

Supplementary Material

1 SUPPLEMENTARY DATA

Table S1. Number of raw, trimmed and mapped reads of DNA and RNA sequencing, as well as their mapping efficiency.

	L1	L2	L3	A1	A2	A3	
Rawreads (pairs)	52 476 690	47 746 583	50 885 234	41 580 001	52 373 828	64 044 560	DNA
Readpairs after trimming	52 062 252	47 389 807	50 242 034	41 240 772	52 036 318	63 184 551	
Mappedreads (pairs)	23 598 870	20 337 026	21 025 544	21 656 667	23 075 867	29 588 511	
Mappedreads (singles)	12 570 731	11 163 879	10 170 005	8 124 080	13 555 979	15 502 292	
Mapping efficiency (%)	57.4	54.7	52.0	62.4	57.4	59.1	
Rawreads (pairs)	58 727 029	39 633 003	40 789 126	39 075 561	40 619 470	42 458 845	RNA
Mappedreads (pairs)	37 149 163	25 469 813	26 608 634	22 673 790	23 366 997	24 409 086	
Mappedreads (singles)	7 375 724	5 251 009	5 817 922	5 512 917	5 046 240	5 503 114	
Mapping efficiency (%)	69.5	70.9	72.4	65.1	63.7	64.0	

Table S2. Overview of methylated positions in larvae and adults: Number of all cytosines(n(C), number of all methylated cytosine (n(5mC)); single larvae sample (L1-3), single adult sample (A1-3), larvae sample in total (L), adult sample in total (A), positions which are specific for larvae (larvae) or imagoes (adults) or which they have in common (both).

Context	Sample							
	L1	L2	L3	A1	A2	A3	L	A
N(C)	131 294 294	131 294 294	131 294 294	131 294 294	131 294 294	131 294 294	131 294 294	131 294 294
N(C) in CpG context	26 147 173	26 147 173	26 147 173	26 147 173	26 147 173	26 147 173	26 147 173	26 147 173
N(C) in CHG context:	20 727 654	20 727 654	20 727 654	20 727 654	20 727 654	20 727 654	20 727 654	20 727 654
N(C) in CHH context	84 419 467	84 419 467	84 419 467	84 419 467	84 419 467	84 419 467	84 419 467	84 419 467
N(5mC) in CpG context	116 635	70 829	79 830	87 521	93 376	180 024	38 244	47 456
N(5mC) in CHG context	50 234	27 187	33 917	37 967	35 032	79 004	3408	3864
N(5mC) in CHH context	231 042	133 377	162 071	132 284	167 494	375 192	20 062	19 851
N(5mC)	397 911	231 393	275 818	257 772	295 902	634 220	61 714	71 171
5mC in CpG context (%)	0.45	0.27	0.31	0.33	0.36	0.69	0.15	0.18
5mC in CHG context (%)	0.24	0.13	0.16	0.18	0.17	0.38	0.02	0.02
5mC in CHH context (%)	0.27	0.16	0.19	0.16	0.20	0.44	0.02	0.02
5mC (%)	0.30	0.18	0.21	0.20	0.23	0.48	0.05	0.05

Table S3. Summary of the methylated positions according to their sequence context (context type), and their sample:positions only in larvae (larvae), positions only in imagoes (adults), positions in larvae and adults (both).

Sample	Context type	N(5mC)	5mC(%)
larvae		27 523	27.89
adults		36 980	37.47
both		34 191	34.64
	CpG	61 484	62.30
	CHG	5982	6.06
	CHH	31 228	31.64

Table S4. Summary of the 4946 differential methylated loci (DMLs) and 4960 regions (DMRs) according to their position in the genome (region type) and in which sample they occur: total number (n), percentage (%), all following regions combined (all), gene body (gb), regulatory region (rr), intergenic region (ir).

Sample	Region type	N(DMLs)	DMLs (%)	N(DMRs)	DMRs (%)
larvae	all	3531		3020	
larvae	gb	1318	37.3	1526	50.5
larvae	rr	1563	44.3	1312	43.4
larvae	ir	650	18.4	182	6.0
adults	all	1415		1940	
adults	gb	665	47.0	1060	54.6
adults	rr	641	45.3	783	40.4
adults	ir	109	7.7	97	5.0

Table S5. Differential expression analysis: Upregulated (up), downregulated (down) in larvae compared to imagoes, not differential expressed (n.s.), undetected (n.d.) genes/ transcripts.

Expression type	Number of genes	Fraction of genes %	Number of transcripts	Fraction of transcripts %
Up	3729	23.99	6608	24.00
Down	4077	26.23	6536	23.74
N.s.	2299	14.79	4075	14.80
N.d.	5437	34.98	10 311	37.45

Table S6. Significantly enriched GO terms, which exclusively can be found for up regulated (up), down regulated (down) expressed genes of larvae compared to adults, or for genes being not differential expressed (n.s.). Genes terms, which have a stage specific function for imagoes (*).

Namespace	ID	Name	Expression type
	GO:0007568	aging	dn
	GO:0097190	apoptotic signaling pathway	
	GO:0034599	cellular respon.se to oxidative stress	
	GO:0048588	developmental cell growth	
	GO:0048589	developmental growth	
	GO:0015985	energy coupled proton tran.sport, down electrochemical gradient	
	GO:0051156	glucose 6-phosphate metabolic process	
	GO:0019682	glyceraldehyde-3-phosphate metabolic process	
	GO:0006119	oxidative phosphorylation	
	GO:0031099	regeneration	
	GO:0006465	signal peptide processing	
	GO:0042246	tissue regeneration	
	GO:0006099	tricarboxylic acid cycle	
	GO:0072350	tricarboxylic acid metabolic process	
	GO:0006437	tyrosyl-tRNA aminoacylation	
	GO:0048469	cell maturation	
	GO:0030030	cell projection organization	
	GO:0002181	cytoplasmic translation	
	GO:0002182	cytoplasmic translational elongation	
	GO:0009263	deoxyribonucleotide biosynthetic process	
	GO:0046385	deoxyribose phosphate biosynthetic process	
	GO:0019692	deoxyribose phosphate metabolic process	
	GO:0043650	dicarboxylic acid biosynthetic process	
	GO:0006270	DNA replication initiation	
	GO:0007306*	eggshell chorion assembly	
	GO:0009790*	embryo development	
	GO:0048598*	embryonic morphogenesis	
	GO:0044419	interspecies interaction between organisms	
	GO:0099118	microtubule-based protein tran.sport	
	GO:0048666	neuron development	
	GO:0030182	neuron differentiation	
	GO:0006490	oligosaccharide-lipid intermediate biosynthetic process	
	GO:0007309*	oocyte axis specification	

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Namespace	ID	Name	Expression type
Biological process	GO:0007308*	oocyte construction	down
	GO:0048599*	oocyte development	
	GO:0009994*	oocyte differentiation	
	GO:0043543	protein acylation	
	GO:0061512	protein localization to cilium	
	GO:0031503	protein-containing complex localization	
	GO:2000765	regulation of cytoplasmic translation	
	GO:0006275	regulation of DNA replication	
	GO:1902652	secondary alcohol metabolic process	
	GO:0007600	sensory perception	
	GO:0016073	snRNA metabolic process	
	GO:0016125	sterol metabolic process	
	GO:0006903	vesicle targeting	
	GO:0048199	vesicle targeting, to, from or within Golgi	
	GO:0001525	angiogenesis	
	GO:0001568	blood vessel development	
	GO:0034219	carbohydrate transmembrane transport	
	GO:0008643	carbohydrate transport	
	GO:0045165	cell fate commitment	
	GO:0001709	cell fate determination	
	GO:0060322	head development	
	GO:0048232	male gamete generation	
	GO:0006044	N-acetylglucosamine metabolic process	
	GO:0000726	non-recombinational repair	
	GO:0070071	Proton-transporting two-sector ATPase complex assembly	
	GO:0070507	regulation of microtubule cytoskeleton organization	
	GO:0001101	response to acid chemical	
	GO:0035295	tube development	
	GO:0035239	tube morphogenesis	
		GO:0044769	ATPase activity, coupled to transmembrane movement of ions, rotational mechanism
	GO:0016778	diphosphotransferase activity	
	GO:0004726	non-membrane spanning protein tyrosine phosphatase activity	
	GO:0004576	oligosaccharyl transferase activity	
	GO:0042803	protein homodimerization activity	

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Namespace	ID	Name	Expression type
Molecular function	GO:0046933	Proton-transporting ATP synthase activity, rotational mechanism	up
	GO:0016748	succinyltransferase activity	
	GO:0016769	transferase activity, transferring nitrogenous groups	
	GO:0004831	tyrosine-tRNA ligase activity	
	GO:0022843	voltage-gated cation channel activity	
	GO:0010997	anaphase-promoting complex binding	down
	GO:0004407	histone deacetylase activity	
	GO:0016799	hydrolase activity, hydrolyzing N-glycosyl compounds	
	GO:0099516	ion antiporter activity	
	GO:0060090	molecular adaptor activity	
	GO:0050664	oxidoreductase activity, acting on NAD(P)H, oxygen as acceptor	
	GO:0034212	peptide N-acetyltransferase activity	
	GO:0009982	pseudouridine synthase activity	
	GO:0017056	structural constituent of nuclear pore	
	GO:0044389	ubiquitin-like protein ligase binding	
	GO:0015144	carbohydrate transmembrane transporter activity	n.s.
	GO:0051010	microtubule plus-end binding	
	GO:0008448	N-acetylglucosamine-6-phosphate deacetylase activity	
	GO:0016653	oxidoreductase activity, acting on NAD(P)H, heme protein as acceptor	
	GO:0015295	solute:proton symporter activity	
GO:0019789	SUMO transferase activity	dn	
GO:0042162	telomeric DNA binding		
GO:0005885	Arp2/3 protein complex		
GO:1990204	oxidoreductase complex		
GO:0045259	Proton-transporting ATP synthase complex		
GO:0045261	Proton-transporting ATP synthase complex, catalytic core F(1)		
GO:0045263	Proton-transporting ATP synthase complex, coupling factor F(o)		
GO:0033178	proton-transporting two-sector ATPase complex, catalytic domain		
GO:0000214	tRNA-intron endonuclease complex		

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Namespace	ID	Name	Expression type
Cellular component	GO:0033588	Elongator holoenzyme complex	down
	GO:0019898	extrinsic component of membrane	
	GO:0032039	integrator complex	
	GO:0042555	MCM complex	
	GO:0097373	MCM core complex	
	GO:0034708	methyltransferase complex	
	GO:0044450	microtubule organizing center part	
	GO:0005643	nuclear pore	
	GO:0000808	origin recognition complex	
	GO:1903293	phosphatase complex	
	GO:0008287	protein serine/threonine phosphatase complex	
	GO:0031932	TORC2 complex	
	GO:0008023	transcription elongation factor complex	
	GO:0043527	tRNA methyltransferase complex	
	GO:0071203	WASH complex	
	GO:0043564	Ku70:Ku80 complex	n.s.
	GO:0000323	lytic vacuole	
	GO:0005846	nuclear cap binding complex	
GO:0005968	Rab-protein geranylgeranyltransferase complex		
GO:0034518	RNA cap binding complex		

Table S7. Genes of interest.



Table S8. Expression and differential methylation profile of genes of interest.

