGTTTGTACAAAAAAGCAGGC TGGATGCGCTTTGGAAAACGTAGAGGAC GAGAACTTTTCGAAAACGTAAATAACA CCCAGCTTTCTTGTACAAAGTGGTCCCC	GGGGACCACTTTGTACAAGAAAGCTGG GTGTTATTTACGTTTTCGAAAAGTTCTC GTCCTCTACGTTTTCGAAAAGCGCATCCA GCCTGCTTTTTTGTACAAACTTGTCCCC
TRH-1B	GGGGACAAGTTTGTACAAAAAAGCAGGC TGGATGCGCTTTGGAAAACGTCGTGCCA ATGAACTTTTCGTTAAACGTAAATAACA CCCAGCTTTCTTGTACAAAGTGGTCCCC	GGGGACCACTTTGTACAAGAAAGCTGG GTGTTATTTACGTTTACCGAAAAGTTCA TTGGCACGACGTTTTCGAAAAGCGCATCC AGCCTGCTTTTTTGTACAAACTTGTCCCC
INS-6	CAATGCCACGAGCAAGTAGTGTTCCAG CACCAG	CTGGTGCTGGAACACTACTTGCTCGTGG CATTG
PDF-1A	GGGGACAAGTTTGTACAAAAAAGCAGGC TGGGTTCAAGTTCGTAACACGCAACG CCGAGCTTATCAACGGACTCATCGGAAT GGATTTGGGAAAATTGTCAGCTGTCGGA AAACGCTGACACCCAGCTTTCTTGTACA AAGTGGTCCCC	GGGGACCACTTTGTACAAGAAAGCTGG GTGTCAGCGTTTTCGACAGCTGACAAT TTTCCCAAATCCATTCCGATGAGTCCGT GATAAGCTCGGCGTTGCTGCGTTTTACG AACTGAACCCAGCCTGCTTTTTTGTACA AACTTGTCCCC
PDF-1B	GGGGACAAGTTTGTACAAAAAAGCAGGC TGGGTTCAAGTTCGTAACACGCTCAAACG CGGAAGTTATCAACGGTCTTCTCAGCAT GAACCTCAACAAATTGCTGGAGCTGGT CGACGATGACACCCAGCTTTCTTGTACA AAGTGGTCCCC	GGGGACCACTTTGTACAAGAAAGCTGG GTGTCATCGTCGACCAGCTCCAGACAAT TTGTTGAGGTTTCATGCTGAGAAGACCGT TGATAAGTTCCGCGTTTGGAGCGTTTAC GAACTGAACCCAGCCTGCTTTTTTGTAC AACTTGTCCCC

b)

Peptide	Sequence (COOH-x-NH ₂)
SCRAMBLE	NSKLHRGGGRSRTSGSTGSMASHARGSPGLQ
TRH-1A	GRELF
TRH-1B	ANELF
INS-6	VPAPGETRACGRKLISLVMVCGDLCNPQEGKDIAATECCGNQCSDDYIRSACCP
PDF-1A	VQFVKRSNAELINGLIGMDLGKLSAVGKR
PDF-1B	VQFVKRSNAELINGLLSMNLNKLSGAGR

Supplementary Table 1. Neuropeptide sequences used in this study. a) Sequences of forward and reverse oligos used in generation of plasmids containing the peptide of interest for functional rescue of the neuropeptides b) Processed peptide amino acid sequences used for bacterial rescue experiments.



Supplementary Figure 1. TRH-1 peptide feeding affects body length, width, and area of

trh-1 mutants. **a)** Relative body volume of *E. coli* OP50 fed worms, displaying all data points

(Mann-Whitney test, ** $p = 0.0012$). **b)** Relative body volume of peptide worms, all data points

wild-type fed scramble, *trh-1* fed scramble, TRH-1A, TRH-1B, or both TRH-1A+B. **c)** Relative

body length of peptide fed worms. **d)** Relative body length of peptide fed worms, all data points.

e) Relative body width of worms fed peptide. **f)** Relative body width of peptide fed worms, all

data points. **g)** Relative body area of peptide fed worms. **h)** Relative body area of peptide fed

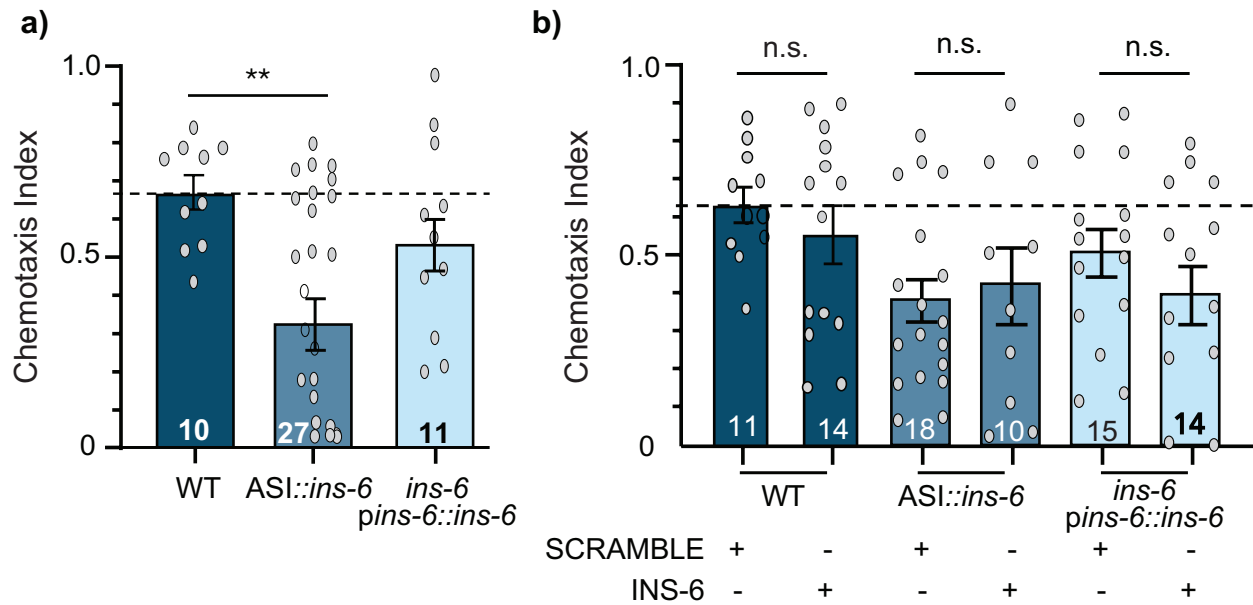
worms displaying all data points. n for all panels wild-type = 454, *trh-1* fed scramble = 559, fed

TRH-1A = 483, fed TRH-1B = 441, fed TRH-1A+B = 221. Error bars denote SEM. (b-g)

Kruskal-Wallis followed by Dunn's multiple comparison tests, */# $p < 0.05$, **/##, $p < 0.01$,

/### $p < 0.001$, */#### $p < 0.0001$, *denote mutant worms compared to wild-type control,

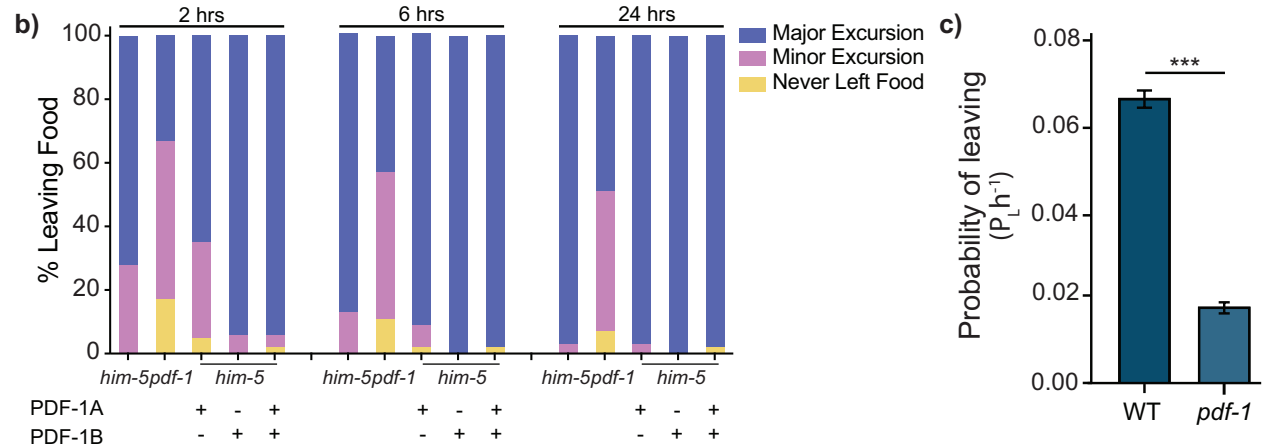
#denote mutant worm condition compared to mutant worms fed scramble peptide.



Supplementary Figure 2. Chemotaxis Index of partially rescued *ins-6* *lof* worms to 750mM NaCl without and with peptide feeding. **a)** Chemotaxis Index to 750 mM NaCl of wild-type, *ins-6* rescued specifically in ASI neurons and a complete genetic rescue of *ins-6* animals. Rescue of *ins-6* in ASI neurons does not result in NaCl chemotaxis, suggesting that this neuron does not play a role in this behavior. **b)** Chemotaxis Index of INS-6 peptide-fed wild-type and genetically rescued *ins-6* worms. Overexpression of INS-6 by feeding does not affect chemotaxis in wild type animals and the different rescue lines of *ins-6*. *n* values denoted in graphs. Error bars denote SEM. One-Way ANOVA, followed by Bonferroni's Correction. * $p < 0.05$, ** $p < 0.01$.

a)

Genotype	Treatment	P_L (confidence interval)	[n]
<i>him-5</i>	OP50	0.06553819 (0.05587592 , 0.07687131)	[72]
<i>him-5</i>	SCRAMBLE	0.07727273 (0.06531852 , 0.09141473)	[55]
<i>him-5</i>	PDF-1A	0.06979167 (0.05892111 , 0.08266776)	[60]
<i>him-5</i>	PDF-1B	0.08712121 (0.0754002 , 0.1006643)	[66]
<i>him-5</i>	PDF-1A+B	0.07720588 (0.06483647 , 0.09193512)	[51]
<i>pdf-1</i>	OP50	0.01785714 (0.0134571 , 0.02369585)	[84]
<i>pdf-1</i>	SCRAMBLE	0.03537736 (0.02746855 , 0.04556329)	[53]
<i>pdf-1</i>	PDF-1A	0.03286638 (0.02557212 , 0.04224128)	[58]
<i>pdf-1</i>	PDF-1B	0.05821078 (0.04760713 , 0.07117621)	[61]
<i>pdf-1</i>	PDF-1A+B	0.04473039 (0.03556126 , 0.0562637)	[51]



Supplementary Figure 3. Leaving behavior of *him-5* and *pdf-1* males fed PDF-1 peptides. a)

Table displaying the calculated Probability of Leaving P_L for each peptide feeding condition in *him-5* and *pdf-1* males. The table also depicts the confidence interval of the data and the sample size for each condition. **b)** Percentage of *him-5* males leaving food after being fed PDF-1 peptides on *E. coli* OP50. PDF-1B feeding results in complete rescue of food-leaving behavior of *pdf-1* *lof* males at all three timepoints **c)** Computation of Probability of leaving for *him-5* and *pdf-1* males fed *E. coli* OP50, *n* noted in **a)** [n] column. Error bars denote SEM. **b)** ANOVA, followed by Bonferroni's Correction, **c)** Two-tailed *t*-test of samples of equal variance. *** $p < 0.001$.