

Supplemental Table 1: Transcriptome metrics for C2 and whole-brain samples.

	Average number of reads (2x100)	Total number of Trinity genes predicted
<i>Tritonia</i> C2	13,524,704	10,151
<i>Hermisenda</i> C2	15,644,670	9,867
<i>Pleurobranchaea</i> C2-1 (Swimmer)	22,684,348	6,917
<i>Pleurobranchaea</i> C2-2 (Non-Swimmer)	14,043,212	4,789
<i>Pleurobranchaea</i> Whole Brain	106,288,872	147,024

Supplemental Table 2: Primers used for initial amplification and plasmid cloning.

Species	Gene	FW Primer	Rv Primer
Tritonia	5-HT1a _{Trit}	CCAGACTACATTCAACCTAAACCAGAGC	GTGGTCAGTTTATGCCTCTGGCTTG
Tritonia	5-HT1b _{Trit}	GGTAATGGTAGTTTAATCAGCGTG	GTTCCGGTAAACAGTTAAAGCTCTAG
Tritonia	5-HT2a _{Trit}	CAGGGTCAAGGAACCTCTCGGCAATC	GATTCGGAGATCCACAAGCAGTAAGTCG
Tritonia	5-HT2b _{Trit}	CCCATTGCTGAGAGGTCTGTCTG	CAGATGGCACCCTTAATCGCTATTGATGG
Tritonia	5-HT4 _{Trit}	CAATAGCGACATTGTGATCCTTGCGCC	GCGGAGGTGGAGGAACACGAAAAG
Tritonia	5-HT6 _{Trit}	GCCTTTCTCCAGACGCCTGCTG	GGGTGGCTTCTGCAGATTTAAGAATTATC
Tritonia	5-HT7 _{Trit}	GACTATCACTCTCACCGATTCTAACACGG	CCCTAGATAACCCAGCGCGAATATTTTAC
Hermisenda	5-HT1a _{Herm}	GTGCCCACGATTCAAGTAATTGG	ATAGAATAGCATAGTGCAATCTTAGTCG
Hermisenda	5-HT1b _{Herm}	CCATGATGAGTAACGTCACGCTACCG	CAGGAGAGTCGTTTCGATGATGCGTC
Hermisenda	5-HT2a _{Herm}	GCTTTAACGGAACGACGGTGATTCTG	CAGGGGAGGAATAAGTTTTACGGAGGAG
Hermisenda	5-HT2b _{Herm}	CACGCACCACTACAACAACATCATCCTC	GGTCATCGGGAAAATCCTGTTCTGTGTG
Hermisenda	5-HT4 _{Herm}	CCAACACAGCGGCCCCACC	CCATGGCAAGTCGCTCTATGGTTAGG
Hermisenda	5-HT6 _{Herm}	GCAGATTCCCAGTAGAGTGGTCAACG	CATAGCATCACCGCTGTCCCTAACC
Hermisenda	5-HT7 _{Herm}	CCCCTTCCAATCAATCGGTTG	CCCGACACAACACGGAGTCT
Pleurobranchaea	5-HT2a _{Pleu}	GACACCTTCCACTCCAGCAACAT	TACGGCTCCATCTTCGCTTTCTTCAT
Pleurobranchaea	5-HT7 _{Pleu}	GAGACCAGGTCACCATGACCAGC	CACACTGCCGGTACACACGGG
Tritonia	SCP _{Trit}	GTTGTGCGCAGGTCAGGCTTGATAC	GTTGTCTACTCAATGTGTGTTCTGACG
Hermisenda	SCP _{Herm}	TCCAGCCCGAAGTCCAACAGTAATG	CCAACCACCATCCCGTCTATTGTAG
Pleurobranchaea	SCP _{Pleu}	CAAGCAGAACCCTCCAAGACACAATG	GTTCCGGCATTTCATCACAGTTGAAGG

Supplemental Table 3: Primers used for absolute qPCR.

Species	Gene	FW Primer	Rv Primer
Tritonia	5-HT1a _{Trit}	GATTTCAATGGATGTCTTGTGCTGC	GCATTCTACTGTCTCTGATTCTCA
Tritonia	5-HT1b _{Trit}	CAGCCAGGTATGGTTTCTCCAC	GAAACGAAGTGCCAGGCGGATT
Tritonia	5-HT2a _{Trit}	ATTACGGCTCTCACCCGAAGAAG	CGATTCCACGAACGACAAGCAG
Tritonia	5-HT2a _{Trit-1}	CAGGGTCAAGGAACTCTCGGCAATC	GATTCGGAGATCCACAAGCAGTAAGTCG
Tritonia	5-HT2a _{Trit-2}	CGACTCAGATTCTTCGTGGGATAG	CACCACCGCTGCTGTTGTTG
Tritonia	5-HT2b _{Trit}	GTTATGCCCTTTGGAATGGTAGTAG	ATGGGACTCGTAGATAGGACAG
Tritonia	5-HT4 _{Trit}	CGTGTTTGAAAATAGCGTGGTCATT	CAGACAGATAAACAGCAACGACTG
Tritonia	5-HT6 _{Trit}	CAGAACCCTCTCCAACCTCTTTAT	GAGCATTTGTGCCATCAGCTTC
Tritonia	5-HT7 _{Trit}	GTTATTGCTCTTATCCTCGGTGTGC	GTTCTGGGCTCGGTTGTCTGT
Hermisenda	5-HT1a _{Herm}	ATGCCGCTAAGTCTCATCTATG	TACTGGGCGGTTTCTAATATCG
Hermisenda	5-HT1b _{Herm}	CCAACGGAAATAGCAATACCAACAT	CGAGTATCGTTCTGGGACTGTTC
Hermisenda	5-HT2a _{Herm-1}	GACTACTTCTGTGATTTCTTCGTGAC	GCTTCTCAAAATAGTCTGGTGTGG
Hermisenda	5-HT2a _{Herm-2}	GAGAAGGCTCCAGAACATCACCAT	TGTATCGCTCCGCCGAAATGAAG
Hermisenda	5-HT2b _{Herm}	CGCTAACCCTCATCATTCTGTGC	CCGAACATGCGTGTGTGTGGG
Hermisenda	5-HT4 _{Herm}	GTCCCAATCCTGACCGTGTT	CATTTTCCATCTTTCTTGCTCTCGCC
Hermisenda	5-HT6 _{Herm}	CAGCAGCACCTTGAGAACCAT	GCGATCATAACGCCTCTACG
Hermisenda	5-HT7 _{Herm}	GCTATGGTGAAGCGTCTCCAATC	TACTTCGTCATCACTCAGCCGTTTC
Pleurobranchaea	5-HT2a _{Pleu}	CGAGCACTTCATCATCTACGGC	TCCACCAGCCGCCGTAAC
Pleurobranchaea	5-HT7 _{Pleu}	GACAAAGGAGTAGAGTTCCAAGTGG	GAAGCCACCAAGACTCTGGGAAT
Tritonia	SCP _{Trit}	GAAATGACAATGCCCCGAGCAAC	CACGGCATCAGTGTCTGTGTG
Hermisenda	SCP _{Herm}	GAGAATGTTGTGGCATCGGACTC	ACCCAGTTCCCTGACCAAG
Pleurobranchaea	SCP _{Pleu}	GTCCAAAGCCGACGCATCG	CGACGAGAAACAGGACGGC

Supplemental Table 4: *Pleurobranchaea* whole brain transcriptome component identification numbers.

Gene	Component ID
5-HT1a _{Pleu}	comp77412_c0_seq1
5-HT1b _{Pleu}	comp71667_c0_seq4
5-HT2a _{Pleu}	comp68325_c1_seq1 and comp70123_c2_seq3
5-HT2b _{Pleu}	comp53876_c0_seq1
5-HT4 _{Pleu}	comp159323_c0_seq1 and comp48526_c1_seq1
5-HT6 _{Pleu}	comp68387_c1_seq1
5-HT7 _{Pleu}	comp73047_c11_seq1

Supplemental Table 5. Statistical comparison of gene expression with swimming

Receptor	Cramer's V	Confidence Interval	
		Low	High
5-HT1a	0.82 Non-swim	0.19	1.00
5-HT1b	0.30 Not correlated	0.00	0.86
5-HT2a	0.78 Swim	0.38	1.00
5-HT2b	0.05 Not correlated	0.00	0.51
5-HT4	1.00 Non-swim	0.35	1.00
5-HT6	1.00 Non-swim	0.41	1.00
5-HT7	0.85 Swim	0.31	1.00
SCP	0.49 Not correlated	0.08	0.89

Supplemental Figure 1: SCP alignment from whole brain and C2 transcriptomes.
Tritonia whole brain (WB) short and long SCP sequences were aligned with the corresponding C2 neuron transcriptome sequence.

WB long SCP	MEMTMPRATVSLTLLFVICTVDAMNYLAFPRMGRSDLTRVKRHVDMFPWNMIFPRKRGGM
WB short SCP	MEMTMPRATVSLTLLFVICTVDAMNYLAFPRMGRS-----
C2 SCP	MEMTMPRATVSLTLLFVICTVDAMNYLAFPRMGRS----- *****
WB long SCP	LPENFIFPRKRGSMLPGNFIFPRKRQNGYLAFPRMGRSQAKAGTAE AIDTECCGIGLKSE
WB short SCP	-----GYLAFPRMGRSQAKAGTAE AIDTECCGIGLKSE
C2 SCP	-----GYLAFPRMGRSQAKAGTAE AIDTECCGIGLKSE *****
WB long SCP	FAVSDDGKEELHNICTASVSVCCGLRELADKPNQVVSVCVDPVSKMPSSYNKLKRL
WB short SCP	FAVSDDGKEELHNICTASVSVCCGLRELADKPNQVVSVCVDPVSKMPSSYNKLKRL
C2 SCP	FAVSDDGKEELHNICTASVSVCCGLRELADKPNQVVSVCVDPVSKMPSSYNKLKRL *****