

Supporting information S1.

Figure A. Histone H3 sequence alignment (untreated and bisulfite treated sequence) to detect methylated cytosines.

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H3      TATGGCTCGTTACCAAGCAGACCGCACGTAATCGACTGGAGGAAAGGCTCCCCGCAAGCAGCTTGCCACCAAGGCTGCTAGGAAGAGTGCTCCAGCCA
H3 bisulfite -----T..TT..TT.

H3      *          *          *          *
CGGGGGAGTCAAGAAGCCCCACAGGTACAGGCCCGGAACGTGTGCACTCCGTGAGATTAGGAGATACCAGAAATCCACAGAGCTCCTTATCCGCAAGCT
H3 bisulfite .....T.....TTTT.T....T..TT.....T....T..T.T.....TT.....TT.T....T.TT....T..T...T.

H3      *          *          *          *
CCCTTCCAGCGTCTTGTAGAGAGATTGCCAGGACTTTAAGACCGATCTTCGCTTCCAGTCCTCTGCCGTCATGGCTCTTCAGGAGGCTAGTGAGGCA
H3 bisulfite TTT..TT....T.....TTT....T.....T....T...T..TT..TT.T..T...T..T..T.....T.....T.

H3      ATCTTGTGGTCTGTTTGAAGACACCAACCTGTGTGCCATCCATGCCAAGCGTGTCACCATCATGCC
H3 bisulfite ..T.....T.....

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A 367 bp fragment of the histone H3 (H3) gene from *Pygospio elegans* aligned with a 230 bp sequence obtained from the same DNA treated with bisulfite conversion. In the H3 bisulfite sequence, dots (.) are used when the sequence is identical to the untreated sequence above, T is for sites where unmethylated cytosines have been converted to thymines, and asterisks (*) are used to point out the eight methylated cytosines which have not been converted during the bisulfite treatment (all at CpG sites).

Figure B. Variation in methylation (measured by CpG o/e ratio) of 8658 ORF containing transcripts from *P. elegans* assigned to discrete biological processes. The number of contigs in each category is shown in parenthesis after the category name. Bars represent mean CpG o/e ratio for ORF containing contigs in each GO Slim category ± 1 standard error of the mean.

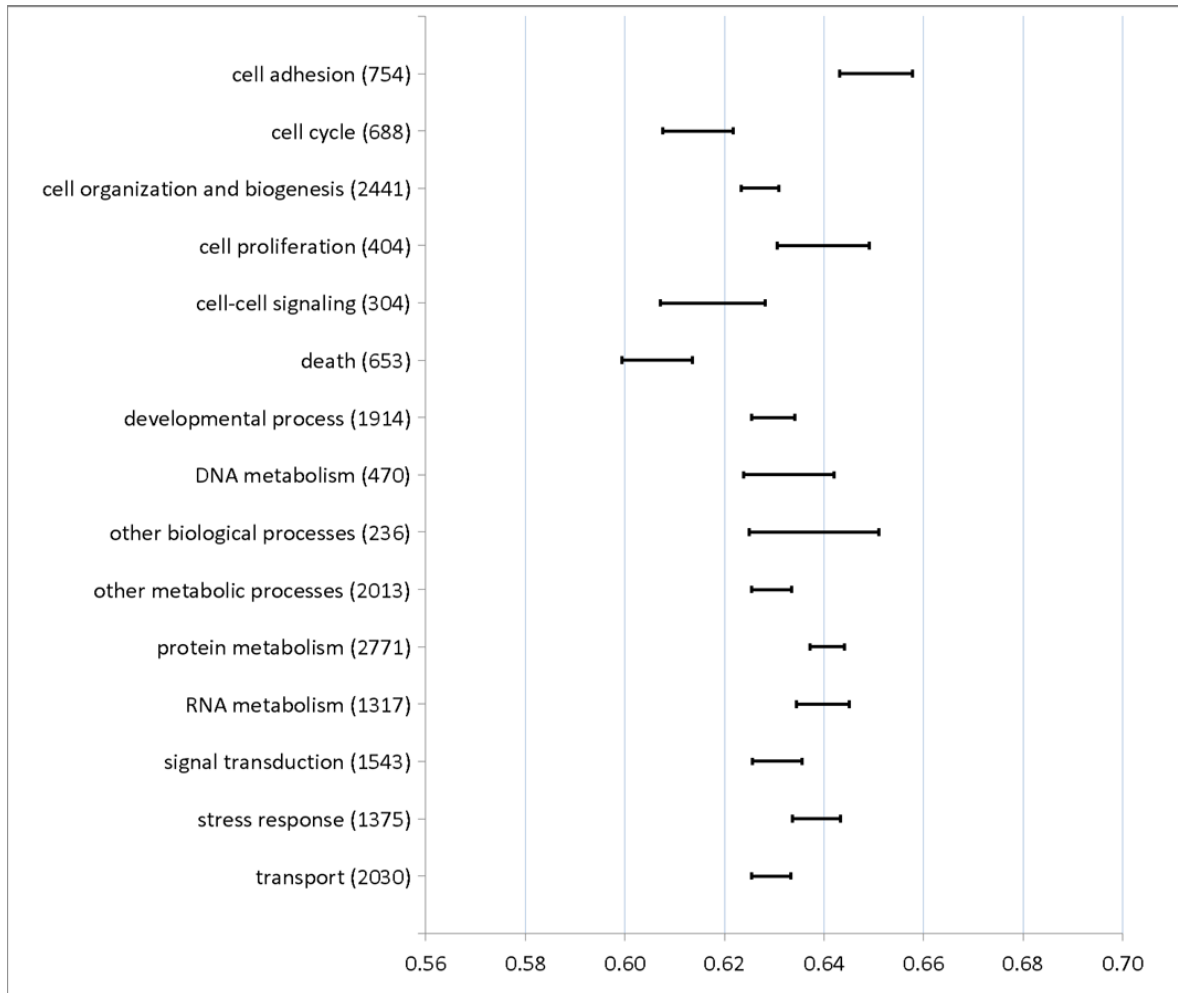


Figure C. Neighbor-joining phylogenetic tree for Dnmt1 sequences (with bootstrap support values of >70% on the nodes).

Species and accession numbers: *Bombyx mori* (NP_001036980.1), *Daphnia pulex* (EFX80183.1), *Nasonia vitripennis* (NP_001164521.1), *Apis mellifera* (XP_006562865.1), *Helobdella robusta* (ESN92163.1), *Aplysia californica* (XP_012936221.1, XP_005095277.1), *Biomphalaria glabrata* (XP_013075419.1), *Crassostrea gigas* (EKC23761.1), *Capitella teleta* (ELT93682.1), *Danio rerio* (NP_571264.2), *Homo sapiens* (AAI26228.1), *Mus musculus* (AAH53047.1)

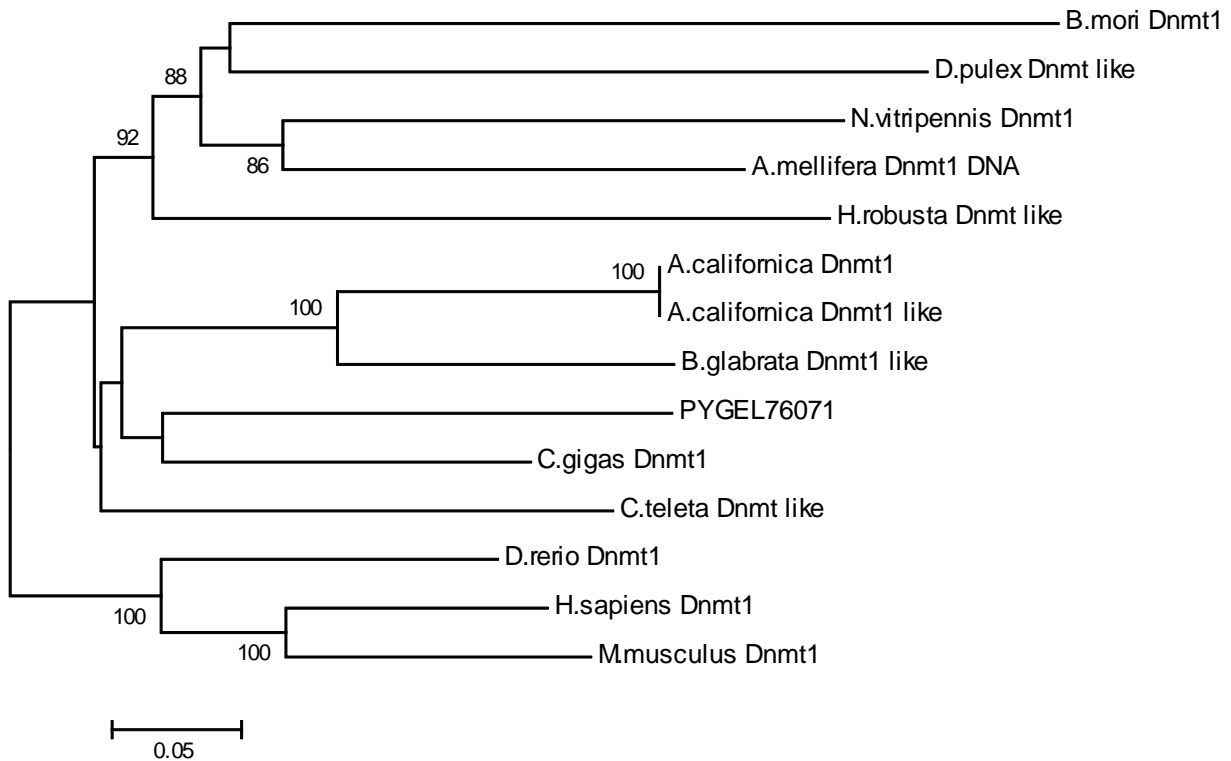


Figure D. Neighbor-joining phylogenetic tree for MBD sequences (MBD2, 3 & 2/3) (with bootstrap support values of >70% on the nodes).

Species and accession numbers: *Aplysia californica* (XP_005103642.1), *Biomphalaria glabrata* (NP_001298225.1), *Crassostrea gigas* (EKC32831.1), *Lottia gigantea* (XP_009048664.1), *Lingula anatina* (XP_013380854.1), *Capitella teleta* (ELU03991.1), *Ixodes scapularis* (XP_002407962.1), *Daphnia pulex* (EFX83334.1), *Apis mellifera* (XP_392422.2), *Drosophila melanogaster* (AF171098_1), *Homo sapiens* (MBD1: NP_001191066.1, MBD2: AAC68871.1, MBD3: AAC68876.1), *Danio rerio* (MBD2: NP_997933.1, MBD3: NP_997934.1), *Mus musculus* (MBD2: NP_034903.2, MBD3: NP_038623.1), *Xenopus laevis* (NP_001083787.1)

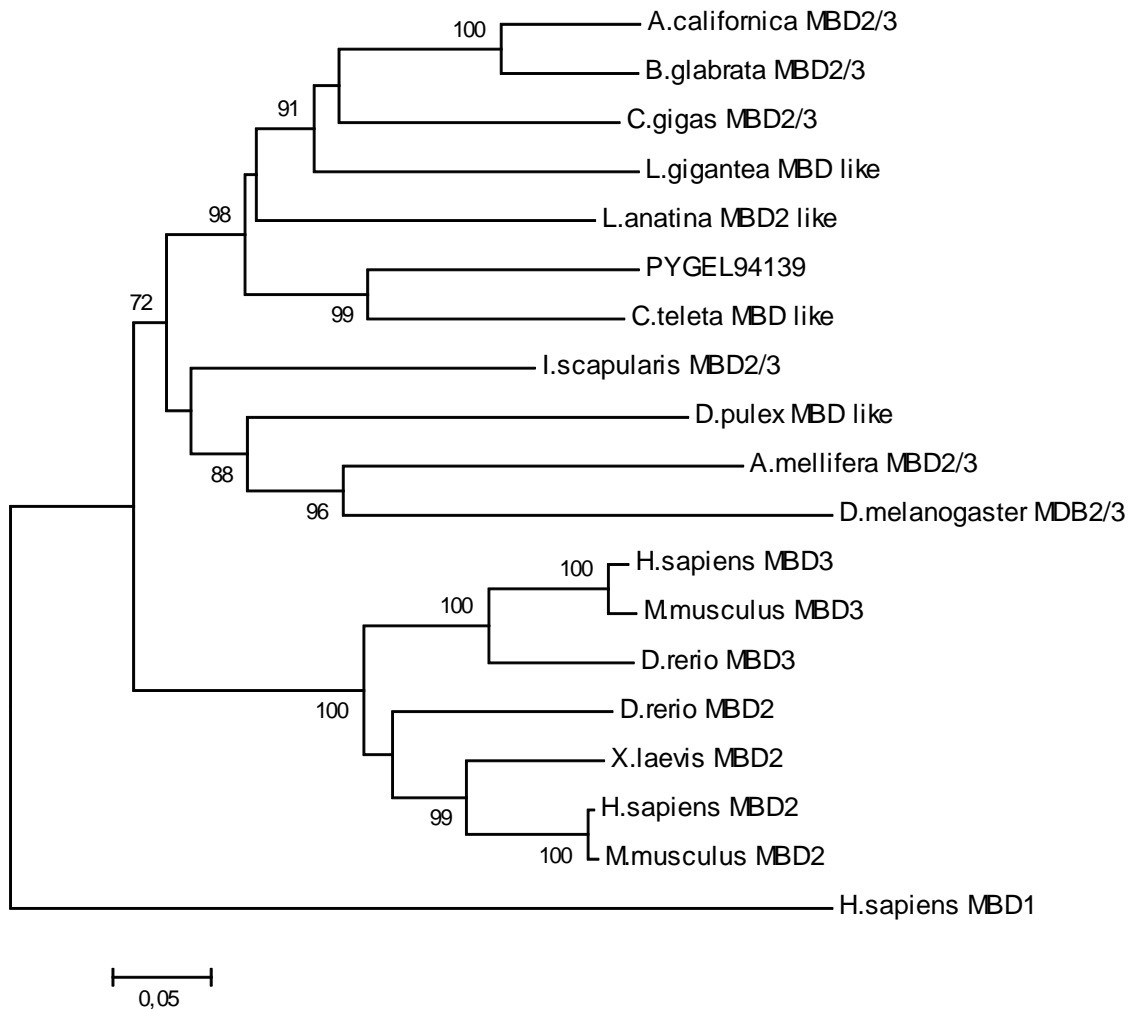


Figure E. Neighbor-joining phylogenetic tree for TET sequences (with bootstrap support values of >70% on the nodes).

Species and accession numbers: *Crassostrea gigas* (XP_011419066.1, EKC18772.1), *Lottia gigantea* (XP_009060167.1), *Biomphalaria glabrata* (XP_013078194.1), *Aplysia californica* (XP_005092662.1), *Capitella teleta* (ELU00383.1), *Drosophila melanogaster* (NP_001246581.1), *Nasonia vitripennis* (XP_008203342.1), *Apis mellifera* (XP_006561262.1), *Melipona quadrifasciata* (KOX75173.1), *Habropoda laboriosa* (KOC60806.1), *Homo sapiens* (TET1: NP_085128.2, TET2: NP_001120680.1, TET3: NP_001274420.1), *Mus musculus* (NP_898961.2)

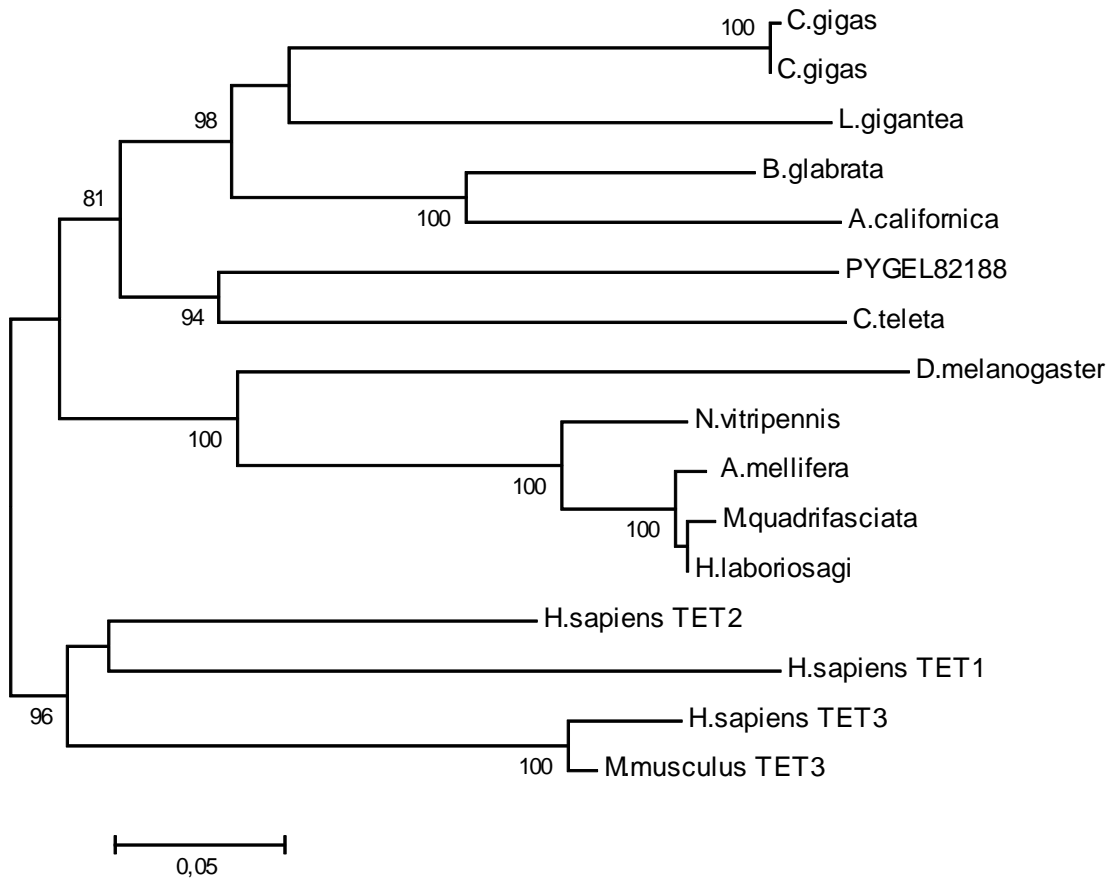


Table B. p-values (significance threshold < 0.05) for comparisons between GO Slim categories for benthic vs. planktonic gravid females. Comparison is based on difference in CpG o/e ratios.

GO category (#ORF benthic planktonic)	p-value
cell adhesion (338 348)	0.04716
cell cycle (261 361)	0.38904
cell organization and biogenesis (1061 1379)	0.04173
cell proliferation (185 224)	0.31999
cell-cell signaling (140 167)	0.78075
death (333 376)	0.09763
developmental process (882 1038)	0.46346
DNA metabolism (138 226)	0.30446
other biological processes (102 117)	0.34460
other metabolic processes (1040 1224)	0.00104
protein metabolism (1144 1663)	0.01190
RNA metabolism (437 671)	0.58868
signal transduction (701 872)	0.11549
stress response (591 743)	0.34359
transport (938 1211)	0.05586

Table C. Primers used in the selective PCR of the methylation-sensitive AFLP protocol

Six different primer combinations used in the selective PCR of the MS-AFLP protocol, number of fragments (loci) produced by each primer pair, number of methylation-sensitive loci (MSL), estimated with the msap program.

EcoRI	HpaII/MspI	Fragments	MSL
5'-GACTGCGTACCAATTCACA-3'	5'-GATGAGTCTAGAACGGTGT-3'	22	13
5'-GACTGCGTACCAATTCACA-3'	5'-GATGAGTCTAGAACGGTAC-3'	33	14
5'-GACTGCGTACCAATTCAGA-3'	5'-GATGAGTCTAGAACGGTAC-3'	26	15
5'-GACTGCGTACCAATTCATC-3'	5'-GATGAGTCTAGAACGGTTA-3'	31	16
5'-GACTGCGTACCAATTCATC-3'	5'-GATGAGTCTAGAACGGTGT-3'	25	19
5'-GACTGCGTACCAATTCATC-3'	5'-GATGAGTCTAGAACGGTAC-3'	28	21