

Supplemental Table 1. List of supplementary strains included in this study

	Isolation					
	n°	Origin <sup>a</sup>	Host	Clinical manifestation	Year	Source <sup>b</sup>
<i>M. agalactiae</i>	4023	FR 82	Ovine	none	1988	BTM
<i>M. agalactiae</i>	4054	FR 58	Caprine	Mastitis	1986	Milk
<u><i>M. agalactiae</i></u>	<u>4206</u>	<u>FR 64</u>	<u>Ovine</u>	<u>Mastitis</u>	<u>1981</u>	<u>Milk</u>
<i>M. agalactiae</i>	4212	FR 84	Ovine	Mastitis	1986	Milk
<i>M. agalactiae</i>	4867	Spain	Caprine	Mastitis	<1990	Milk
<b><i>M. agalactiae</i></b>	<b>4908</b>	<b>FR 73</b>	<b>Caprine</b>	<b>Mastitis</b>	<b>&lt;1990</b>	<b>Milk</b>
<i>M. agalactiae</i>	5650	FR 81	Ovine	Mastitis	<1991	Milk
<i>M. agalactiae</i>	6355	Spain	Ovine	Arthritidis	1992	Joint
<i>M. agalactiae</i>	6833	Italy	Caprine	Mastitis	<1993	Milk
<i>M. agalactiae</i>	7320	Greece	Ovine	Mastitis	1987	Milk
<i>M. agalactiae</i>	7337	Greece	Caprine	Mastitis	1987	Milk
<i>M. agalactiae</i>	13462	FR 07	Caprine	Pneumonia	2003	Lung
<i>M. agalactiae</i>	14668	FR 84	Caprine	Pneumonia	2006	Lung
<i>M. agalactiae</i>	14780	FR 64	Ovine	none	2007	BTM
<i>M. agalactiae</i>	14793	Italy (Sardinia)	Ovine	Mastitis	nk	Milk
<u><i>M. agalactiae</i></u>	<u>14843</u>	<u>FR 64</u>	<u>Ovine</u>	<u>none</u>	<u>2007</u>	<u>BTM</u>
<i>M. agalactiae</i>	14962	FR 20	Ovine	Mastitis	2008	Mammary gland
<i>M. agalactiae</i>	14968	FR 69	Caprine	Mastitis	2008	BTM
<i>M. agalactiae</i>	15122	FR 84	Caprine	Mastitis	2008	Milk
<i>M. agalactiae</i>	15241	FR 14	Caprine	Mastitis	2009	BTM
<i>M. agalactiae</i>	15242	FR 86	Caprine	Mastitis	2009	Milk
<b><i>M. agalactiae</i></b>	<b>15291</b>	<b>FR 73</b>	<b>Caprine</b>	<b>none</b>	<b>1987</b>	<b>Joint</b>
<i>M. bovis</i>	6817	Italy	Bovine	Pneumonia	<1993	Lung
<i>M. bovis</i>	PG45 (TS)	US	Bovine	Mastitis	1962	nk

<sup>a</sup> Origin refers to the country of isolation and when in France (FR) also to the code of the department (administrative division).

<sup>b</sup> BTM, Bulk Tank Milk; nk, not known

Strains in bold are representative of the last outbreak of CA in the Savoy department (FR 73). Underscored strains are representative of the current ongoing CA episode in the Pyrénées Atlantiques department (FR 64).

Supplemental Table S2. CDSs identification in the prophage of *M. agalactiae* 14628

Mnemonic (strand) <sup>a</sup>	Product <sup>b</sup>	Length <sup>c</sup>	<i>M. conjunctivae</i> (similarity %) <sup>d</sup>	<i>M. bovigentialium</i> (similarity %) <sup>d</sup>
MAGb_2930 (+)	HP	204		
MAGb_2940 (-)	CHP, putative HNH endonuclease, predicted transmembrane protein	174	MCJ_005070 (49.7) <sup>f</sup>	MBVG_6650 (55.4)
MAGb_2950 (+)	CHP, predicted ATP-dependent endonuclease of the OLD family	346		MBVG_6660 (46.7)
MAGb_2960 (+)	CHP, putative metallo-hydrolase/oxidoreductase	278		MBVG_6670 (50.2)
MAGb_2970 (+)	CHP	121		MBVG_6680 (46.9)
MAGb_2980 (+)	CHP, putative DEAD-like helicase	447	MCJ_005050 (50.9)	MBVG_6690 (72.0)
MAGb_2990 (+)	N6-adenine DNA methyltransferase	294		MBVG_6720 (37.4)
MAGb_3000 (+)	CHP	206		
MAGb_3010 (+)	CHP, putative C-5 cytosine-specific DNA methyltransferase <sup>e</sup>	256		MBVG_6630 (41.8)
MAGb_3020 (+)	CHP	85		
MAGb_3030 (+)	HP	91		
MAGb_3040 (+)	HP	87		
MAGb_3050 (+)	CHP, putative DUF669 family phage protein	175	MCJ_005040 (41.6)	MBVG_6710 (59.3)
MAGb_3060 (+)	CHP, putative phage DNA polymerase	669		MBVG_6730 (80.4)
MAGb_3070 (+)	CHP, putative phage/plasmid primase	676	MCJ_005030 (51.3)	MBVG_6740 (70.6)
MAGb_3080 (-)	CHP, predicted transmembrane protein	201		MBVG_6750 (65.6)
MAGb_3090 (-)	CHP, predicted transmembrane protein	191	MCJ_004770 (41.7)	MBVG_6760 (75.5)
MAGb_3100 (-)	HP	66		
MAGb_3110 (-)	CHP	125	MCJ_004790 (50.4)	MBVG_6770 (68.0)
MAGb_3120 (+)	CHP	1008		MBVG_6780 (45.7)
MAGb_3130 (+)	HP	170		
MAGb_3140 (-)	CHP	531	MCJ_004820 (50.7)	MBVG_6800 (70.6)
MAGb_3150 (-)	CHP	307	MCJ_004830 (61.7)	MBVG_6810 (75.0)
MAGb_3160 (-)	CHP	132	MCJ_004850 (43.0)	MBVG_6830 (61.8)
MAGb_3170 (-)	CHP	154	MCJ_004860 (57.6)	MBVG_6840 (67.3)
MAGb_3180 (-)	CHP	146	MCJ_004870 (46.4)	MBVG_6850 (62.3)
MAGb_3190 (-)	CHP	184		MBVG_6860 (54.7)
MAGb_3200 (-)	CHP	882	MCJ_004890 (35.8)	MBVG_6870 (36.8)
MAGb_3210 (-)	CHP	112	MCJ_004900 (55.6)	MBVG_6880 (57.1)
MAGb_3220 (-)	CHP, putative phage prohead protein	618	MCJ_004910 (41.5)	MBVG_6890 (74.4)
MAGb_3230 (-)	CHP, putative phage portal protein	404	MCJ_004920 (57.5)	MBVG_6900 (79.8)
MAGb_3240 (-)	CHP, putative terminase large subunit	601	MCJ_004940 (64.7)	MBVG_6910 (79.3)
MAGb_3250 (-)	CHP	199		MBVG_6920 (63.4)
MAGb_3260 (-)	CHP	53		MBVG_6930 (53.2)
MAGb_3270 (-)	Integrase – recombinase ( <i>Xer</i> )	250		MBVG_6940 (64.8)

<sup>a</sup> CDSs are designated by the mnemonic codification used in the Molligen 3.0 database and their orientation in the genome is indicated in parenthesis.

<sup>b</sup> Hypothetical proteins (HP) have no homolog outside the *M. agalactiae* species. Conserved hypothetical proteins (CHP) share sequence similarity with proteins of unknown function identified in mollicutes or other bacteria.

<sup>c</sup> CDS length is given in nucleotides.

<sup>d</sup> Homolog CDSs in *M. conjunctivae* HRC/581T and *M. bovigentialium* cl-51080. The global similarity was calculated using the EMBOSS Needle global alignment tool. Pairwise alignments of protein sequences were run with default parameters. Protein alignments with a score < 35% are not indicated.

<sup>e</sup> Similar to C5 methylase MarMP of *M. arthritidis* bacteriophage MAV1 (global similarity = 59.2 %).

<sup>f</sup> An alternative start codon was chosen for MCJ\_005070 (genomic position 582226 instead of 582628).

Supplemental Table S3. List of CDSs from *M. agalactiae* strain 14628 that are not present in the already sequenced *M. agalactiae* strains PG2 and 5632.

Mnemonic	Feature type	Product	Mycoplasmal species and strains with BBH <sup>a</sup>	
			All genomes	Without <i>M. agalactiae</i> /
MAGb_0410	pseudogene	HP (N-term), no predicted function	<i>M. bovis</i> PG45 (_0475)	No hit
MAGb_0420	pseudogene	HP (C-term), no predicted function	<i>M. bovis</i> PG45 (_0475)	No hit
MAGb_0630	CDS	Type III RMS, endonuclease	<i>M. pulmonis</i> UAB CTIP	<i>M. pulmonis</i> UAB CTIP
MAGb_0640	CDS	Type III RMS, methylase	<i>M. pulmonis</i> UAB CTIP	<i>M. pulmonis</i> UAB CTIP
MAGb_0900	CDS	Type II RMS, endonuclease	<i>M. bovis</i> PG45 (_0366)	no
MAGb_0910	CDS	Type II RMS, methylase	<i>M. bovis</i> PG45 (_0365)	<i>MmmSC</i> PG1
MAGb_2500	pseudogene	CHP (N-term), predicted lipoprotein	No hit	No hit
MAGb_2520	pseudogene	HP (N-term), predicted lipoprotein	No hit	No hit
MAGb_2530	pseudogene	HP (C-term), predicted lipoprotein	No hit	No hit
MAGb_2610	pseudogene	Transposase	<i>M. bovis</i> PG45 (_0570)	No hit
MAGb_2620	CDS	Type II RMS, endonuclease	<i>M. bovis</i> PG45 (_0721)	<i>Mmc</i> GM12
MAGb_2630	CDS	Type II RMS, methylase	<i>M. bovis</i> PG45 (_0722)	<i>Mmc</i> GM12
MAGb_2640	CDS	Type II RMS, methylase	<i>M. bovis</i> PG45 (_0723)	<i>Mcc</i> California Kid
MAGb_2810	pseudogene	Type II RMS, endonuclease (C-term)	<i>M. anatis</i> 1340	<i>M. anatis</i> 1340
MAGb_2820	pseudogene	Type II RMS, endonuclease (central region)	<i>MmmSC</i> Gladysdale	<i>MmmSC</i> Gladysdale
MAGb_2830	pseudogene	Type II RMS, endonuclease (N-term)	<i>MmmSC</i> PG1	<i>MmmSC</i> PG1
MAGb_2840	CDS	Type II RMS, methylase	<i>M. bovis</i> Hubei-1	<i>M. anatis</i> 1340
MAGb_6200	CDS	Type II RMS, methylase	<i>M. bovis</i> PG45 (_0663)	<i>M. leachii</i> 99/014/6
MAGb_6210	pseudogene	CHP (N-term), no predicted function	<i>M. bovis</i> PG45 (_0662)	<i>M. leachii</i> 99/014/6
MAGb_6220	pseudogene	CHP (C-term), no predicted function	<i>M. leachii</i> PG50	<i>M. leachii</i> PG50
MAGb_8010	CDS	CHP, no predicted function	<i>M. bovis</i> PG45 (_0189)	<i>M. leachii</i> 99/014/6
MAGb_8020	CDS	CHP, no predicted function	<i>M. bovis</i> PG45 (_0190)	<i>M. leachii</i> PG50
MAGb_8030	CDS	CHP, no predicted function	<i>M. leachii</i> PG50	<i>M. leachii</i> PG50

<sup>a</sup>Protein blast and tblastn analysis were run against Molligen 3.0 databasis using individual CDS from *M. agalactiae* 14628. For clarity, the 35 CDS from the prophage already shown in the supplemental Table S2 are not listed here. E-value >e-8 were considered non significant (no hit). When the best blast hit (BBH) was obtained in *M. bovis* strain PG45, the corresponding mnemonic is mentioned in brackets (MBOVPG45\_). CHP, Conserved Hypothetical Protein; HP, Hypothetical Protein; RMS, Restriction Modification System

Supplemental Table S4. List of CDSs from *M. agalactiae* 14628 strain predicted as candidates for HGT.

Mnemonic	Product	BBH in <i>M. agalactiae/bovis</i> (without strain 14628)	BBH in Mollicutes (without <i>M. agalactiae/bovis</i> ) <sup>a</sup>	BBH in Spiroplasma group	Homologs in PG2 (if any) previously predicted as HGT <sup>b</sup>
<b>HGT predicted with the <i>Spiroplasma</i> phylogenetic group (that includes the “M. mycoides” cluster)</b>					
MAGb_0020	CDS19, ICE-related	MAGa6960	MCAP_0567	MCAP_0567	No homolog in PG2
MAGb_0090	CDS5, ICE-related	MAGa4920	MLC_2130	MLC_2130	<b>Only distant homolog (MAG4040)</b>
MAGb_0140	CDSA, ICE-related	MAGa2990	MCAP_0560	MCAP_0560	No homolog in PG2
MAGb_0150	CDS1, ICE-related	MBOVPG45_0213	MCAP0554	MCAP0554	No homolog in PG2
MAGb_0160	CDS19, ICE-related	MMB_0376	MLC_2230	MLC_2230	<b>MAG3880 and MAG 3890</b>
MAGb_0170	CDS22, ICE-related	MAG3860	MCAP0571	MCAP0571	<b>MAG3360 and 3860</b>
MAGb_0450	Oxidoreductase	MAGa4360	MMCAP2_0258	MMCAP2_0258	<b>MAG4120</b>
MAGb_0460	CHP, predicted lipoprotein	MAGa4370	MCAP0846	MCAP0846	<b>MAG4130</b>
MAGb_0540	CHP, predicted lipoprotein, DUF285 family	MAGa4450	MLEA_007790	MLEA_007790	<b>MAG4220</b>
MAGb_0550	CHP, predicted lipoprotein	MAGa4460	MCAP0843	MCAP0843	<b>MAG4230 and MAG4240</b>
MAGb_0620	CHP, DUF285 family	MAG4310	MSC_1005	MSC_1005	<b>MAG4310</b>
MAGb_0790	Glycerol kinase	MAG4470	MCAP0218	MCAP0218	<b>MAG4470</b>
MAGb_0800	Glycerol facilitator factor	MAG4480	MPUT_0572	MPUT_0572	<b>MAG4480</b>
MAGb_1010	ABC transporter, permease	MAGa5070	MMCAP2_0869	MMCAP2_0869	<b>MAG4600</b>
MAGb_1020	ABC transporter, ATP-binding	MAG4610	MSB_A0801	MSB_A0801	<b>MAG4610</b>
MAGb_1030	CHP	MAG4620	MMCAP2_0868	MMCAP2_0868	<b>MAG4620</b>
MAGb_1040	CHP, predicted lipoprotein	MAG4630	MPUT_0375	MPUT_0375	<b>MAG4630</b>
MAGb_1050	CHP, predicted lipoprotein	MAGa5110	MPUT_0245	MPUT_0245	<b>MAG4640</b>
MAGb_1250	Esterase/lipase	MAGa5310	MCAP0445	MCAP0445	<b>MAG4840 and 4850</b>
MAGb_1260	Glycosyl transferase	MMB_0523	MCAP0063	MCAP0063	<b>MAG4860 and 4870</b>
MAGb_1280	Malate permease	MAGa5360	MMCAP2_0037	MMCAP2_0037	<b>MAG4890</b>
MAGb_1330	CHP, predicted lipoprotein	MAGa5410	MMS_A0858	MMS_A0858	<b>MAG4940</b>
MAGb_1340	CHP, predicted lipoprotein	MAGa5420	MCAP0723	MCAP0723	<b>MAG4950 and 4960</b>
MAGb_1350	Hexosephosphate transport protein	MAG4970	MLEA_003800	MLEA_003800	<b>MAG4970</b>
MAGb_2060	Type I RMS, specificity subunit	MBOVPG45_0238	MSB_A0090	MSB_A0090	Not predicted (MAG5720)
MAGb_2070	Type I RMS, methylase	MAGa6290	MSB_A0088	MSB_A0088	Not predicted (MAG5730)
MAGb_2100	5-formyltetrahydrofolate cyclo-ligase	MAG5750	MMCAP2_0443	MMCAP2_0443	<b>MAG5750</b>
MAGb_2110	CHP, predicted transmembrane protein	MAG5760	MMCAP2_0112	MMCAP2_0112	<b>MAG5760</b>
MAGb_2120	CHP, predicted lipoprotein	MAG5770	MMCAP2_0293	MMCAP2_0293	<b>MAG5770</b>
MAGb_2370	CHP, predicted lipoprotein, Spma family	MAGa6720	MMCAP2_0092	MMCAP2_0092	No homolog in PG2
MAGb_2410	CHP, predicted lipoprotein, Spma family	MAGa6790	MLC_0790	MLC_0790	MAG6060
MAGb_2420	CHP, predicted lipoprotein, Spma family	/	MLC_0820	MLC_0820	No homolog in PG2
MAGb_2620	Type II RMS, endonuclease	MBOVPG45_0721	MMCAP2_0756	MMCAP2_0756	No homolog in PG2

MAGb_2630	Type II RMS, methylase	MMB_0669	MMCAP2_0755	MMCAP2_0755	No homolog in PG2
MAGb_2640	Type II RMS, methylase	MBOVPG45_0723	MCAP0049	MCAP0049	No homolog in PG2
MAGb_2820	Type II RMS (pseudo, central region), endonuclease		MSC_0949	MSC_0949	No homolog in PG2
MAGb_2830	Type II RMS (pseudo, N-term), endonuclease	MMB_0687	MSC_0949	MSC_0949	No homolog in PG2
MAGb_2840	Type II RMS, methylase	MMB_0688	MMS_A1042	MMS_A1042	Only distant homolog (MAG6680)
MAGb_2850	CHP, predicted lipoprotein	MAGa7370	MMCAP2_0396	MMCAP2_0396	<b>MAG6410</b>
MAGb_2880	CDSE, ICE-related	MAGa7400	MMS_A0526	MMS_A0526	<b>MAG6440</b>
MAGb_2890	CHP	MAG6460	MCAP0494	MCAP0494	<b>MAG6450</b>
MAGb_2900	CHP, predicted lipoprotein, DUF285 family	MAG6460	MSC_1005	MSC_1005	<b>MAG6460</b>
MAGb_2910	CHP, Fic protein family	MAGa7460	MSC_0192	MSC_0192	<b>MAG6470</b>
MAGb_2920	CHP, predicted lipoprotein, DUF285 family	MAGa7470	MLEA_008710	MLEA_008710	<b>MAG6480 and 6490</b>
MAGb_3010	CHP, putative methylase (marMP)	MAG4030	MCAP0556	MCAP0556	<b>MAG4030</b>
MAGb_3280	CHP, potentially truncated in N-term	MAGa7480	MSC_1031d	MSC_1031d	Not predicted (MAG6510)
MAGb_3290	CHP (Pseudo, C-term), predicted lipoprotein	MAGa7490	MSC_0519	MSC_0519	<b>MAG6520</b>
MAGb_3300	CHP (Pseudo, N-term), predicted lipoprotein	MAG6520	MLEA_002640	MLEA_002640	<b>MAG6520</b>
MAGb_3310	Glycerol ABC transporter, permease	MAG6530	MLEA_002650	MLEA_002650	<b>MAG6530</b>
MAGb_3320	Glycerol transporter subunit B	MAGa7510	MSB_A0465	MSB_A0465	<b>MAG6540</b>
MAGb_3330	Glycerol ABC transporter, ATP-binding	MAGa7520	MCAP0454	MCAP0454	<b>MAG6550</b>
MAGb_3340	CHP, predicted transmembrane protein	MAG6570	MLC_5290	MLC_5290	<b>MAG6560 and 6570</b>
MAGb_3900	Abortive infection protein AbiGI	MAGa8140	MMCAP2_0161	MMCAP2_0161	No homolog in PG2
MAGb_3910	Abortive infection protein AbiGII	MAGa8130	MSC_0175	MSC_0175	No homolog in PG2
MAGb_4000	CHP, predicted lipoprotein	MAGa8260	MLEA_004250	MLEA_004250	<b>MAG7130</b>
MAGb_4140	CHP, DUF285 family, potentially truncated in N-term	MAG7270	MSC_1005	MSC_1005	<b>MAG7270</b>
MAGb_4450	Esterase/lipase	MAGa0050	MLC_5050	MLC_5050	<b>MAG0030</b>
MAGb_4460	Esterase/lipase	MAG0040	MCAP0445	MCAP0445	<b>MAG0040</b>
MAGb_4470	NADH dependent flavin oxidoreductase	MAG0050	MMCAP2_0463	MMCAP2_0463	<b>MAG0050</b>
MAGb_4480	Lipoate-protein ligase A	MAGa0080	MLC_5020	MLC_5020	<b>MAG0060</b>
MAGb_4490	Lipoate-protein ligase A	MAGa0090	MLEA_002600	MLEA_002600	<b>MAG0070</b>
MAGb_4500	CHP	MAGa0100	MPUT_0111	MPUT_0111	<b>MAG0080</b>
MAGb_4510	Glycine cleavage system H protein	MAGa0110	MSB_A0457	MSB_A0457	<b>MAG0090</b>
MAGb_4520	CHP	MAGa0120	MPUT_0109	MPUT_0109	Not predicted (MAG0100)
MAGb_4610	CHP, predicted lipoprotein	MBOVPG45_0024	MLEA_001450	MLEA_001450	<b>MAG0190 and 0200</b>
MAGb_4620	CHP, predicted lipoprotein, DUF285 family	MAGa0220	MPUT_0237	MPUT_0237	<b>MAG0210 to MAG0240</b>
MAGb_4630	CHP, predicted lipoprotein, DUF285 family	MAGa0250	MLEA_005940	MPUT_0237	<b>MAG0210 to MAG0240</b>
MAGb_4720	Oligopeptide ABC transporter, ATP-binding	MAG0340	MLC_8220	MLC_8220	<b>MAG0340</b>
MAGb_4730	Oligopeptide ABC transporter, ATP-binding	MAG0350	MLC_8230	MLC_8230	<b>MAG0350</b>
MAGb_4740	Oligopeptide ABC transporter, permease	MAGa0360	MSC_0966	MSC_0966	<b>MAG0360</b>
MAGb_4750	Oligopeptide ABC transporter, permease	MAGa0370	MCAP0117	MCAP0117	<b>MAG0370</b>
MAGb_4760	Oligopeptide ABC transporter, substrate-	MAGa0380	MCAP0116	MCAP0116	<b>MAG0380</b>

	binding protein, predicted lipoprotein				
MAGb_4770	CHP, predicted transmembrane protein	MAGa0390	MMS_A1055	MMS_A1055	<b>MAG0390</b>
MAGb_5050	Aspartate-ammonia ligase	MAG0640	MSC_1015	MSC_1015	<b>MAG0640</b>
MAGb_5760	CHP, DUF285 family	MAGa1390	MMCAP2_0891	MMCAP2_0891	<b>MAG1330 and 1340</b>
MAGb_5890	Glucose-inhibited division protein	MAGa1510	MPUT_0540	MPUT_0540	<b>MAG1470</b>
MAGb_5910	D-lactate dehydrogenase	MAGa1530	MSC_0034	MSC_0034	<b>MAG1490</b>
MAGb_5920	Esterase/lipase	MAGa1540	MLC_3540	MLC_3540	<b>MAG1500</b>
MAGb_5990	ABC transporter, ATP binding protein	MAGa1640	MLC_5110	MLC_5110	Not predicted (MAG1630)
MAGb_6000	CHP, predicted membrane protein	MAG1640	MLC_5120	MLC_5120	<b>MAG1640</b>
MAGb_6030	CHP, predicted lipoprotein	MAG1670	MMCAP2_0015	MMCAP2_0015	<b>MAG1670</b>
MAGb_6110	CHP, predicted transmembrane protein	MAGa1750	MMCAP2_0594	MMCAP2_0594	Not predicted (MAG1770/MAG1760)
MAGb_6200	RMS, Cytosin-specific methylase, potentially truncated in N-term	MMB_0186	MLEA_007530	MLEA_007530	No homolog in PG2
MAGb_6210	CHP (Pseudo, N-term)	MMB_0187	MLEA_007540	MLEA_007540	No homolog in PG2
MAGb_6220	CHP (Pseudo, C-term)	MMB_0187	MLEA_007540	MLEA_007540	No homolog in PG2
MAGb_6520	Exodeoxyribonuclease V alpha chain	MBOVPG45_0626	MLC_4920	MLC_4920	<b>MAG2150 and 2160</b>
MAGb_6530	CHP	MAGa2190	MLEA_000570	MLEA_000570	<b>MAG2170</b>
MAGb_6650	CHP, predicted lipoprotein	MAG2220	MCAP0451	MCAP0451	<b>MAG2220</b>
MAGb_6780	Esterase/lipase	MAGa2460	MSB_A0456	MSB_A0456	<b>MAG2300</b>
MAGb_6790	Glycerol ABC transporter, ATP-binding	MAG2310	MMCAP2_0456	MMCAP2_0456	<b>MAG2310</b>
MAGb_6800	Glycerol ABC transporter, permease	MAGa2480	MPUT_0596	MPUT_0596	<b>MAG2320</b>
MAGb_6810	Glycerol ABC transporter, permease	MAGa2490	MCAP0452	MCAP0452	<b>MAG2330</b>
MAGb_6820	CHP, predicted lipoprotein	MAGa2500	MLEA_002640	MLEA_002640	<b>MAG2340</b>
MAGb_6950	NADH oxidase	MAG2460	MMS_A1115	MMS_A1115	<b>MAG2460</b>
MAGb_6960	Proton/glutamate symporter	MAGa2640	MLC_8900	MLC_8900	<b>MAG2470</b>
MAGb_7150	Alkylphosphonate ABC transporter, substrate-binding protein, predicted lipoprotein	MAGa2820	MMCAP2_0708	MMCAP2_0708	<b>MAG2690</b>
MAGb_7160	ABC transporter, ATP-binding	MAGa2830	MMCAP2_0707	MMCAP2_0707	<b>MAG2700</b>
MAGb_7170	Alkylphosphonate ABC transporter, permease	MAG2710	MPUT_0398	MPUT_0398	<b>MAG2710</b>
MAGb_7180	CHP	MAGa2850	MMCAP2_0080	MMCAP2_0080	<b>MAG2720</b>
MAGb_7290	CHP, predicted transmembrane protein	MAGa2960	MSC_0626	MSC_0626	<b>MAG2830</b>
MAGb_7300	CHP, predicted lipoprotein	MAG2840	MPUT_0333	MPUT_0333	<b>MAG2840</b>
MAGb_7310	CHP, predicted transmembrane protein	MAGa3230	MCAP0349	MCAP0349	<b>MAG2850</b>
MAGb_7320	CHP, predicted transmembrane protein with mycoplasma virulence signal region	MAGa3240	MMS_A0697	MMS_A0697	<b>MAG2860</b>
MAGb_7330	CHP, predicted lipoprotein	MAGa3250	MPUT4160	MPUT4160	<b>MAG2870</b>
MAGb_7340	CHP, predicted transmembrane protein	MAGa3260	MSB_A0365	MSB_A0365	<b>MAG2880</b>
MAGb_7350	CHP	MAGa3270	MPUT_0337	MPUT_0337	<b>MAG2890</b>
MAGb_7360	CHP	MAG2900	MSB_A0367	MSB_A0367	<b>MAG2900</b>

MAGb_7370	CHP	MAGa3290	MLEA_001690	MLEA_001690	<b>MAG2910</b>
MAGb_7380	CHP, predicted transmembrane protein	MAGa3300	MLEA_001700	MLEA_001700	<b>MAG2920</b>
MAGb_7390	ATP synthase alpha chain	MAG2930	MMCAP2_0576	MMCAP2_0576	<b>MAG2930</b>
MAGb_7400	ATP synthase beta chain	MAG2940	MMS_A0679	MMS_A0679	<b>MAG2940</b>
MAGb_7420	CHP, predicted lipoprotein	MAG2960	MLC_8090	MLC_8090	<b>MAG2960</b>
MAGb_7590	CHP, predicted lipoprotein , DUF31 family	MAG3120	MLC_2660	MLC_2660	<b>MAG3120</b>
MAGb_7600	Peptide methionine sulfoxide reductase	MAG3130	MPUT_0643	MPUT0800	<b>MAG3130</b>
MAGb_7730	CHP	MAGa3660	MLC_0350	MLC_0350	<b>MAG3330 and 3340</b>
MAGb_7920	RMS, Cytosine-specific methyltransferase	MAGa3950	MLC_2020	MLC_2020	No homolog in PG2
MAGb_7930	HP	/	MLC_2030	MLC_2030	No homolog in PG2
MAGb_7950	Type III RMS (Pseudo, N-term), methylase	MAGa1580	MMCAP2_0590	MMCAP2_0590	Not predicted (MAG1530)
MAGb_7960	Type III RMS (Pseudo, C-term), methylase	MAG1530	MMCAP2_0590	MMCAP2_0590	Not predicted (MAG1530)
MAGb_7980	CDSG, ICE-related	MAGa6910	MLC_2270	MLC_2270	No homolog in PG2
MAGb_8000	CDSF, ICE-related	MAGa6930	MPUT_0387	MPUT_0387	No homolog in PG2
MAGb_8010	CHP	MBOVPG45_0189	MLEA_005850	MLEA_005850	No homolog in PG2
MAGb_8020	CHP	MBOVPG45_0190	MSB_A0621	MSB_A0621	No homolog in PG2
MAGb_8030	CHP	/	MSB_A0622	MSB_A0622	No homolog in PG2
MAGb_8040	CDSE, ICE-related	MAGa6950	MMS_A0526	MMS_A0526	<b>MAG6440</b>
MAGb_0580	CHP	MAGa4490	Q9RQS4	Q9RQS4	<b>MAG4270</b>

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**HGT predicted with the *Hominis* phylogenetic group**

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MAGb_0630	Type III RMS, endonuclease	MAGa3840	MYPU_3950	/	No homolog in PG2
MAGb_0640	Type III RMS, methylase	MMB_0697	MYPU_3960	/	No homolog in PG2
MAGb_2610	Transposase (Pseudo)	MMB_0522	/	/	No homolog in PG2
MAGb_2940	CHP, putative HNH endonuclease, in prophage	/	Mbvg_6650	/	No homolog in PG2
MAGb_2950	CHP, predicted ATP-dependent endonuclease of the OLD family, in prophage	/	Mbvg_6660	/	No homolog in PG2
MAGb_2960	CHP, putative metallo-hydrolase/oxidoreductase, in prophage	/	Mbvg_6670	/	No homolog in PG2
MAGb_2970	CHP, in prophage	/	Mbvg_6680		No homolog in PG2
MAGb_2980	CHP, putative helicase, in prophage	/	Mbvg_6690	MLC_3520	No homolog in PG2
MAGb_3000	CHP, in prophage	MMB_0367	/	/	No homolog in PG2
MAGb_3050	CHP, putative DUF669 family, in prophage	/	Mbvg_6710	/	No homolog in PG2
MAGb_3060	CHP, putative DNA polymerase, in prophage	/	Mbvg_6730	/	No homolog in PG2
MAGb_3070	CHP, putative primase, in prophage	/	Mbvg_6740	/	No homolog in PG2
MAGb_3080	CHP, predicted transmembrane protein, in prophage	/	Mbvg_6750	/	No homolog in PG2
MAGb_3090	CHP, predicted transmembrane protein, in	/	Mbvg_6760	/	No homolog in PG2



prophage					
MAGb_3110	CHP, in prophage	/	Mbvg_6770	/	No homolog in PG2
MAGb_3120	CHP, in prophage	MMB_0391	Mbvg_6780	/	No homolog in PG2
MAGb_3130	HP, in prophage	MMB_0390	/	/	No homolog in PG2
MAGb_3140	CHP, in prophage	MMB_0389	MCJ_004820	/	No homolog in PG2
MAGb_3150	CHP, in prophage	/	Mbvg_6810	/	No homolog in PG2
MAGb_3170	CHP, in prophage	/	Mbvg_6840	/	No homolog in PG2
MAGb_3180	CHP, in prophage	/	Mbvg_6850	/	No homolog in PG2
MAGb_3190	CHP, in prophage	/	Mbvg_6860	/	No homolog in PG2
MAGb_3200	CHP, in prophage	/	Mbvg_6870	/	No homolog in PG2
MAGb_3210	CHP, in prophage	/	Mbvg_6880	/	No homolog in PG2
MAGb_3220	CHP, putative phage prohead protein	/	Mbvg_6890	/	No homolog in PG2
MAGb_3230	CHP, putative phage portal protein	/	Mbvg_6900	/	No homolog in PG2
MAGb_3240	CHP, putative phage terminase	/	Mbvg_6910	SPIC11_028	No homolog in PG2
MAGb_3250	CHP, in prophage	/	Mbvg_6920	/	No homolog in PG2

#### HGT predicted with the *Pneumoniae* phylogenetic group

MAGb_2990	Type II RMS, methylase, in prophage	/	MPN198	/	No homolog in PG2
MAGb_3270	Integrase-recombinase, in prophage	MBOVPG45_0822	UUR10_0216	/	Only distant homolog (MAG7110)

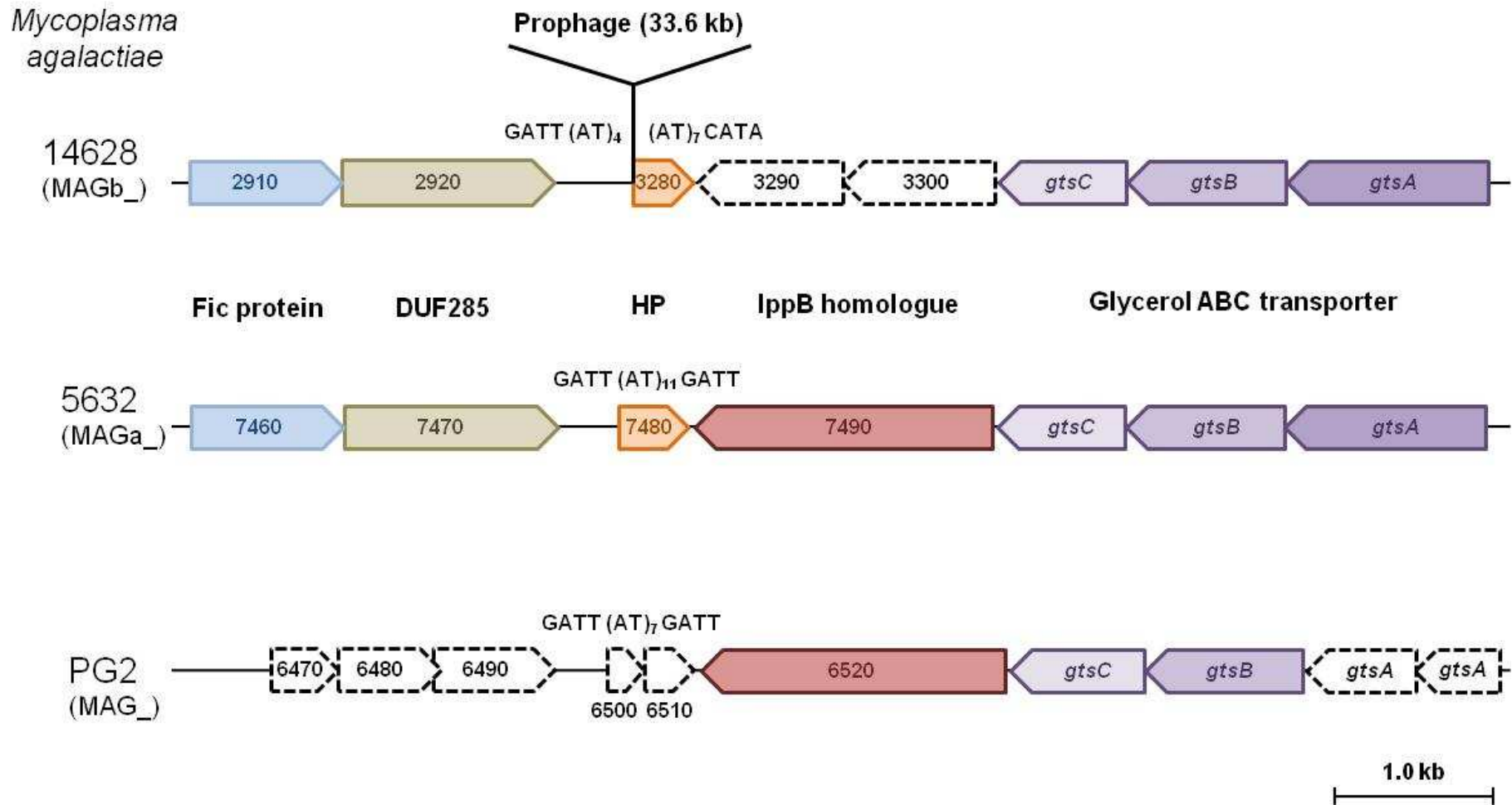
#### HGT predicted outside the Mollicutes

MAGb_4640	CHP YeiN, indigoidine synthase A-like protein	MAG0250	ACL_0802	/	Not predicted (MAG0250)
MAGb_4650	CHP YeiC, ribokinase/pfkB sugar kinase	MAGa0270	ACL_1374	/	Not predicted (MAG0260)
MAGb_3460	N4-cytosine or N6-adenine DNA methylase	MAGa7650	MCRO_0563	MLC_1630	<u>MAG6680</u>
MAGb_7200	Alcohol dehydrogenase	MAGa2870	MYPE4620	SPIC102_019	<u>MAG2740</u>
MAGb_2810	Type II RMS (pseudo, C-term), endonuclease	/	GIG_00827	/	No homolog in PG2
MAGb_0900	Type II RMS, endonuclease	No homolog in PG2	MBOVPG45_0366	/	/
MAGb_0910	Type II RMS, methylase	Only distant homolog (MAG6680)	MBOVPG45_0365	/	MSC_0186

<sup>a</sup> A BlastP analysis was run against all Mollicutes genomes available in databases except those from *M. agalactiae* and *M. bovis* but including the CDSs from the *M. bovis* genitalium prophage (this work). The best blast hit (BBH) is indicated by its mnemonic.

<sup>b</sup> For comparison purpose, homologs in PG2 are also reported with their predictive status for HGT as determined previously (Sirand-Pugnet et al., PLoS Genet. 3:e75, 2007). The mnemonic for PG2 CDS are in bold when HGT was predicted with the "*M. mycoides*" cluster and underlined when predicted outside Mollicutes.





Supplemental Figure 1. Genomic location of the prophage insertion site in *M. agalactiae* 14628 and corresponding locations in strains 5632 and PG2. The 33.6 Kb prophage insertion site and flanking CDSs in the 14628 chromosome are represented along with the corresponding regions in the genome of 5632 and PG2. For each strain, CDSs are indicated according to their orientation in the chromosome and approximate sizes. CDS numbers refer to the mnemonic codification used for each strain in the Molligen 3.0 database. Homologous CDSs are indicated by a colour code. Pseudogenes are indicated in dotted lines. Genes belonging to the glycerol ABC transporter and CDSs products homologous to *M. mycoides* subsp. *mycoides* LppB lipoprotein were predicted to be involved in horizontal gene transfer between *M. agalactiae* and mycoplasmas of the “*M. mycoides*” cluster ((Sirand-Pugnet et al., PLoS Genet. 3:e75, 2007). Other CDS products belong to the FIC protein family (FIC protein), contain a domain of unknown function 285 (DUF285), or are hypothetical proteins (HP) with no homolog outside the *M. agalactiae* species. CDS Magb\_3280 (3280) is potentially truncated at N-terminal as the result of the prophage insertion at an AT rich region.