

# Chapter 8

## Appendices

### *Appendix A: Aquarium water, 10X*

(Prepared by the Wellcome Trust Sanger Institute (WTSI) media team)

<b>Reagents</b>	<b>Amount for 5 litre</b>	<b>Final conc.</b>
<b>CaCl<sub>2</sub></b>	2.78 g	5.00 mM
<b>MgSO<sub>4</sub>·7H<sub>2</sub>O</b>	6.14 g	4.98 mM
<b>K<sub>2</sub>SO<sub>4</sub></b>	0.215 g	0.25 mM
<b>NaHCO<sub>3</sub></b>	2.1 g	5.00 mM
<b>*FeCl<sub>3</sub>·6H<sub>2</sub>O</b>	240 ul	0.009 mM

\*The FeCl<sub>3</sub>·6H<sub>2</sub>O is prepared by dissolving 5 g in 100 ml water.

The final pH of the aquarium water is pH 7 +/- 0.5.

## **Appendix B: Reference genomes used by NPG QC**

The NPG QC team at the WTSI constantly adds more species to the pool of reference genomes. The species listed below are those included during the work of this thesis.

<i>Acinetobacter baumannii</i>	<i>Escherichia coli</i>
<i>Actinobacillus pleuropneumoniae</i>	<i>Felis catus</i>
<i>Aeromonas hydrophila</i>	<i>Giardia intestinalis</i>
<i>Anopheles gambiae</i>	<i>Gorilla beringei</i>
<i>Aspergillus fumigatus</i>	<i>Gorilla gorilla</i>
<i>Bacillus thuringiensis</i>	<i>Haemonchus contortus</i>
Betacoronavirus	<i>Haemophilus influenzae</i>
<i>Bordetella bronchiseptica</i>	<i>Haemophilus parasuis</i>
<i>Bordetella pertussis</i>	<i>Heligmosomoides polygyrus</i>
<i>Bos taurus</i>	Hepatitis C
<i>Brucella abortus</i>	<i>Heterocephalus glaber</i>
<i>Brucella canis</i>	HIV 1
<i>Brugia malayi</i>	<i>Homo sapiens</i>
<i>Brugia pahangi</i>	Human herpesvirus 1
<i>Burkholderia cenocepacia</i>	Human herpesvirus 2
<i>Burkholderia gladioli</i>	Human herpesvirus 3
<i>Burkholderia pseudomallei</i>	Human herpesvirus 4
<i>Caenorhabditis elegans</i>	Human herpesvirus 5
<i>Callithrix jacchus</i>	Human herpesvirus 6
<i>Campylobacter fetus</i>	Human herpesvirus 7
<i>Campylobacter jejuni</i>	Human herpesvirus 8
<i>Canis familiaris</i>	<i>Human papillomavirus</i>
<i>Cavia porcellus</i>	<i>Hymenolepis microstoma</i>
<i>Chlamydia trachomatis</i>	Influenza A
<i>Chlamydophila abortus</i>	<i>Klebsiella pneumoniae</i>
<i>Citrobacter rodentium</i>	<i>Lactobacillus casei</i>
<i>Clostridium difficile</i>	Lambda
<i>Cryptococcus neoformans</i>	<i>Legionella pneumophila</i>
<i>Cryptosporidium parvum</i>	<i>Leishmania braziliensis</i>
<i>Danio rerio</i>	<i>Leishmania donovani</i>
<i>Dracunculus medinensis</i>	<i>Leishmania infantum</i>
<i>Drosophila melanogaster</i>	<i>Leishmania major</i>
<i>Echinococcus granulosus</i>	<i>Leptospira interrogans</i>
<i>Echinococcus multilocularis</i>	<i>Macaca fascicularis</i>
<i>Enterococcus casseliflavus</i>	<i>Melissococcus plutonius</i>
<i>Enterococcus faecalis</i>	MERS coronavirus
<i>Enterococcus faecium</i>	<i>Monodelphis domestica</i>
<i>Enterococcus hirae</i>	<i>Mus musculus</i>
<i>Equus caballus</i>	<i>Mustela putorius</i>

*Mycobacterium abscessus*  
*Mycobacterium africanum*  
*Mycobacterium avium*  
*Mycobacterium bovis*  
*Mycobacterium tuberculosis*  
*Mycobacterium ulcerans*  
*Mycoplasma agalactiae*  
*Mycoplasma bovis*  
*Mycoplasma genitalium*  
*Mycoplasma hyopneumoniae*  
*Myotis lucifugus*  
*Neisseria gonorrhoeae*  
*Neisseria meningitidis*  
 Norwalk virus  
*Onchocerca volvulus*  
*Oryctolagus cuniculus*  
*Oryzias latipes*  
*Ovis aries*  
*Paenibacillus larvae*  
*Pan troglodytes*  
*Plasmodium berghei*  
*Plasmodium chabaudi*  
*Plasmodium falciparum*  
*Plasmodium knowlesi*  
*Plasmodium vivax*  
*Propionibacterium acnes*  
*Proteus mirabilis*  
*Pseudomonas aeruginosa*  
*Pseudomonas fluorescens*  
*Rattus norvegicus*  
*Rhabditophanes sp*  
*Rhesus macaque*  
*Saccharomyces cerevisiae*  
*Saliva composite*  
*Salmonella bongori*  
*Salmonella enterica*  
*Salmonella pullorum*  
*Sarcophilus harrisii*  
*Schistocephalus solidus*  
*Schistosoma mansoni*  
*Schizosaccharomyces pombe*  
*Serratia proteamaculans*  
*Shigella boydii*  
*Shigella flexneri*  
*Shigella sonnei*  
*Staphylococcus aureus*  
*Staphylococcus haemolyticus*  
*Staphylococcus saprophyticus*  
*Streptococcus agalactiae*  
*Streptococcus dysgalactiae*  
*Streptococcus equi*  
*Streptococcus pneumoniae*  
*Streptococcus pyogenes*  
*Streptococcus suis*  
*Streptococcus uberis*  
*Streptomyces coelicolor*  
*Streptomyces venezuelae*  
*Strongyloides ratti*  
*Sus scrofa*  
*Teladorsagia circumcincta*  
*Trichobilharzia regenti*  
*Trichobilharzia szidati*  
*Trichuris muris*  
*Trypanosoma brucei*  
*Tupaia belangeri*  
*Tursiops truncatus*  
*Vibrio cholerae*  
*Wolbachia endosymbiont of Drosophila melanogaster*  
*Xenopus tropicalis*  
*Yersinia enterocolitica*  
*Yersinia pseudotuberculosis*

## *Appendix C: Versions of R packages used in data analysis*

R version 3.3.1 (2016-06-21)  
Platform: x86\_64-apple-darwin13.4.0 (64-bit)  
Running under: OS X 10.11.1 (El Capitan)

locale:  
en\_GB.UTF-8/en\_GB.UTF-8/en\_GB.UTF-8/C/en\_GB.UTF-8/en\_GB.UTF-8

attached base packages:  
grid parallel stats4 stats graphics grDevices utils datasets methods base

other attached packages:

ape_3.5	VennDiagram_1.6.17	futile.logger_1.4.3
RColorBrewer_1.1-2	gridExtra_2.2.1	dplyr_0.5.0
vioplot_0.2	sm_2.2-5.4	kohonen_2.0.19
MASS_7.3-45	class_7.3-14	heatmap_1.0.8
topGO_2.24.0	SparseM_1.7	GO.db_3.3.0
AnnotationDbi_1.34.4	graph_1.50.0	ggplot2_2.2.1
DESeq2_1.12.3	SummarizedExperiment_1.2.3	Biobase_2.32.0
GenomicRanges_1.24.2	GenomeInfoDb_1.8.1	IRanges_2.6.1
S4Vectors_0.10.2	BiocGenerics_0.18.0	

loaded via a namespace (and not attached):

genefilter_1.54.2	locfit_1.5-9.1	splines_3.3.1
survival_2.39-5	XML_3.98-1.4	foreign_0.8-66
lambda.r_1.1.9	matrixStats_0.50.2	plyr_1.8.4
gtable_0.2.0	latticeExtra_0.6-28	geneplotter_1.50.0
xtable_1.8-2	scales_0.4.1	Hmisc_3.17-4
tools_3.3.1	magrittr_1.5	lazyeval_0.2.0
futile.options_1.0.0	Formula_1.2-1	cluster_2.0.4
assertthat_0.1	R6_2.1.2	rpart_4.1-10
colorspace_1.2-6	munsell_0.4.3	XVector_0.12.0
BiocParallel_1.6.2	acepack_1.3-3.3	RSQLite_1.0.0
lattice_0.20-33	Rcpp_0.12.9.1	Matrix_1.2-6
DBI_0.4-1	annotate_1.50.0	nlme_3.1-128
zlibbioc_1.18.0	tibble_1.2	data.table_1.10.0
nnet_7.3-12		

***Appendix D: Basch media components***

0.5 gr	Lactalbumin hydrolysate powder (L9010, Sigma)
250ul	Hypoxanthine (1 mM) (H9377, Sigma)
500ul	Insulin (8mg/ml) (I0516, Sigma)
500ul	Hydrocortisone (1 mM) (H0888, Sigma)
500ul	Triiodothyronine (0.2 mM) (T5516, Sigma)
2.5 ml	MEM Vitamins (100X) (M6895, Sigma)
25 ml	Schneiders Drosophila Medium (21720024, Invitrogen)
5 ml	Hepes Buffer (sc-286961, Santa Cruz Biotechnology, Inc.)
50 ml	Fetal bovine serum (F0926, Sigma)
10 ml	Antibiotic-Antimycotic (15240-062, Invitrogen)
to 500 ml	DMEM (D6546, Sigma)

Filter through 0.22  $\mu$ m membrane and store at 4 °C.

**Appendix E: *S. mansoni* genetrees downloaded from WormBase ParaSite release 9**

The genetrees are grouped by chapters and ordered by *S. mansoni* gene identifier.

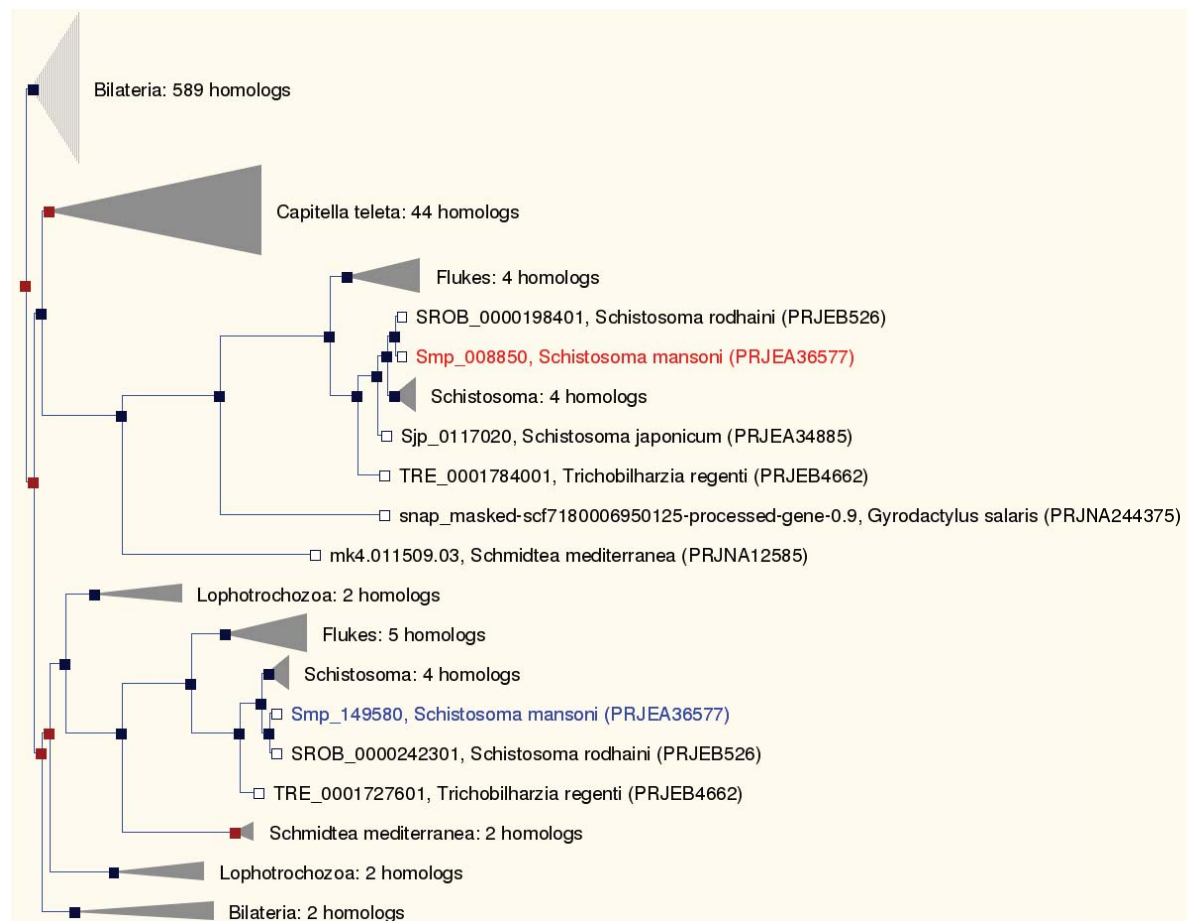
Legend for all genetrees

Branch Length	Genes	Nodes
— x1 branch length	Gene ID gene of interest	□ gene node
⋯ x10 branch length	Gene ID within-sp. paralog	■ speciation node
⋯ x100 branch length		■ duplication node
		■ ambiguous node
		■ gene split event
Collapsed Nodes		
◀	collapsed sub-tree	
▶	collapsed (paralog)	
▶	collapsed (gene of interest)	
Collapsed Alignments		
□	0 - 33% Aligned AA	□ Gap
■	33 - 66% Aligned AA	■ Aligned AA
■	66 - 100% Aligned AA	
Expanded Alignments		
		□ Gap
		■ Aligned AA

Smp\_008850 (chapter 3)

A fully expanded tree is available at

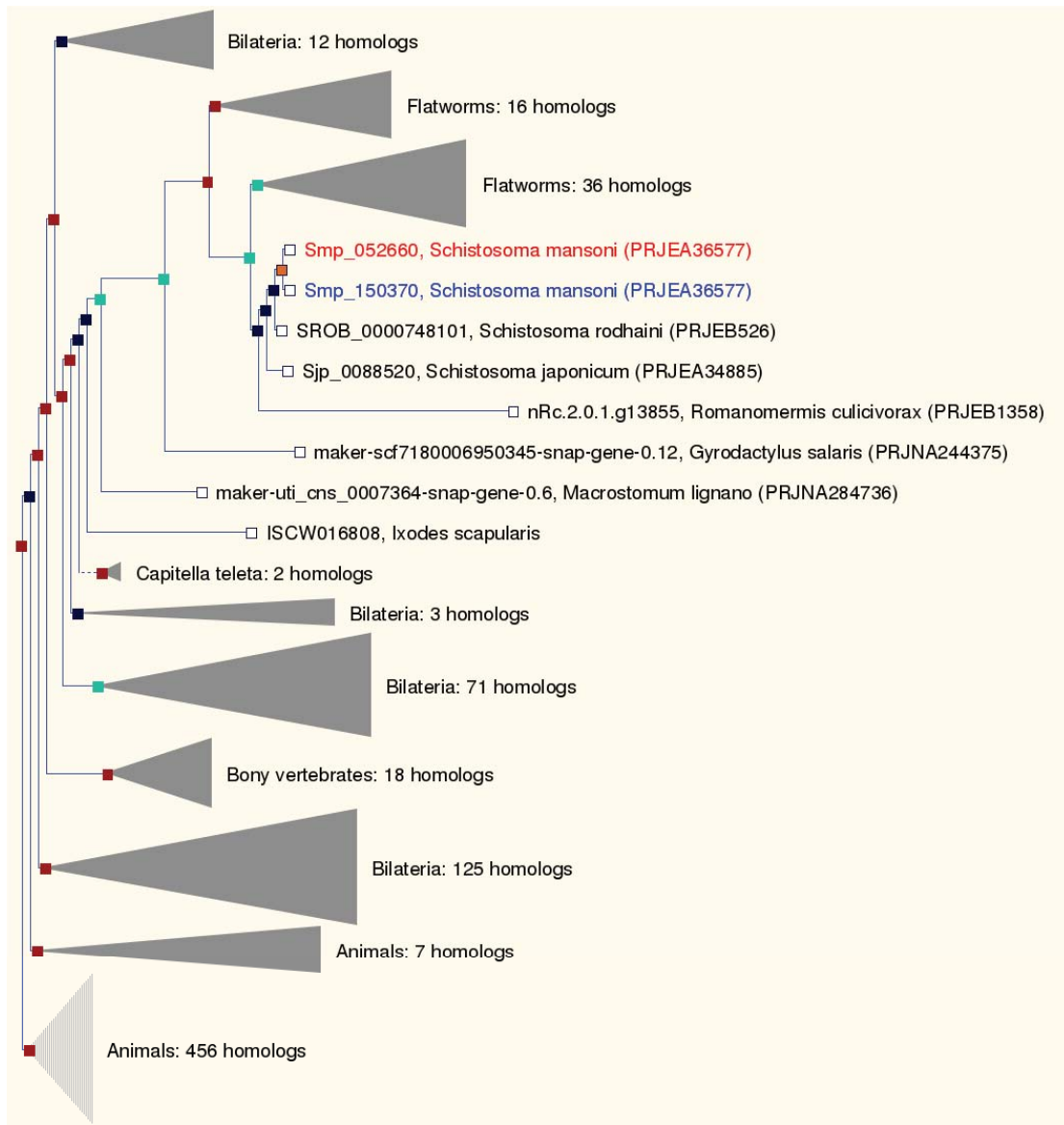
<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBGT00840000210809>



Smp\_052660 (chapter 3)

A fully expanded tree is available at

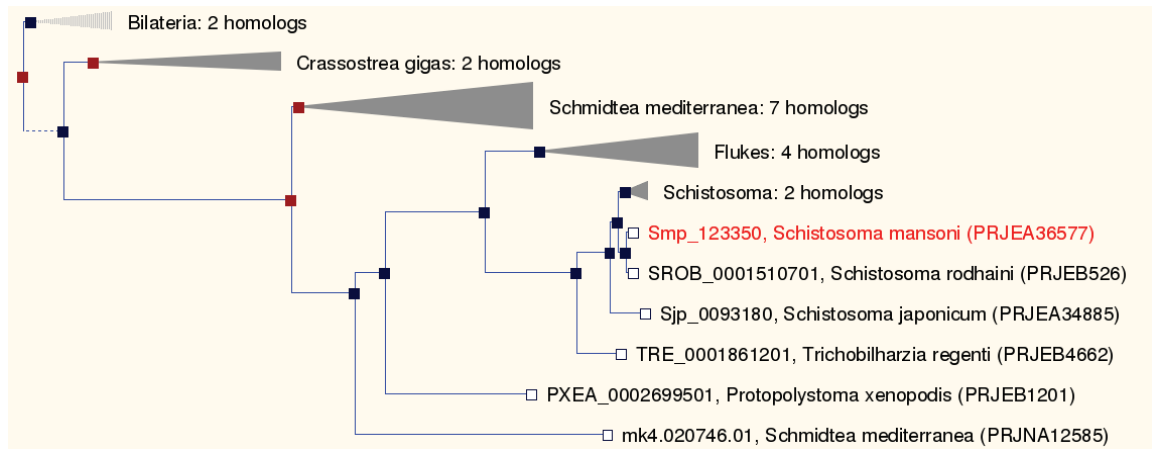
<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBG00800000173209>



Smp\_123350 (chapter 3)

A fully expanded tree is available at

<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBG00000000014882>

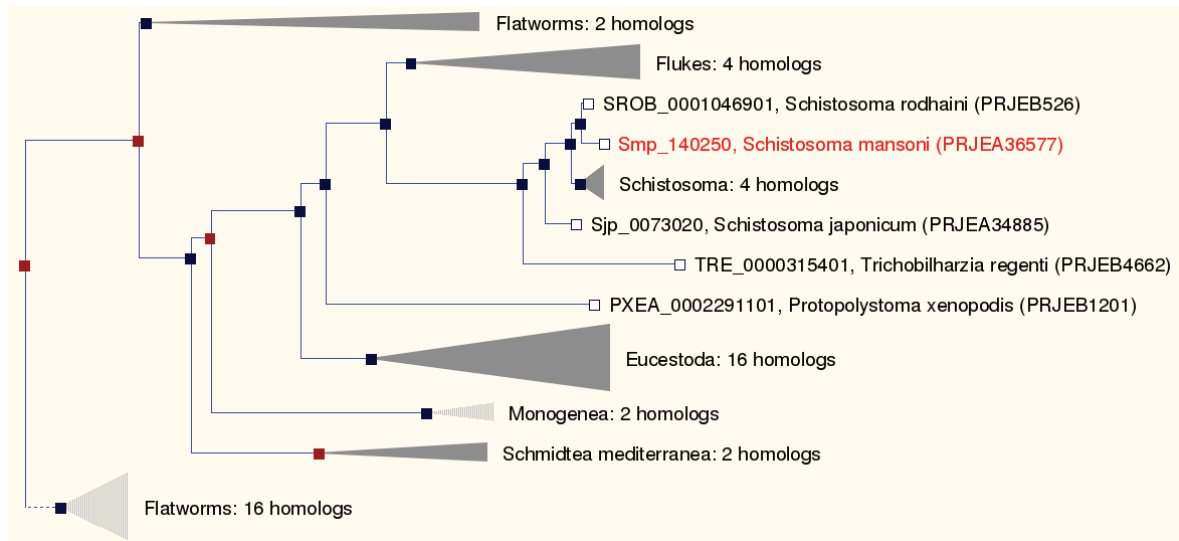




Smp\_140250 (chapter 3)

A fully expanded tree is available at

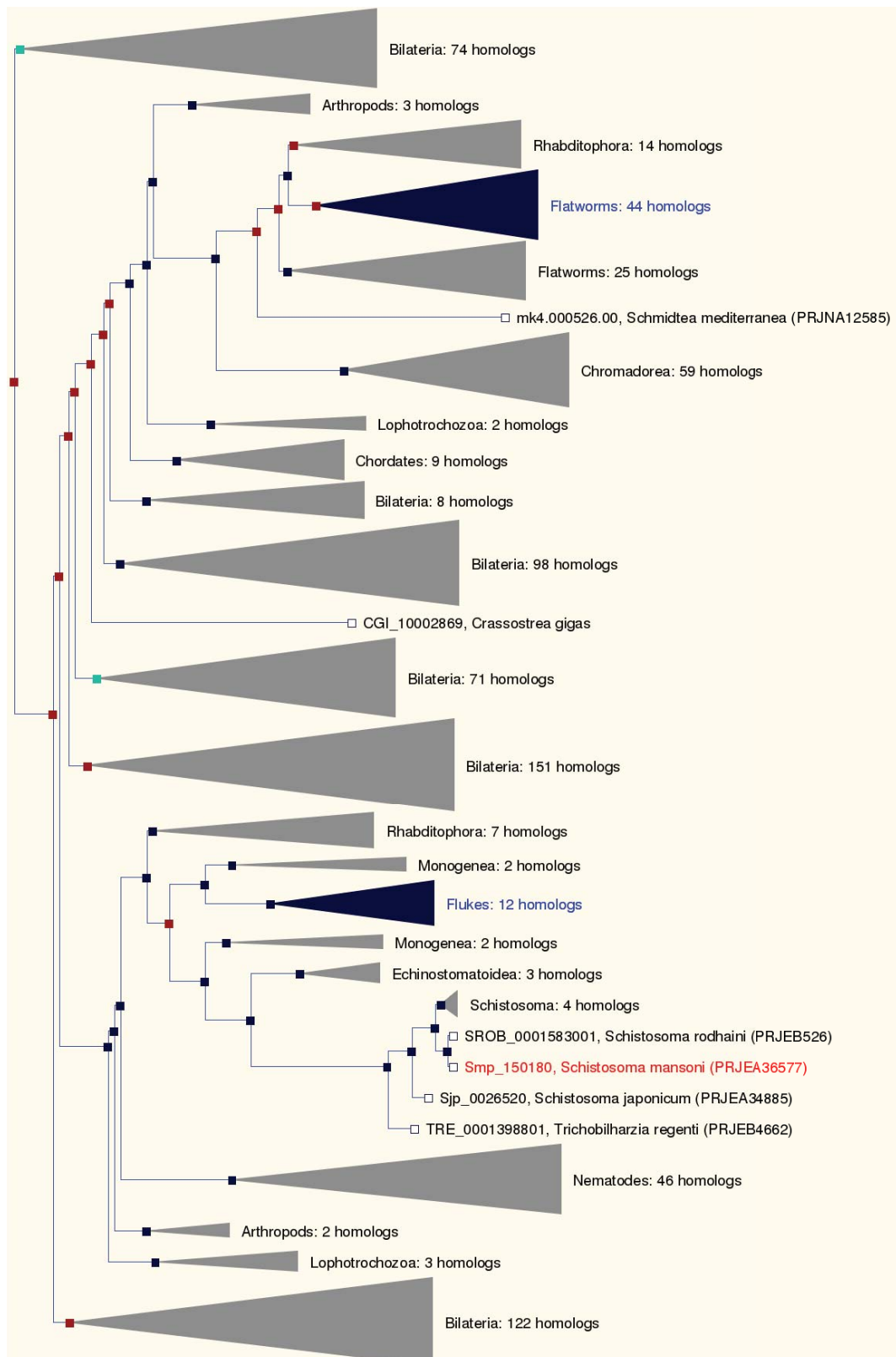
<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBG00000000006409>



Smp\_150180 (chapter 3)

A fully expanded tree is available at

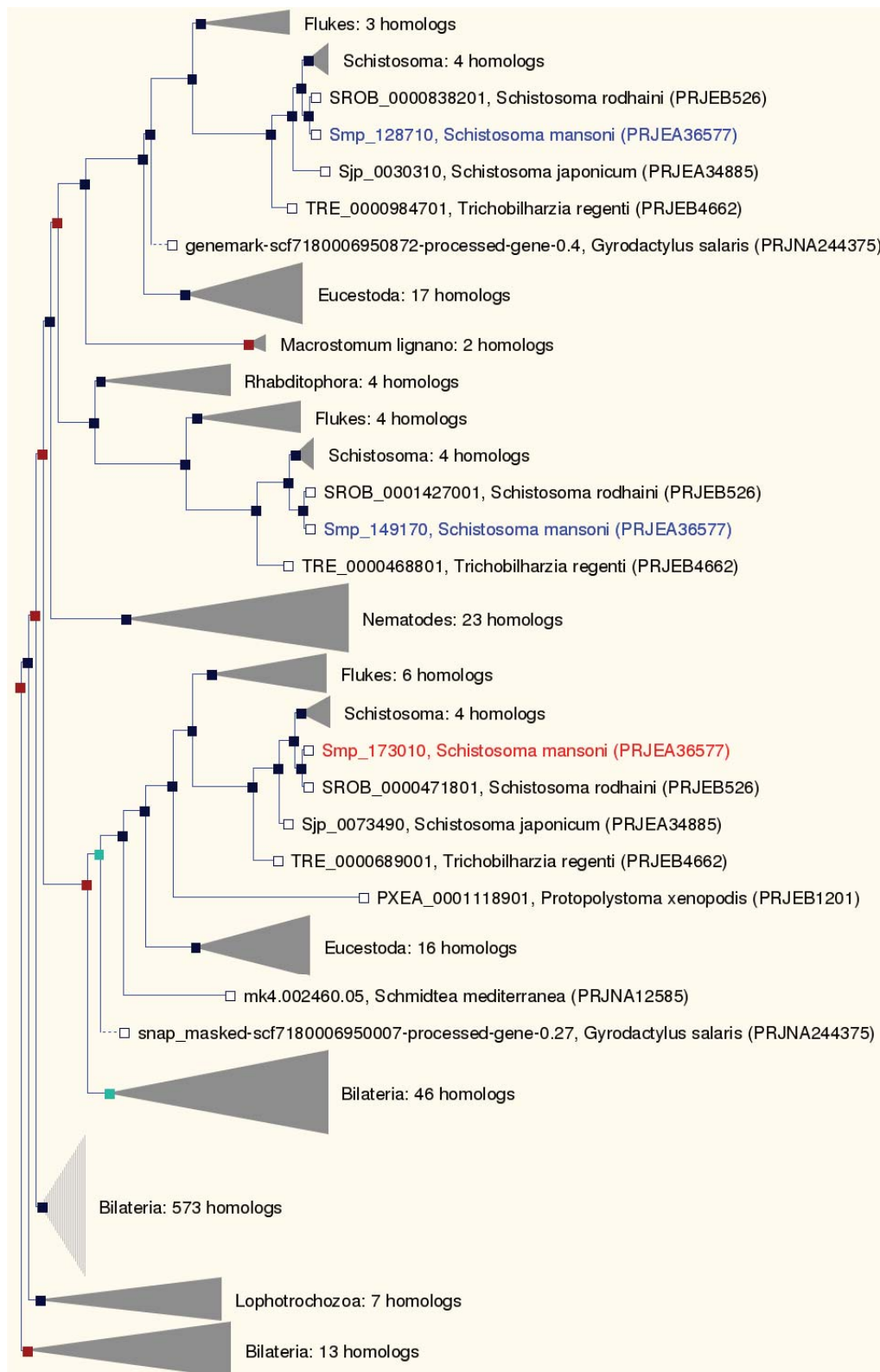
<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBG00800000173094>



Smp\_173010 (chapter 3)

A fully expanded tree is available at

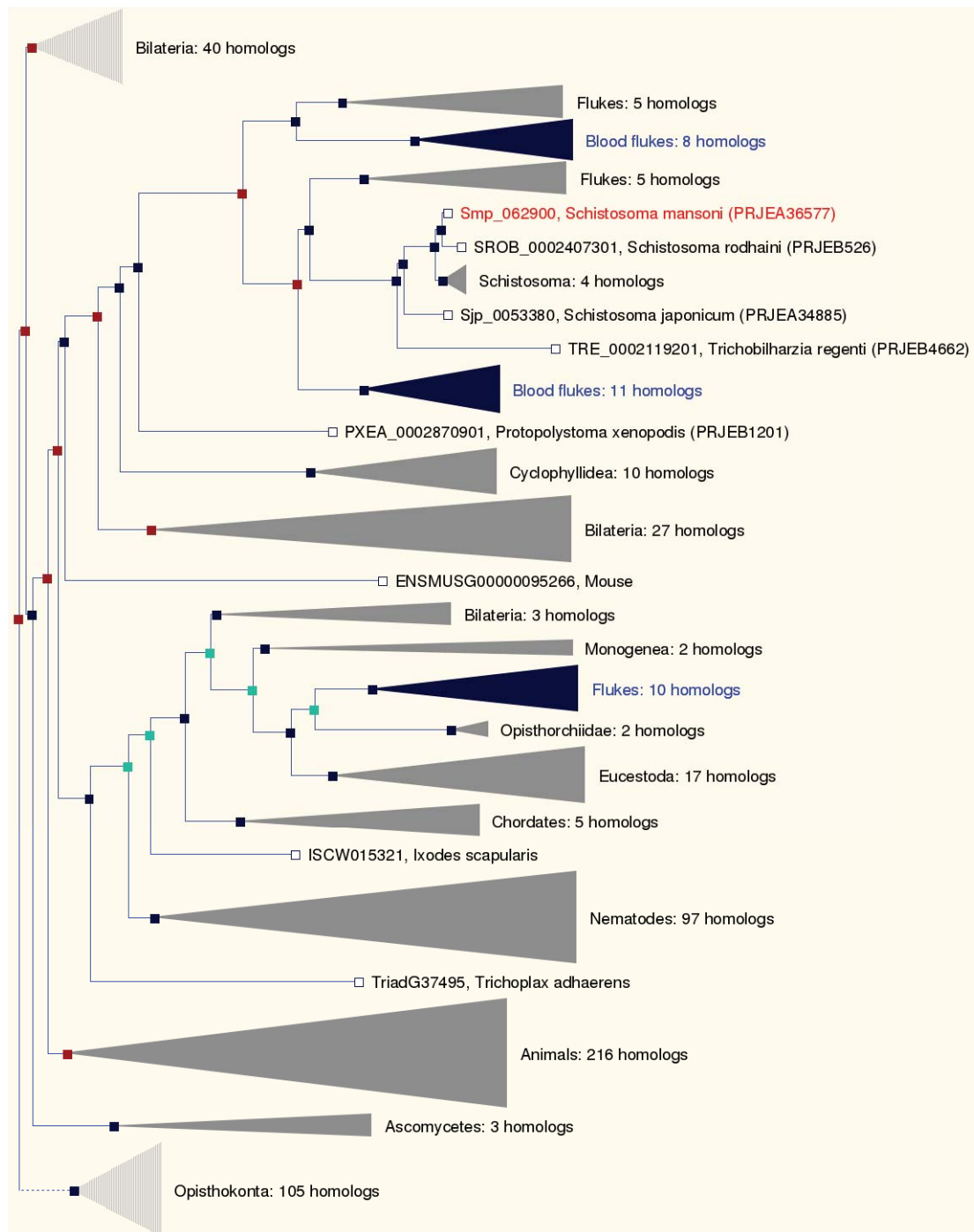
<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBG00800000173235>



Smp\_062900 (chapter 4)

A fully expanded tree is available at

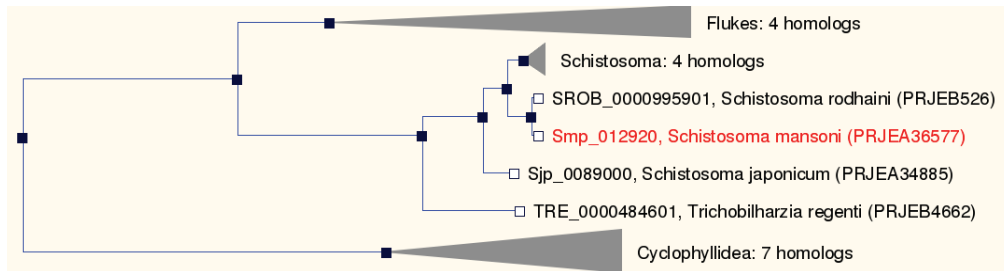
<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBG00800000173326>



Smp\_012920 (chapter 4)

A fully expanded tree is available at

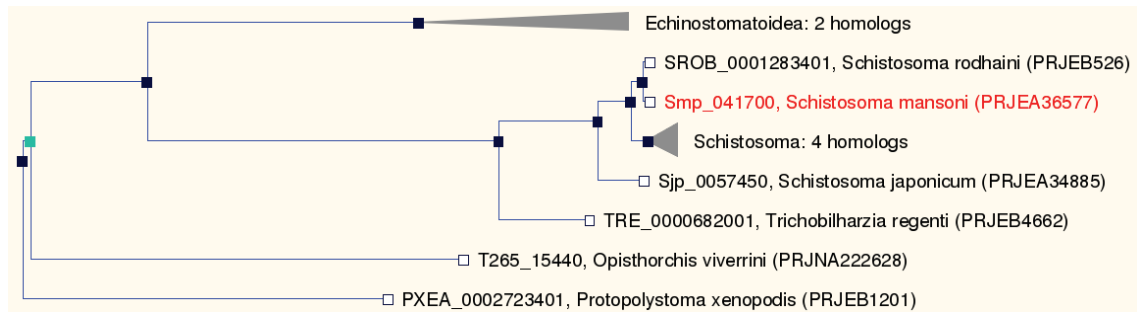
<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBGT00000000012253>



Smp\_041700 (chapter 4)

A fully expanded tree is available at

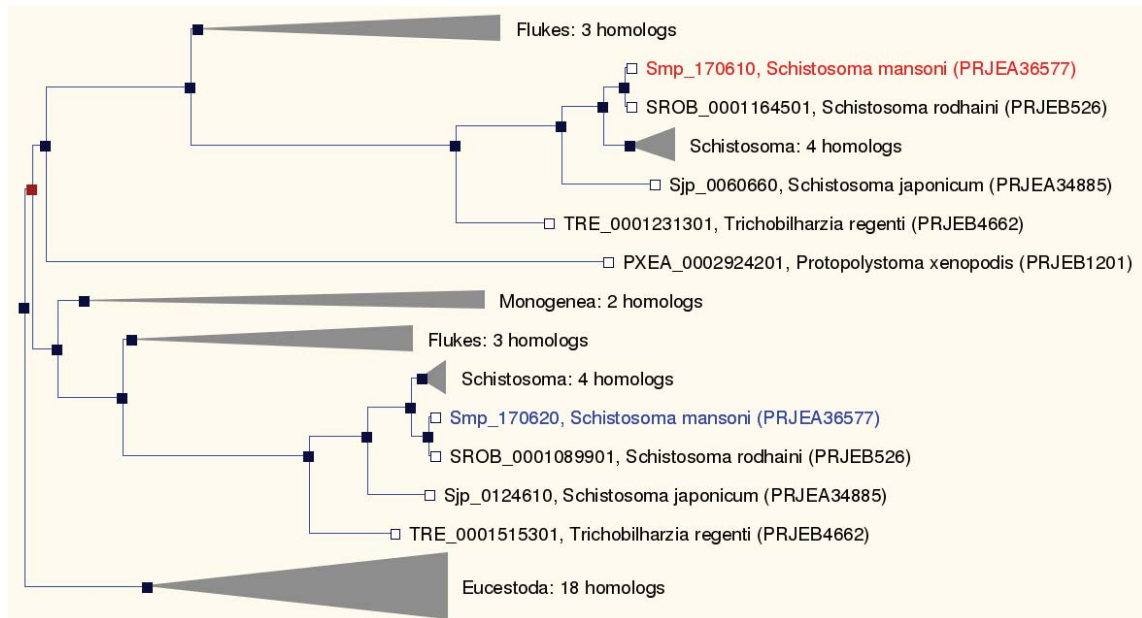
<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBG00880000282628>



Smp\_170610 (chapter 4)

A fully expanded tree is available at

<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBG00000000007301>



Smp\_201210 (chapter 4)

The tree is available at

<http://parasite.wormbase.org/Multi/GeneTree/Image?gt=WBGT00860000263648>

▣ SROB\_0002124301, *Schistosoma rodhaini* (PRJEB526)

▣ Smp\_201210, *Schistosoma mansoni* (PRJEA36577)



*Appendix F: Orthologues of Smp\_041700 downloaded from WormBase ParaSite release 8*

<b>Gene stable identifier of orthologues</b>	<b>Gene description</b>	<b>Genome project</b>
A_02595	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	schistosoma_haematobium_prjna78265
CapteG188111		capitella_teleta
CapteG190027		capitella_teleta
CapteG192465		capitella_teleta
CapteG197443		capitella_teleta
CapteG200779		capitella_teleta
CapteG206916		capitella_teleta
CapteG207119		capitella_teleta
CapteG208374		capitella_teleta
CapteG211298		capitella_teleta
CGI_10000319	Growth hormone secretagogue receptor type 1	crassostrea_gigas
CGI_10009331	Growth hormone secretagogue receptor type 1	crassostrea_gigas
CGI_10012154	C-C chemokine receptor type 5	crassostrea_gigas
CGI_10022932	Neuromedin-U receptor 1	crassostrea_gigas
CGI_10025276	Growth hormone secretagogue receptor type 1	crassostrea_gigas
CGI_10026655		crassostrea_gigas
D915_05109	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	fasciola_hepatica_prjna179522
ECPE_0000219101	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	echinostoma_caproni_prjeb1207
PXEA_0002723401	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	protopolystoma_xenopodis_prjeb1201
SCUD_0000629701	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	schistosoma_curassoni_prjeb519
Sjp_0057450	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	schistosoma_japonicum_prjea34885
SMRZ_0001551401	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	schistosoma_margrebowiei_prjeb522
SMTD_0000981201	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	schistosoma_mattheei_prjeb523
SROB_0001283401	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	schistosoma_rodhaini_prjeb526
T265_15440		opisthorchis_viverrini_prjna222628
TRE_0000682001	Rhodopsin-like orphan GPCR,putative (inferred by orthology to a <i>S. mansoni</i> protein)	trichobilharzia_regenti_prjeb4662

*Appendix G: List of endothelial cell surface marker from Durr et al., 2004*

<b>Product description</b>	<b>Produce name</b>	<b>Species</b>	<b>Uniprot identifier</b>	<b>Ensembl identifier of Human orthologue</b>
Endothelial plasminogen activator inhibitor	SERPINE1	R	P20961	ENSG00000106366
Endothelial actin-binding protein	FLNA	H	P21333	ENSG00000196924
MUC18	MCAM	R	x	ENSG00000076706
Integrin alpha-5 (CD49e)	ITGA5	M	P11688	ENSG00000161638
Platelet endothelial tetraspan antigen-3	CD151	R	Q9QZA6	ENSG00000177697
Vascular endothelial-cadherin 1	CDH5	M	P55284	ENSG00000179776
EDG-1	S1PR1	R	P48303	ENSG00000170989
MAC-inhibitor (CD59)	CD59	R	P27274	ENSG00000085063
MRP-1 (CD9)	CD9	R	P40241	ENSG00000010278
Platelet-endothelial cell adhesion molecule-1 (CD31)	PECAM1	R	x	ENSG00000261371
Aminopeptidase N (CD13)	ANPEP	R	P97449	ENSG00000166825
Von Willebrand factor	VWF	R	P04275	ENSG00000110799
Caveolin-1	CAV1	R	Q8VIK9	ENSG00000105974
Endothelial cell-selective adhesion molecule	ESAM	M	Q925F2	ENSG00000149564
Integrin beta-1 (CD29)	ITGB1	R	P49134	ENSG00000150093
Annexin V	ANXA5	R	P14668	ENSG00000164111
Podocalyxin	PODXL	R	x	ENSG00000128567
Intercellular adhesion molecule-1 (I-CAM1; CD54)	ICAM1	R	Q00238	ENSG00000090339
Endothelial differentiation-related factor 1	EDF1	H	O60869	ENSG00000107223
Intercellular adhesion molecule-2 (I-CAM2; CD102)	ICAM2	M	P35330	ENSG00000108622
Na+K+ transporting ATPase alpha 1	ATP1A1	M	P06685	ENSG00000163399
Vascular endothelial cell specific protein 11	SEPT2	R	x	ENSG00000168385

Integrin alpha V	ITGAV	M	P43406	ENSG00000138448
Endothelin converting enzyme	ECE1	R	P42893	ENSG00000117298
Thrombomodulin	THBD	R	x	ENSG00000178726
Scavenger receptor class F	SCARF1	H	x	ENSG00000074660
Microvascular endothelial differentiation gene 1	DNAJB9	R	P97554	ENSG00000128590
Integrin alpha-3 (CD49c)	ITGA3	M	Q62470	ENSG00000005884
5'-nucleotidase (CD73)	NT5E	R	P21588	ENSG00000135318
EDG-2	BUD31	H	P41223	ENSG00000106245
H-CAM (CD44)	CD44	R	O08779	ENSG00000026508
Tyrosine-protein kinase receptor TIE-2	TEK	R	Q9QW24	ENSG00000120156
Transferrin receptor (CD71)	TFRC	H	P02786	ENSG00000072274
Angiotensin-converting enzyme (CD134)	ACE	R	P12821	ENSG00000159640
Tight junction protein 2 - ZO2	TJP2	R	Q9UDY2	ENSG00000119139
Endothelial collagen	COL8A1	H	P27658	ENSG00000144810
Sialomucin (CD34)	CD34	M	Q64314	ENSG00000174059
Tumor endothelial marker 4 (TEM4)	ARHGEF17	H	Q96PFE	ENSG00000110237
APC protein	APC	H	P25054	ENSG00000134982
PAR-1B alpha	MARK2	H	x	ENSG00000072518
Endomucin	EMCN	R	x	ENSG00000164035
Annexin IV	ANXA4	R	P55260	ENSG00000196975
Vascular cell adhesion protein 1	VCAM1	R	P29534	ENSG00000162692
Chemokine receptor CCX CKR	ACKR4	M	-	ENSG00000129048
Vascular endothelial-cadherin 2	PCDH12	H	Q9NPG4	ENSG00000113555
Tight junction protein 1 - ZO1	TJP1	M	P39447	ENSG00000104067
Purinergic receptor 5	LPAR6	H	P43657	ENSG00000139679
Ecto-apyrase (CD39)	ENTPD1	M	P55772	ENSG00000138185
MECA32, PV-1	PLVAP	M, R	x, Q9WV78	ENSG00000130300
Scavenger receptor class B type I	SCARB1	R	x	ENSG00000073060

RAGE	AGER	R	Q63495	ENSG00000204305
Tumor endothelial marker 6 (TEM6, Tensin 3)	TNS3	H	Q96PE0	ENSG00000136205
MDR 1A	ABCB1	R	P21447	ENSG00000085563
Integrin alpha-1 (CD49a)	ITGA1	R	P18614	ENSG00000213949
Alpha-2 macroglobulin	A2M	M	P28666	ENSG00000175899
Vascular endothelial junction-associated molecule	JAM2	H	P57087	ENSG00000154721
Aquaporin-CHIP	AQP1	R	P29975	ENSG00000240583
Dipeptidyl peptidase IV (CD26)	DPP4	R	P14740	ENSG00000197635
Nicotinic acetylcholine receptor alpha 3	CHRNA3	M	-	ENSG00000080644
Platelet-derived growth factor receptor	PDGFRB	M	P05622	ENSG00000113721
P2Y purinoceptor 6	P2RY6	R	-	ENSG00000171631
Nitric-oxide synthase	NOS1	R	P29476	ENSG00000089250
Scavenger receptor (CD36)	CD36	R	x	ENSG00000135218
EGF	EGF	H	x	ENSG00000138798
Vascular adhesion protein-1	AOC3	M	O70423	ENSG00000131471
Muscarinic acetylcholine receptor M3	CHRM3	R	x	ENSG00000133019
Angiotensin II receptor (1 or 2)	AGTR1	R	-	ENSG00000144891
Nicotinic acetylcholine receptor beta 3	CHRNA3	R	P12391	ENSG00000147432
Carbonic anhydrase IV	CA4	R	P48284	ENSG00000167434
Bradykinin receptor B2	BDKRB2	R	-	ENSG00000168398
Adrenergic receptor alpha 2B	N/A	H	x	ENSG00000222040

### ***Appendix H: List of supplementary tables***

Supplementary files listed below and this list are available at

<https://doi.org/10.17863/CAM.10322> in .csv and .pdf format.

#### **Chapter 3**

Table S3.1	D13 vs D6 down-regulated genes.csv
Table S3.2	D13 vs D6 up-regulated genes.csv
Table S3.3	D13 vs D6 GO term enrichment.csv
Table S3.4	Genes with high expression during liver stages D13 to D21.csv
Table S3.5	GO term enrichment of genes with high expression during liver stages.csv
Table S3.6	D28 vs D21 down-regulated genes.csv
Table S3.7	D28 vs D21 up-regulated genes.csv
Table S3.8	D28 vs D21 GO term enrichment.csv
Table S3.9	D35 vs D28 down-regulated genes.csv
Table S3.10	D35 vs D28 up-regulated genes.csv
Table S3.11	D35 vs D28 GO term enrichment.csv

#### **Chapter 4**

Table S4.1	GO term enrichment of genes differentially expressed at day 17.csv
Table S4.2	HEPG2 vs non-HEPG2 up-regulated genes.csv
Table S4.3	HEPG2 vs non-HEPG2 down-regulated genes.csv
Table S4.4	HEPG2 vs non-HEPG2 GO term enrichment.csv

#### **Chapter 5**

Table S5.1	HUVEC co-culture vs worm-free up-regulated genes.csv
Table S5.2	HUVEC co-culture vs worm-free down-regulated genes.csv
Table S5.3	HUVEC co-culture vs worm-free GO enrichment.csv
Table S5.4	HUVEC co-culture vs worm-free pathway enrichment.csv
Table S5.5	HEPG2 co-culture vs worm-free up-regulated genes.csv
Table S5.6	HEPG2 co-culture vs worm-free down-regulated genes.csv
Table S5.7	HEPG2 co-culture vs worm-free GO enrichment.csv

Table S5.8	HEPG2 co-culture vs worm-free pathway enrichment.csv
Table S5.9	GripTite co-culture vs worm-free up-regulated genes.csv
Table S5.10	GripTite co-culture vs worm-free down-regulated genes.csv
Table S5.11	GripTite co-culture vs worm-free GO enrichment.csv
Table S5.12	GripTite co-culture vs worm-free pathway enrichment.csv