

Supplementary Table S1. This file is provided as a separate Excel file, and it contains a list of published QTL in threespine (*G. aculeatus*, *G. nipponicus*) and ninespine (*P. pungitius*) stickleback. Physical positions for markers and confidence intervals are given for three threespine stickleback assembly versions [14,17,18]. QTL were loosely grouped into nine functional categories: feeding (teeth, gill rakers, branchial bones, jaw, epaxial muscle), body shape (univariate measures, landmark x- and y-coordinates), defence (lateral plate number and size, dorsal spines, pelvic spines and girdle), behaviour/sensory system (behaviours, lateral line), swimming (vertebrae number, pterygiophore position and number, fin ray position and number), pigmentation (reproductive coloration, melanisation, barring), respiration (opercle size and shape), body size (centroid size, standard length, body weight), and reproduction (testes size, sperm number).

Species	Crosses	Ecotypes	Trait categories	Cross type	Number of progeny	Gene identification	QTL refs
<i>G. aculeatus</i>	BC (Priest benthic female x Priest limnetic male) x BC Priest benthic female	benthic-limnetic	defence, feeding	BCx	92		[10]
<i>G. aculeatus</i>	JP Pacific Ocean marine female x BC Paxton benthic male	marine-freshwater	defence	F2	360	Plate morph: <i>Eda</i> [64]; Plate size: <i>Gdf6</i> [70]	[23]
<i>G. aculeatus</i>	AK Bear Paw, Boot, Whale lakes x AK Rabbit Slough marine	marine-freshwater	defence	F2	98, 116, 77	Plate morph: <i>Eda</i> [64]; Pelvic reduction: <i>Pitx1</i> [68]	[24]
<i>G. aculeatus</i>	JP Pacific Ocean marine female x BC Paxton benthic male	marine-freshwater	defence	F2	375	Pelvic reduction: <i>Pitx1</i> [25,68]	[25]
<i>G. aculeatus</i>	AK Bear Paw lake x AK Rabbit Slough marine	marine-freshwater	respiration	F2	98		[26]
<i>G. aculeatus</i>	SC Loch Fada, North Uist female x SC River Kelvin, Glasgow male	lake-stream	defence	F2	177	Pelvic reduction: <i>Pitx1</i> [68]	[27]
<i>G. aculeatus</i>	JP Pacific Ocean marine female x BC Paxton benthic male	marine-freshwater	pigmentation	F2	360	Pigmentation: <i>Kitlg</i> [28]	[28]
<i>G. aculeatus</i>	JP Pacific Ocean marine female x BC Paxton benthic male	marine-freshwater	body shape	F2	372		[29]
<i>G. aculeatus</i> ; <i>G. nipponicus</i>	JP Pacific Ocean marine female x JP Japan Sea marine male; (Japan Sea female x Pacific Ocean male) x JP Pacific Ocean male	marine-marine	body size, behavior and sensory system; defence, reproduction	F2, BCx	70, 76		[30]
<i>G. aculeatus</i>	BC Hotel Lake female x BC Little Campbell marine male	marine-freshwater	body size, pigmentation	F2	176		[31]
<i>G. aculeatus</i>	BC Enos Lake benthic-limnetic admixed population	benthic-limnetic	body shape, pigmentation	admix	508		[32]
<i>G. aculeatus</i>	BC Little Campbell marine female x BC Paq, Graham, Hoggan, Cranby lake males	marine-freshwater	body shape, defence, feeding	F2	374, 361, 290, 374		[33]
<i>G. aculeatus</i>	BC Paxton benthic female x JP Pacific Ocean marine male	marine-freshwater	behavior and sensory system, defence	F2	234	Lateral line: <i>Eda</i> [66]	[34]
<i>G. aculeatus</i>	BC Paxton benthic female x JP Pacific Ocean marine male	marine-freshwater	behavior and sensory system	F2	229	Schooling: <i>Eda</i> [67]	[35]
<i>G. aculeatus</i>	BC Paxton limnetic x BC Paxton benthic	benthic-limnetic	body shape, defence, feeding	F2	530		[36]
<i>G. aculeatus</i>	CH Lake Geneva stream female x CH Lake Constance lake male	lake-stream	body shape, defence, feeding, swimming	F2	377		[37]

<i>G. aculeatus</i>	JP Pacific Ocean marine female x BC Paxton benthic male	marine-freshwater	feeding	F2	272	Tooth number: <i>Bmp6</i> [38]	[38]
<i>G. aculeatus</i>	BC Little Campbell marine female x WA Fish Trap Creek male; BC Little Campbell marine female x BC Paxton benthic male	marine-freshwater	feeding	F2	279, 270		[39]
<i>G. aculeatus</i>	WA Fish Trap Creek female x BC Little Campbell marine male; BC Little Campbell marine female x AK Bear Paw lake, BC Paxton benthic males	marine-freshwater	feeding	F2	273, 384, 418		[40]
<i>G. aculeatus</i>	FI Baltic Sea marine female x FI Lake Pulmanki male	marine-freshwater	body shape, body size, defence	F2	190		[41]
<i>G. aculeatus</i>	JP Pacific Ocean marine female x BC Paxton benthic male	marine-freshwater	body shape, defence, feeding, respiration, swimming	F2	370		[42]
<i>G. aculeatus</i> ; <i>G. nipponicus</i>	JP (Japan Sea female x Pacific Ocean male) x JP Pacific Ocean male	marine-marine	defence	BCx	76		[43]
<i>G. aculeatus</i>	BC Paxton limnetic female x BC Paxton benthic male; BC Priest limnetic female x BC Priest benthic male	benthic-limnetic	body shape, defence, feeding	F2	403, 323		[44]
<i>G. aculeatus</i>	BC Little Campbell marine female x CA Cerrito Creek freshwater male	marine-freshwater	feeding	F2	171		[45]
<i>G. aculeatus</i>	WA Fish Trap Creek female x BC Little Campbell marine male; BC Little Campbell marine female x AK Bear Paw lake male	marine-freshwater	defence, feeding	F2	360, 363		[18]
<i>G. aculeatus</i>	BC Paxton benthic female x JP Pacific Ocean marine male	marine-freshwater	behavior and sensory system	F2, BCx	376, 95		[46]
<i>G. aculeatus</i>	BC Paxton, Priest, Enos benthic females x BC Little Campbell marine male	marine-freshwater	body shape, defence, feeding	F2	186, 180, 180		[47]
<i>G. aculeatus</i>	(CA Matadero stream female x BC Paxton limnetic male) x CA Matadero female	stream-limnetic	pigmentation	BCx	429		[48]
<i>P. pungitius</i>	NWT Fox Holes Lake female x AK Pt McKenzie creek	lake-stream	body shape, defence	F1	120		[49]
<i>P. pungitius</i>	FI Baltic Sea female marine x FI Ryttilampi pond male	marine-freshwater	body size	F2	283		[50]

<i>P. pungitius</i>	FI Baltic Sea female marine x FI Rytilampi pond male	marine-freshwater	defence	F2	283	[51]
<i>P. pungitius</i>	FI Baltic Sea female marine x FI Rytilampi pond male	marine-freshwater	behavior and sensory system	F2	283	[52]

Supplementary Table S2. Published QTL studies in threespine (*G. aculeatus*, *G. nipponicus*) and ninespine (*P. pungitius*) stickleback. The mapping crosses used in each study are indicated, including the geographic origin of the populations: Alaska, USA (AK), British Columbia, Canada (BC), California, USA (CA), Switzerland (CH), Finland (FI), Japan (JP), Northwest Territories, Canada (NWT), Scotland, UK (SC), Washington, USA (WA). The ecotype comparisons, trait categories, cross type (backcross (BCx), F2 intercross (F2), admixture mapping (admix)), and number of progeny in the crosses for each study are indicated. If a gene has been identified that underlies a QTL found in a particular study, the trait name, gene name, and reference is indicated. Reference numbers refer to those in the main text.

Trait	Species	Chr	PVE	Gene	Gene ID method and evidence	Coding vs regulatory	Same vs different gene or mutation	Standing variation vs new mutation	References
Plate morph	<i>G. aculeatus</i>	IV	77.6	<i>Eda</i>	a, b, e, f	regulatory	same gene	standing variation	[23,64,65]
Lateral line patterning	<i>G. aculeatus</i>	IV	64.8	<i>Eda</i>	a, f	regulatory?	not tested	unknown	[34,66]
Schooling body position	<i>G. aculeatus</i>	IV	13.5	<i>Eda</i>	a, f	regulatory?	not tested	unknown	[35,67]
Pelvic loss	<i>G. aculeatus</i>	VII	100	<i>Pitx1</i>	a, b, e, f	regulatory	same gene; different mutations	new mutation	[25,68]
Pelvic loss	<i>P. pungitius</i>	VII	ND	<i>Pitx1</i>	c, e	regulatory	same and different genes	unknown	[49,51,69]
Plate size	<i>G. aculeatus</i>	XX	9.9	<i>Gdf6</i>	a, d, e, f	regulatory	same gene?	standing variation?	[23,70]
Tooth number	<i>G. aculeatus</i>	XXI	30	<i>Bmp6</i>	a, e	regulatory	different gene	unknown	[38,42,45]
Pigmentation	<i>G. aculeatus</i>	XIX	56.1	<i>Kitlg</i>	a, e	regulatory	same gene	standing variation	[28]

Supplementary Table S3. Identification of genes that underlie phenotypic diversity in threespine stickleback. A combination of ^ahigh resolution linkage mapping, ^bassociation mapping, ^ccomplementation crosses, and ^dgenome scans have been used to identify candidate genes; ^eallele specific expression and ^ftransgenic manipulation have been used to provide evidence that the gene does have an effect on the phenotype of interest. Reference numbers refer to those in the main text.

Chr	Length	Genes	Number of QTL									
			Total	Feeding	Shape	Defence	Behaviour	Swimming	Pigmentation	Respiration	Body size	Reproduction
I	29.63	1,328	92	26	46 ^{a,b}	7	0	5	4	2	1	1
II	23.70	907	31 ^c	15	8 ^c	6	0	1	0	1	0	0
III	17.80	1,004	12 ^{c,d}	4 ^{c,d}	6 ^d	2	0	0	0	0	0	0
IV	34.14	1,410	205 ^{a,b}	89 ^{a,b}	58 ^{a,b}	43 ^{a,b}	4	8	3	0	0	0
V	15.56	861	25 ^d	12	6	2	4	1	0	0	0	0
VI	18.85	760	14 ^{c,d}	2 ^{c,d}	3 ^{c,d}	2	2	0	4	0	1	0
VII	30.84	1,481	119 ^{a,b}	40	41	32 ^{a,b}	0	0	4	2	0	0
VIII	20.53	924	43	26	11	4	1	1	0	0	0	0
IX	20.58	1,016	26 ^{c,d}	9	4 ^{c,d}	8	4	0	1	0	0	0
X	18.03	931	28 ^d	14	4 ^{c,d}	8	1	1	0	0	0	0
XI	17.64	1,108	27 ^d	10	3 ^{c,d}	9	4	0	1	0	0	0
XII	20.76	1,138	57	11	40 ^{a,b}	1	2	0	1	2	0	0
XIII	20.74	1,014	31 ^{c,d}	6 ^{c,d}	19	5	1	0	0	0	0	0
XIV	16.17	792	30	17	13	0	0	0	0	0	0	0
XV	17.32	823	13 ^{c,d}	8	4 ^{c,d}	0	0	1	0	0	0	0
XVI	19.52	864	72 ^{a,b}	17	50 ^{a,b}	5	0	0	0	0	0	0
XVII	20.25	1,064	36	23	7 ^d	2	1	1	0	1	1	0
XVIII	15.99	739	24	6	14	2	0	0	0	1	1	0
XIX	20.61	1,086	35 ^d	9	9	4	0	0	2	2	5	4
XX	20.45	990	78 ^{a,b}	45 ^{a,b}	18	10	2	3	0	0	0	0
XXI	17.35	614	106 ^{a,b}	33 ^{a,b}	35 ^{a,b}	23 ^{a,b}	9	5	0	0	1	0
Total	436.46	20,854	1,104	422	399	175	35	27	20	11	10	5

Supplementary Table S4. Distribution of QTL on stickleback chromosomes. For each chromosome (Chr), the chromosome length in megabases and number of genes are based on the assembly of Glazer *et al.* [18], and the number of QTL in each trait category are taken from electronic supplementary material table S1. Using a goodness-of-fit test (chisq.test in R), we found that the observed number of QTL per chromosome differs significantly from the expected number of QTL, given either the length of the chromosome in megabases or the number of genes on the chromosome, for all QTL (length: $\chi^2_{20} = 446.19$, $p < 2.2\text{e-}16$; genes: $\chi^2_{20} = 611.33$, $p < 2.2\text{e-}16$), feeding QTL (length: $\chi^2_{20} = 217.18$, $p < 2.2\text{e-}16$; genes: $\chi^2_{20} = 275.49$, $p < 2.2\text{e-}16$), body shape QTL (length: $\chi^2_{20} = 234.38$, $p < 2.2\text{e-}16$; genes: $\chi^2_{20} = 282.70$, $p < 2.2\text{e-}16$), and defence QTL (length: $\chi^2_{20} = 180.15$, $p < 2.2\text{e-}16$; genes: $\chi^2_{20} = 224.96$, $p < 2.2\text{e-}16$). The remaining trait categories had too few QTL for statistical analyses. Specific chromosomes with more or fewer QTL than expected were identified by examining the standardized residuals for each chromosome; following Agresti [1], chromosomes with a standardized residual of greater than 3 had more QTL than expected based on ^achromosome length or ^bnumber of genes, while chromosomes with a standardized residual of less than -3 had fewer QTL than expected based on ^cchromosome length or ^dnumber of genes.

Supplementary material for Peichel and Marques, 2016, The genetic and molecular architecture of phenotypic diversity in sticklebacks, *Phil. Trans. R. Soc. B.*
doi: 10.1098/rstb.2015.0486

References

1. Agresti A. 2007 *An introduction to categorical data analysis*. Hoboken, NJ: John Wiley & Sons.