

SUPPLEMENTARY MATERIALS FOR “High density Genetic Linkage map of the southern blue-ringed octopus (*Hapalochlaena maculosa*)”

Table S1. Summary of *Hapalochlaena maculosa* families generated in this study. Bolded rows represent families which passed filtering and were included in linkage map generation. n = size of family.

Female	Male	Family_ID	n (pre filtering)	Status
131	127	131_127	37	Included
	128	131_128	10	excluded due to small size
132	142	132_142	43	Included
	143	132_143	5	excluded due to small size
133	132	133_132	33	Included
	Unknown male 2	133_02	16	excluded due to unknown male
	131	133_131	6	excluded due to small size
136	134	136_134	26	Included
	145	136_145	26	Included
137	Unknown male 1	137_01	35	excluded due to unknown male
	136	137_136	13	male removed by filtering
138	133	138_133	37	Included
	135	138_135	8	excluded due to small size
140	155	140_155	27	Included
	149	140_149	14	Included
144	144	144_144	36	Included
	151	144_151	5	excluded due to small size
145	148	145_148	34	Included
	138	145_138	3	excluded due to small size
146	139	146_139	24	Female removed by filtering
	Unknown male 3	146_03	15	excluded due to unknown male
	137	146_137	11	Female removed by filtering
	Unknown male 4	146_04	5	excluded due to unknown male
148	153	148_153	11	Both parents removed by filtering
	156	148_156	6	excluded due to small size
149	Unknown male 5	149_05	35	excluded due to unknown male
	154	149_154	9	excluded due to small size
	158	149_158	4	excluded due to small size

Table S2. Filtering and quality assessment of SNPs produced using DARTQC. Rep average is a metric specific to DArT (Diversity Arrays) specifying the proportion of alleles that give repeatable results over 30 replicates. MAF (Minor Allele Frequency)

FILTER	PARAMETERS	SILENCED SAMPLES	SILENCED SNPS	SILENCED CALLS	CHANGED CALLS
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Read Counts	5	0	0	4510437	0
Min SNP Call Rate	0.7	0	17542	0	0
Count Comparison	['0.05', '0.1']	0	0	11462	4353
DART SNP Metric	('RepAvg', '<', 0.9)	0	0	0	0
Cluster	0.95	0	1166	0	0
MAF	['0.02']	0	1460	0	0
TOTAL SILENCED		0	20168	4521899	4353
TOTAL IN DATASET		548	24199	13261052	13261052

Table S3. SNPs removed through filtering in ten *Haplochlaena maculosa* families using PLINK.

Family	SNPs Post DARTQC filtering	family size	Missing genotype threshold (0.1)	SNPs removed (MAF 0.05)	Mendel errors excluded (SNPs 0.1, individuals 0.1)	individuals retained	SNPs retained
F131_M127	7240	37	1656	3370	289	33	1925
F132_M142	7240	42	1275	3529	275	42	2161
F133_M132	7240	24	2331	2897	305	24	1707
F136_M134	7240	28	2476	3256	184	28	1324
F136_M145	7240	23	2828	2606	315	23	1491
F138_M133	7240	36	1126	3680	426	33	2008

F140_M149	7240	14	2198	2758	0	11	2284
F140_M155	7240	27	1987	3352	299	27	1602
F144_M144	7240	36	1125	3722	363	36	2030
F145_M148	7240	26	1615	3481	395	26	1749

Table S4. Characteristics and summary of *Haplochl​aena maculosa* Chromonomer assembly

LG	Length (Mb)	Markers per Scaffold				Number of genes
		1	2	> 2	Total	
1	112.76	95	7	4	106	444
2	126.98	91	11	2	104	609
3	150.54	101	12	2	115	606
4	79.88	75	12		87	496
5	70.24	71	4		75	255
6	82.68	66	6	2	74	296
7	80.66	74	2		76	366
8	78.33	64	4		68	312
9	64.34	49	4		53	198
10	61.35	57	3		60	271
11	67.60	39	2	3	44	181
12	48.97	49	4	1	54	252
13	40.98	31	3		34	79
14	39.80	30	2		32	80
15	24.50	33	2		35	192
16	76.57	42	2	2	46	246

17	35.90	27			27	207
18	10.72	24	1		25	106
19	21.35	30	1		31	78
20	20.09	32			32	70
21	8.54	17			17	31
22	13.18	23			23	114
23	5.68	11	1		12	29
24	2.39	9			9	17
25	0.63	7			7	6
26	9.91	6	2		8	18
27	9.52	6	1	1	8	66
28	6.48	7			7	8
29	3.45	7			7	30
30	2.67	7	1		8	50
31	5.07	10			10	8
32	0.72	7			7	7
33	1.91	6			6	27
34	1.71	5			5	10
35	1.23	3			3	12
36	0.91	2			2	12
37	0.59	5			5	10
38	4.76	1		1	2	27
39	0.01	1			1	NA
40	0.76	2			2	4
41	3.33	2			2	7
42	4.96	4			4	0

43	0.01	1			1	0
44	3.64	2			2	1
45	1.87	4			4	21
46	0.33	1			1	5
47	1.68	3			3	12
Total	1,390	1239	87	18	1344	5876

Table S5. Recombination rates for shared loci intervals in maternal and paternal maps. LOD (logarithm of odds)

Interval_name	Recombination rate	LOD	N	Family_sex
8777195_8793682	0	8.729	29	F131_M127_F
	0.047	4.575	21	F133_M132_M
8777838_16715831	0.033	7.126	30	F138_M133_F
	0.06	6.657	33	F144_M144_M
8778429_16717674	0	6.622	22	F145_M148_F
	0.029	8.275	34	F144_M144_M
8782658_16719272	0	2.709	9	F140_M149_F
	0	7.525	25	F140_M155_F

	0	5.418	18	F136_M145_M
8786079_8821975	0.1	4.795	30	F131_M127_F
	0	6.622	22	F145_M148_M
8788246_8819402	0.047	4.575	21	F133_M132_F
	0.333	0.811	33	F144_M144_M
8789815_8807862	0.031	7.7	32	F144_M144_F
	0.043	5.137	23	F136_M134_M
8790307_16719120	0	8.428	28	F131_M127_F
	0.03	7.987	33	F144_M144_M
8790488_8813350	0.5	0	18	F133_M132_F
	0.033	7.126	30	F144_M144_M
8791588_8818502	0	9.03	30	F138_M133_F

	0.047	4.575	21	F133_M132_M
8791778_16714510	0	7.525	25	F140_M155_F
	0.05	4.296	20	F136_M145_M
	0	6.622	22	F145_M148_M
8810238_16717194	0.095	3.453	21	F145_M148_F
	0	8.428	28	F138_M133_M
	0	6.321	21	F140_M155_M
8813074_16714430	0	6.622	22	F133_M132_F
	0.047	4.575	21	F136_M134_M
8820902_16718223	0.111	4.037	27	F138_M133_F
	0	6.622	22	F145_M148_M

Table S6. Significant G-values for segregation distortion test performed using LINKMFEX

Marker	G_value	N	Family	Sex	P_value
8797869	4.614	27	F131_M127	F	0.03171198

8777491	4.438	19	F133_M132	F	0.035147399
8779158	3.984	21	F133_M132	F	0.045934359
8780341	5.883	18	F133_M132	F	0.015287752
8782521	3.984	21	F133_M132	F	0.045934359
8782571	4.716	22	F133_M132	F	0.02988318
8788301	5.883	18	F133_M132	F	0.015287752
8813160	4.438	19	F133_M132	F	0.035147399
8814776	3.984	21	F133_M132	F	0.045934359
8819339	5.883	18	F133_M132	F	0.015287752
16716266	5.883	18	F133_M132	F	0.015287752
8802252	6.526	27	F138_M133	F	0.010630876
8819069	4.614	27	F138_M133	F	0.031711198
8781769	3.962	7	F140_M149	F	0.046538379
16716535	3.984	21	F140_M155	F	0.045934359
8790887	3.962	7	F140_M149	M	0.046538379
8801915	3.962	7	F140_M149	M	0.046538379
8779743	3.984	21	F145_M148	M	0.045934359

8798154	4.716	22	F145_M148	M	0.02988318
8801588	4.716	22	F145_M148	M	0.02988318
8819731	5.232	20	F145_M148	M	0.02217502
16715113	4.716	22	F145_M148	M	0.02988318
16716336	4.716	22	F145_M148	M	0.02988318
16716880	3.984	21	F145_M148	M	0.045934359

Table S7. Summary of HOX gene on *H. maculosa* genomic scaffolds. LG (Linkage Group)

Scaffold	Gene id	Hox Gene	Location	Scaffold/LG length
ScBNHFi_16706	g2744.t1	HOX1	Scaffold	360kb
ScBNHFi_31566	g9405.t1	LOX5	Scaffold	3.16Mb
ScBNHFi_22735	g5544.t1	ANTP/LOX2	Scaffold	8.53Mb
ScBNHFi_13142	g1245.t1	LOX4	LG_9	64Mb
ScBNHFi_16629	g2698.t1	POST1	LG_9	64Mb
ScBNHFi_31566	g9400.t1	SCR	LG_9	64Mb

Table S8. The top 30 Pfam domains for genes mapped within the 47 pseudo-chromosomes generated using chromonomer.

Description	Pfam ID	Number of genes
Cadherin domain	PF00028	51

WD domain, G-beta repeat	PF00400	42
Protein kinase domain	PF00069	37
Ankyrin repeats (3 copies)	PF12796	32
von Willebrand factor type A domain	PF00092	32
Zinc finger, C2H2 type	PF00096	32
Cadherin-like	PF08266	31
RNA recognition motif. (a.k.a. RRM, RBD, or RNP domain)	PF00076	27
7 transmembrane receptor (rhodopsin family)	PF00001	24
Neurotransmitter-gated ion-channel ligand binding domain	PF02931	22
Helicase conserved C-terminal domain	PF00271	20
Neurotransmitter-gated ion-channel transmembrane region	PF02932	18
Ras family	PF00071	17
Major Facilitator Superfamily	PF07690	16
SAM domain (Sterile alpha motif)	PF00536	16
DEAD/DEAH box helicase	PF00270	15
Protein tyrosine kinase	PF07714	15
SAM domain (Sterile alpha motif)	PF07647	15
Homeobox domain	PF00046	14

Laminin G domain	PF02210	13
PH domain	PF00169	13
ABC transporter	PF00005	12
Leucine rich repeat	PF13855	12
BTB/POZ domain	PF00651	11
EF-hand domain pair	PF13499	11
PDZ domain (Also known as DHR or GLGF)	PF00595	11
Chitin binding Peritrophin-A domain	PF01607	10
Protein-tyrosine phosphatase	PF00102	10
Trypsin	PF00089	10

Table S9. Genome coverage estimation of sex average, maternal and paternal linkage maps. Genome length estimation method 1(Ge1) and 2 (Ge2).

Method	Map	Ge result	Average between methods Ge1+Ge2/2	% coverage
Ge1 = LG length * (marker number + 1/ marker number - 1)	Sex average	2091.75	2091.75	96.41
Ge2 = LG length +(2*ave_interval)	Sex average	2091.75		
Ge1 = LG length * (marker number + 1/ marker number - 1)	Male	1914.07	1914.06	97.22
Ge2 = LG length +(2*ave_interval)	Male	1914.06		
Ge1 = LG length * (marker number + 1/ marker number - 1)	Female	1949.44	1949.44	93.98
Ge2 = LG length +(2*ave_interval)	Female	1949.44		