

Table S1. Numbers of putative orphan MTases and their predicted activity across several species of bacteriophages

Phage*	Gene/orf	Genbank/NCBI ID	Reference	Number of orphan MTases within phage species*
<i>N6-Methyladenine activity</i>				
<i>Acintobacter buamannii</i> AB1	orf56	ADO14427.1	(103)	2
<i>Aeromonas</i> phage vB AsaM-56	gp11	YP_007007700.1	(104)	2
<i>Aggregatibacter</i> phage S1249	gp20	NC_013597.1	(105)	1
<i>Sinorhizobium</i> phage PBC5	orf6	NC_003324.1	-	1
<i>Stx2</i> converting phage II (<i>E. coli</i>)	orf66	NC_004914.2	(106)	2
<i>Colwellia</i> phage 9A	g088	YP_006489274.1	-	1
<i>Salmonella</i> phage epsilon15	p29	NC_004775	(107)	7
<i>Persicivirga</i> phage P12024L	gp28	NC_018272.1	(108)	2
<i>Shigella</i> phage Shf1a	gp62	YP_004414877.1	-	2
<i>Erwinia</i> phage phiEaH2	orf28	JX316028.1	(109)	2
<i>Providencia</i> phage Redjac	orf14	NC_018832.1	-	2
<i>Cronobacter</i> phage ESP2949-1	gp26	YP_007005414.1	(110)	3
<i>Escherichia</i> phage PBECO 4	orf147	KC295538.1	-	6
<i>Wolbachia</i> phage WO-A	orf263	NP_966069.1	(111)	4
<i>Riemerella</i> phage RAP44	gp58	NC_019490.1	(112)	1
<i>Bacteroides</i> phage B40-8	orf15	NC_011222.1	(113)	1
<i>Haemophilus</i> phage HP2	p12	NP_536818.1	(114)	3
<i>Flavobacterium</i> phage 11b	orf65	NC_006356.2	(115)	1
<i>Yersinia</i> phage PY100	orf33	CAJ28432.1	(72)	2
<i>Caulobacter</i> phage CcrMagneto	gp125	NC_019407.1	(116)	5
<i>Campylobacter</i> phage NCTC12673	gp102	YP_004421654.1	(117)	6
<i>Myxococcus</i> phage Mx8	orf5	NC_003085.1	-	1
<i>Enterobacteria</i> phage phi92	gp155	CBY99584.1	(118)	20
<i>Klebsiella</i> phage KP15	gp229	YP_003580090.1	-	3
<i>Prochlorococcus</i> phage P-SSM2	orf58	YP_214300.1	(119)	-
<i>Synechococcus</i> phage S-RIM2_R1_1999	orf32	YP_007675522.1	-	7
<i>Burkholderia</i> phage Bcep176	gp44	YP_355379.1	-	1
<i>Escherichia</i> phage rv5	gp096/gp230	YP_002003732.1	-	3

<i>Brucella</i> phage Tb	orf43	AEY69712.1	(68)	2
<i>Vibrio</i> phage_henriette_12B8	orf15	YP_007877988.1	-	13
<i>Pseudoalteromonas</i> phage H105/1	gp25	YP_004327118.1	(120)	1
<i>Enterococcus</i> phage phiEf11	orf51	NC_013696.1	(121)	4
<i>Leuconostoc</i> phage1-A4	orf2	ADD71725.1	(122)	2
<i>Lactococcal</i> phage 949	orf96	NC_015263.1	(123)	2
<i>Mycobacterium</i> phage Aeneas	gp62	AFL48073.1	(124)	29
<i>Streptococcus</i> phage 7201	orf13	NP_038314.1	(125)	12
<i>Lactobacillus</i> phage LBR48	orf47	ADF83450.1	(126)	7
<i>Gordonia</i> phage GTE7	gp37	YP_004934738.1	(127)	1
<i>Rhodococcus</i> phage ReqiDocB7	orf36	ADD80822.1	(128)	1
<i>Aeromonas</i> phage phiO18P	orf33	NC_009542.2	(129)	2
<i>Listeria</i> phage PSA	orf33	AJ312240	(130)	2
<i>Staphylococcus</i> phage 80	orf21	DQ908929.1	(131)	3
<i>Tetrasphaera</i> phage TJE1	orf2	HQ225832.1	(132)	1
m4c- Methylcytosine Activity				
<i>Salmonella</i> phage SE2	gp51	YP_005098147.1	(133)	1
<i>Pseudomonas</i> phage B3	orf48	NC_006548.1	(69)	2
<i>Acidithiobacillus caldus</i> AcaML1	orf24	JX507079.1	(134)	1
<i>Cronobacter</i> phage vB_CsaM_GAP32	gp391	YP_006987257.1	-	1
<i>Burkholderia</i> phage BcepIL02	gp41	NC_012743.2	(135)	5
<i>Cyanophage</i> KBS-M-1A	gp86	NC_020486.1	(136)	10
<i>Pectobacterium</i> phage ZF40	gp20	YP_007006929.1	(104)	1
<i>Prochlorococcus</i> phage P-HM1	orf39	YP_004322464.1	(119)	8
<i>Synechococcus</i> phage metaGMbCM1	gp157	YP_007001649.1	-	19
<i>Vibrio</i> phage VP882	gp37	YP_001039848.1	(137)	1
<i>Stenotrophomonas</i> phage S1	gp23	YP_002321474.1	(138)	1
<i>Mycobacterium</i> phage Baka	gp151	AEK08207.1	(124)	17
<i>Listeria</i> phage 2389	gp38	NP_511017.1	(130)	2
<i>Clostridium</i> phage phiCD119	gp70	YP_529621.1	(139)	
m5c-Methylcytosine Activity*				
<i>Klebsiella</i> phage KP15	gp115	YP_003579981.1	-	2
<i>Brucella</i> phage Tb	orf42	YP_007002108.1	(68)	2
<i>Bordetella</i> phage_BIP-1	p29	NP_996640.1	(140)	2
<i>Campylobacter</i> phage_CGC-2007	-	ABU53707.1	(141)	1

<i>Edwardsiella ictaluri</i> eiAU	orf6	HQ824553.1	-	1
<i>Vibrio</i> phage CP-T1	gp12	YP_007003052.1	(104)	6
<i>Caulobacter</i> phage CcrKarma	gp126	NC_019410.1	(116)	6
<i>Shigella</i> phage pSf1	gp21	YP_008059743.1	-	2
<i>Salmonella</i> phage epsilon15	gp41	NC_004775	(107)	3
<i>Pseudomonas</i> phage F116	orf 10	NC_006552.1	(142)	4
<i>Croceibacter</i> phage P2559S	orf55	NC_018276.1	(143)	1
<i>Sinorhizobium</i> phage PBC5	orf15	NC_003324.1	-	1
<i>Xanthomonas</i> phage vB_XveM_DIBBI	orf43	NC_017981.1	-	1
<i>Cyanophage</i> S-TIM5	orf152	NC_019516.1	(144)	1
<i>Rhodobacter</i> phage RcapNL	orf39	JQ066768.1	-	2
<i>Escherichia</i> phage K1-dep	orf31	GU196277.1	(145)	3
<i>Cronobacter</i> phage vB_CsaM_GAP32	gp151	YP_006987497.1	-	2
<i>Enterobacteria</i> phage RB43	orf127c	YP_239103.1	-	8
<i>Synechococcus</i> phage S-CBS2	gp5	YP_004421459.1	(146)	2
<i>Burkholderia</i> phage KL3	gp47	YP_004306459.1	(74)	16
<i>Thermus</i> phage P23-45	p14	YP_001467867.1	(147)	2
<i>Streptococcus</i> phage MM1	orf13	NP_150145.1	(148)	10
<i>Bacillus</i> Phage H2	-	M72412.1	(58)	7
<i>Lactococcal</i> phage 4268	orf14	NP_839905.1	(88)	3
<i>Enterococcus</i> phage phiFL1A	orf24	NC_013646.1	(149)	5
<i>Clostridium</i> phage phiCD27	gp62	YP_002290938.1	(150)	5
<i>Lactobacillus</i> phage LC-Nu	gp40	YP_358798.1	(151)	2
<i>Mycobacterium</i> phage corndog	gp7	NC_004685.1	(124)	68
<i>Streptomyces</i> phage phiBT1	orf9.1	NP_813725.1	(152)	1
<i>Saccharomonospora</i> phage PIS	orf81	JX006077.1	(153)	1
<i>Mycoplasma</i> phage MAV1	orf4	NC_001942.1	(154)	1
<i>Acidithiobacillus caldus</i> AcaML1	orf65	JX507079.1	(134)	1

* Methylation activity and distribution among species determined by blasting putative phage orphan MTases against the REBASE database. One example of a phage encoded MTase for each bacterial species is listed in column 1.

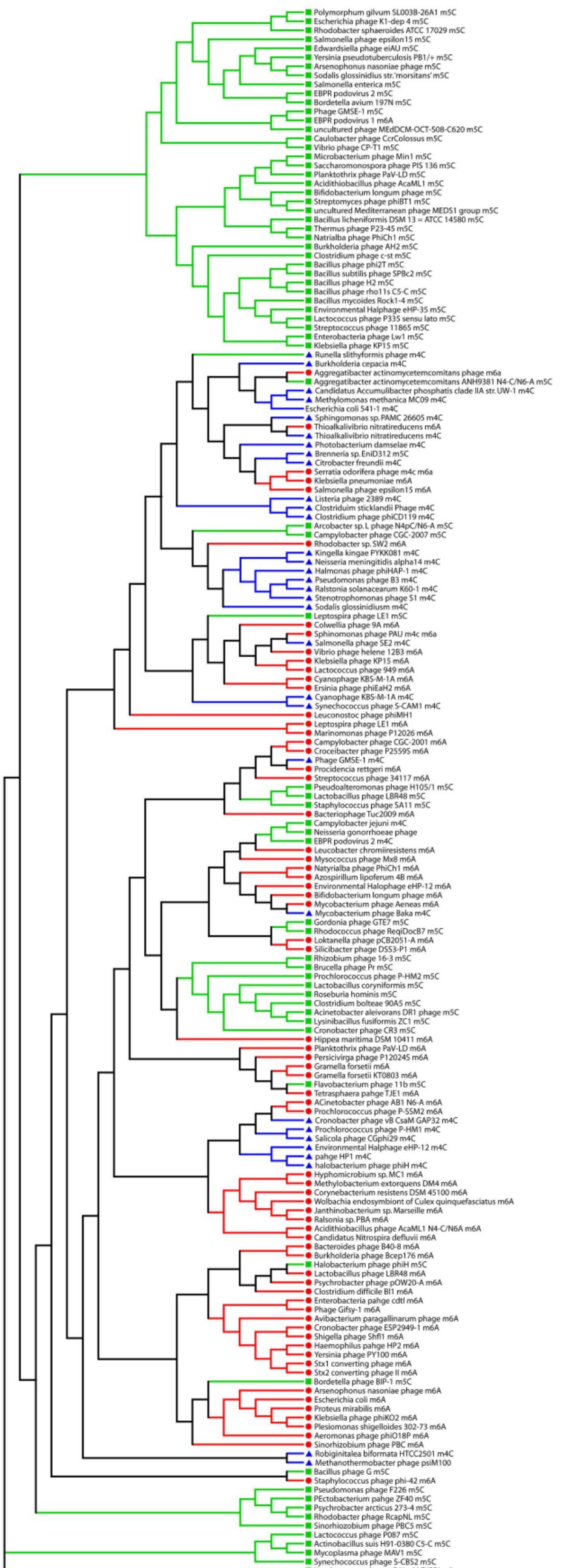


Figure S1. Phylogenetic tree of representative phage orphan methyltransferases highlighting the three functional classes. — = m5c, — = m6a — = m4c. BLASTP analysis was performed on putative bacteriophage methyltransferases against all methyltransferases available on REBASE to assign methyltransferase subtype (e value cut off 10^{-10}). One phage encoding a putative MTase associated with each bacterial species was selected to generate a representative tree. Sequence alignments of the putative MTases were performed using ClustalOmega (<http://www.ebi.ac.uk/Tools/msa/clustalo/>) and visualized using Mega5.

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