

Figure S1

a

```
B1_lim1-int1_ARE 1 GGTGAACTGAGCGGGATAGCTT-----AATGGGGTTAGGGAAACGTCGAATCATGTT 55
Bf_lim1-int1_ARE 1 CA...TT...T...A.....C.....TTAC...A...AATGGAT...T...A.....A 47
Bb_lim1-int1_ARE 1 ---GTA...GTA...A...GAG...A...A.....GTC...CTCG...A.....GAG...A.... 52
Bj_lim1-int1_ARE 1 GAT...GAT...A...GAACTTGG...A...C...CAT...A...A.....A 58

B1_lim1-int1_ARE 56 CCGCGCCAGAGATAATGGAGGCACGGGGCTACTTGACGCGTTC...TAAAGAC... 113
Bf_lim1-int1_ARE 48 GG...T...G...C...G...G...G...G...G...A...G...A...G...A...A..... 105
Bb_lim1-int1_ARE 53 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 112
Bj_lim1-int1_ARE 59 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 116

B1_lim1-int1_ARE 114 TCTACGAAAGATAATCATTATAAACCTCGAGGAACCCGACGGAGTGAAGTGAAGNGC... 173
Bf_lim1-int1_ARE 106 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 165
Bb_lim1-int1_ARE 113 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 171
Bj_lim1-int1_ARE 117 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 176

B1_lim1-int1_ARE 174 TAAAGAGCTTTAAIGAAAGCGGGTAATGGGGCTCTGCAATGAT... 233
Bf_lim1-int1_ARE 166 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 224
Bb_lim1-int1_ARE 172 CTC...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 231
Bj_lim1-int1_ARE 177 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 236

B1_lim1-int1_ARE 234 TGTCCTGAGAACCCATAAACTGGGGCTACGGGGCTATTGTGAT... 288
Bf_lim1-int1_ARE 225 ---TTAACTGGGCTTGTGATTAACATGGTGTATAAGGGTGTTTGTAC... 291
Bb_lim1-int1_ARE 232 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 288
Bj_lim1-int1_ARE 237 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 293

B1_lim1-int1_ARE 289 -----AATTCGGGTTATTGTATAAACATGGTGTATAAGGGTGTTTGTAC... 343
Bf_lim1-int1_ARE 282 TTGGC...A...A...A...A...A...A...A...A...A...A...A...A...A... 317
Bb_lim1-int1_ARE 289 TTGGC...A...A...A...A...A...A...A...A...A...A...A...A...A... 348
Bj_lim1-int1_ARE 294 TTGGC...A...A...A...A...A...A...A...A...A...A...A...A...A... 353

B1_lim1-int1_ARE 344 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 344
Bf_lim1-int1_ARE 318 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 373
Bb_lim1-int1_ARE 349 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 395
Bj_lim1-int1_ARE 354 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 402

B1_lim1-int1_ARE 391 TTTTGTTCTGCTTCGGGACGACACCAAAGAGGAACTAACCCAAAATGAC... 449
Bf_lim1-int1_ARE 374 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 432
Bb_lim1-int1_ARE 396 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 454
Bj_lim1-int1_ARE 403 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 461

B1_lim1-int1_ARE 450 AATTTAIGTCATGTGAAATTATGGGGCTCTGCAATGGGGCTTATG... 507
Bf_lim1-int1_ARE 433 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 491
Bb_lim1-int1_ARE 455 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 512
Bj_lim1-int1_ARE 462 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 519

B1_lim1-int1_ARE 508 ATGACATATACATCTAACCTGGCAATGATGGTACGGGGCTCTGCA... 550
Bf_lim1-int1_ARE 492 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 547
Bb_lim1-int1_ARE 513 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 566
Bj_lim1-int1_ARE 520 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 579

B1_lim1-int1_ARE 551 ATCAGCT----AAATCTTGTATTAATTAACGGGATGGGGCTCTGGAG... 605
Bf_lim1-int1_ARE 548 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 594
Bb_lim1-int1_ARE 567 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 626
Bj_lim1-int1_ARE 580 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 632

B1_lim1-int1_ARE 606 GGGCGGGTGTACGCTAGCTATTTGTTGAACTGGGGTTCTGAT... 654
Bf_lim1-int1_ARE 595 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 640
Bb_lim1-int1_ARE 627 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 678
Bj_lim1-int1_ARE 633 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 688
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b

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B1_chrd-int2_CNS1 1 ACGTAA---TTGGTATGTTAATGATTGTGATGGGGCTCTGCAATGG... 56
Bf_chrd-int2_CNS1 1 ---GCG-TTA...TCATGTTTGATGATGTTGTGATGGGGCTCTGCA... 57
Bb_chrd-int2_CNS1 1 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 58
Bj_chrd-int2_CNS1 1 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 58

B1_chrd-int2_CNS1 57 CGAACTATTAATGGGGCTCTGCAATGGGGCTCTGCAATGG... 107
Bf_chrd-int2_CNS1 58 A...G...G...G...G...G...G...G...G...G...G...G...G...G... 110
Bb_chrd-int2_CNS1 59 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 118
Bj_chrd-int2_CNS1 59 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 118

B1_chrd-int2_CNS1 108 TCTGTTGTAAAGGGGTGGGGCTCTGCAATGGGGCTCTGCA... 163
Bf_chrd-int2_CNS1 111 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 167
Bb_chrd-int2_CNS1 119 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 178
Bj_chrd-int2_CNS1 104 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 161

B1_chrd-int2_CNS1 164 CCGGTGTTGGGGCTCTGCAATGGGGCTCTGCAATGG... 216
Bf_chrd-int2_CNS1 168 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 226
Bb_chrd-int2_CNS1 172 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 231
Bj_chrd-int2_CNS1 162 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 216

B1_chrd-int2_CNS1 217 GTGTGATGTCGTTGGGGCTCTGCAATGGGGCTCTGCA... 263
Bf_chrd-int2_CNS1 227 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 280
Bb_chrd-int2_CNS1 232 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 291
Bj_chrd-int2_CNS1 217 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 268

B1_chrd-int2_CNS1 264 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 318
Bf_chrd-int2_CNS1 281 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 340
Bb_chrd-int2_CNS1 292 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 351
Bj_chrd-int2_CNS1 269 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 328

B1_chrd-int2_CNS1 319 TCGGGATATAATGAATGGGGCTCTGGGGCTCTGCAATGGGG... 378
Bf_chrd-int2_CNS1 341 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 400
Bb_chrd-int2_CNS1 352 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 411
Bj_chrd-int2_CNS1 329 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 388

B1_chrd-int2_CNS1 379 CGTAAAAGCAATGTCATGTTATGGGGCTCTGCAATGGGGCT... 434
Bf_chrd-int2_CNS1 401 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 458
Bb_chrd-int2_CNS1 412 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 468
Bj_chrd-int2_CNS1 389 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 444

B1_chrd-int2_CNS1 435 TTITTGCTAAATAGGTACCTTGATTTACGATGGGGCTCTGCA... 493
Bf_chrd-int2_CNS1 459 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 518
Bb_chrd-int2_CNS1 469 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 527
Bj_chrd-int2_CNS1 445 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 503

B1_chrd-int2_CNS1 494 AATGTTTAGCTGCTGCTGCTGGGGCTCTGCAATGGGGCT... 543
Bf_chrd-int2_CNS1 519 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 575
Bb_chrd-int2_CNS1 528 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 567
Bj_chrd-int2_CNS1 504 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 557

B1_chrd-int2_CNS1 544 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 576
Bf_chrd-int2_CNS1 576 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 604
Bb_chrd-int2_CNS1 568 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 596
Bj_chrd-int2_CNS1 558 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 586
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c

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B1_chrd-int2_CNS2 1 GGTGGCGTGGGGCTGAGACGGGGCTGGATGGGGCTCTGCAATGG... 60
Bf_chrd-int2_CNS2 1 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 60
Bb_chrd-int2_CNS2 1 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 60
Bj_chrd-int2_CNS2 1 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 60

B1_chrd-int2_CNS2 61 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 120
Bf_chrd-int2_CNS2 61 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 120
Bb_chrd-int2_CNS2 61 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 120
Bj_chrd-int2_CNS2 61 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 120

B1_chrd-int2_CNS2 121 GGGCGCCGACCGGGCTGGGGCTCTGCAATGGGGCTCTGCA... 138
Bf_chrd-int2_CNS2 121 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 138
Bb_chrd-int2_CNS2 121 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 138
Bj_chrd-int2_CNS2 121 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 180

B1_chrd-int2_CNS2 138 ----- 138
Bf_chrd-int2_CNS2 138 ----- 138
Bb_chrd-int2_CNS2 138 ----- 138
Bj_chrd-int2_CNS2 241 AAAAAGATCCCAGTCAAGAGAAAAGGGGGATAAGAAGCCCCAAACGCC... 300

B1_chrd-int2_CNS2 139 ----- 155
Bf_chrd-int2_CNS2 139 ----- 155
Bb_chrd-int2_CNS2 139 ----- 155
Bj_chrd-int2_CNS2 301 TACAAAAATAAGCCAAAACCTCAACTAGGGCTCTGCAATGG... 360

B1_chrd-int2_CNS2 156 GGGGGGATTTGGAGGGCTGGGGCTCTGCAATGGGGCTCTGCA... 215
Bf_chrd-int2_CNS2 156 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 223
Bb_chrd-int2_CNS2 156 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 214
Bj_chrd-int2_CNS2 361 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 419

B1_chrd-int2_CNS2 216 TGACAAGCTGCTGCTGGGGCTCTGCAATGGGGCTCTGCA... 247
Bf_chrd-int2_CNS2 214 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 243
Bb_chrd-int2_CNS2 214 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 271
Bj_chrd-int2_CNS2 420 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 459
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d

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B1_gsc ARE 1 GCGGCGCGGTTGGGGCTCTGCAATGGGGCTCTGCAATGGGGCTCTGCA... 52
Bf_gsc ARE 1 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 52
Bb_gsc ARE 1 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 53
Bj_gsc ARE 1 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 51

B1_gsc ARE 53 ----- 107
Bf_gsc ARE 61 TAATGAGCTAATGAGCTGGGGCTCTGCAATGGGGCTCTGCAATGG... 120
Bb_gsc ARE 54 GG---G---CAAGCTCTCTCTCTCTCTCTCTCTCTCTCTCTCTCTCTCT... 109
Bj_gsc ARE 52 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 105

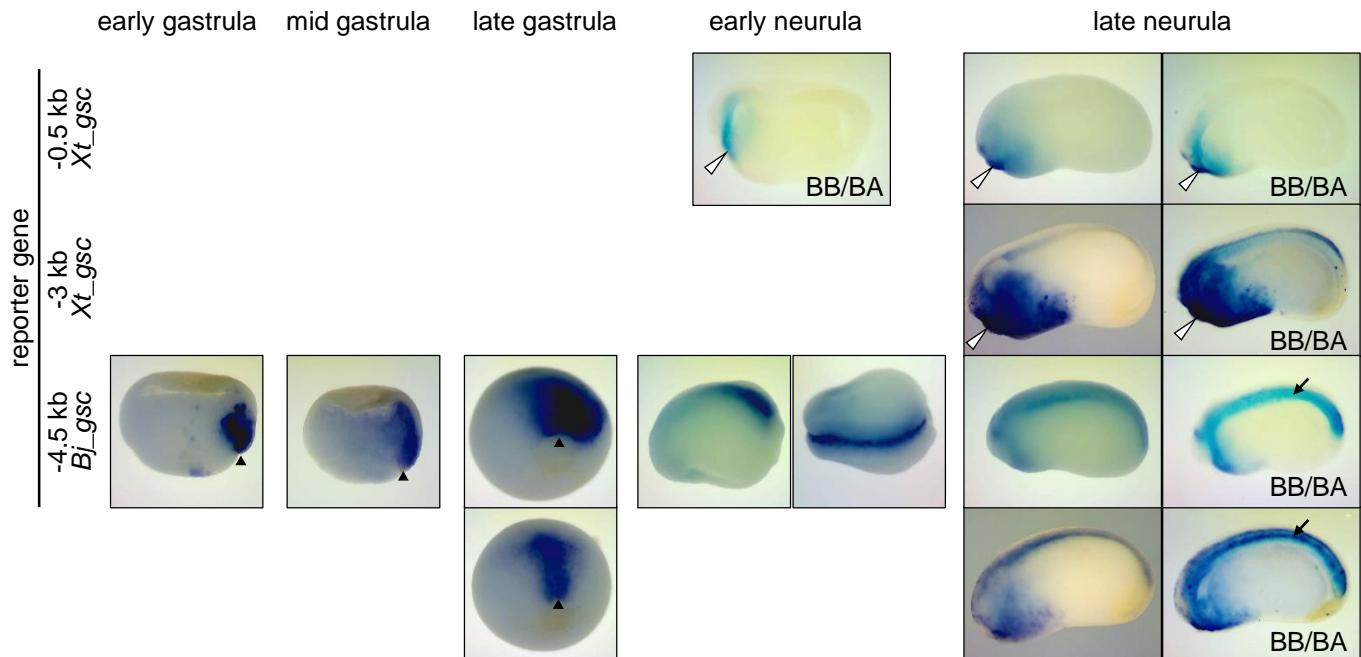
B1_gsc ARE 53 ----- 107
Bf_gsc ARE 61 TAATGAGCTAATGAGCTGGGGCTCTGCAATGGGGCTCTGCAATGG... 120
Bb_gsc ARE 54 GG---G---CAAGCTCTCTCTCTCTCTCTCTCTCTCTCTCTCTCTCTCT... 109
Bj_gsc ARE 52 G...G...G...G...G...G...G...G...G...G...G...G...G...G...G... 105

B1_gsc ARE 108 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 161
Bf_gsc ARE 121 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 158
Bb_gsc ARE 110 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 164
Bj_gsc ARE 106 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 165

B1_gsc ARE 162 CGCTGGGCTCTGCAATGGGGCTCTGCAATGGGGCTCTGCAATGG... 192
Bf_gsc ARE 159 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 211
Bb_gsc ARE 165 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 196
Bj_gsc ARE 166 G...G...G...G...G...G...G...G...G...G...G...G...G...G... 195
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Lim1 motif FoxH1 motif Smad motif

Figure S2



Reporter	Stage	Reproducible expr.	Ectopic expr.	No expr.	Total
-0.5 kb <i>Xt_gsc</i>	Mid-Late Neurula	13 (Mouth)	0	9	22
-3 kb <i>Xt_gsc</i>	Early-Mid gastrula	13 (Organizer)	6	22	41
	Late gastrula	14 (Head organizer)	9	33	56
	Mid-Late Neurula	13 (Mouth + neural tissue)	1	2	16
-4.5 kb <i>Bj_gsc</i>	Early-mid gastrula*	5 (Organizer) 28 (Organizer)	1 14	17 6	23 48
	Late gastrula*	2 (Posterior mesoderm) 17 (Posterior mesoderm)	3 3	26 10	31 30
	Mid-Late Neurula*	19 (notochord + neural tissue) 40 (notochord + neural tissue)	2 12	13 22	34 74

*results of two independent experiments are shown in separate lines