Strains	Description	Reference
RN4220	Laboratory strain, restriction-defective derivative of RN450	(1)
RN450	NTCT8325 cured of $\phi$ 11, $\phi$ 12 and $\phi$ 13	(2)
RN451	RN450 lysogenic for $\phi$ 11	(2)
RN10359	RN450 lysogenic for $\phi$ 80 $lpha$	(3)
JP1794	RN451 (SaPlbov1 <i>tst::tet</i> M)	(4)
JP4028	RN451 Δ <i>rin</i> A	This study
JP4128	JP4028 (SaPlbov1 <i>tst::tet</i> M)	This study
JP4221	JP4028 (pJP740)	This study
JP5961	JP4128 (pJP740)	This study
JP4027	RN451 Δ <i>rin</i> B	This study
JP4127	JP4027 (SaPlbov1 <i>tst∷tet</i> M)	This study
JP3602	RN10359 (SaPI1 <i>tst::tet</i> M)	(5)
JP3603	RN10359 (SaPIbov1 <i>tst::tet</i> M)	(5)
JP4717	RN10359 Δ <i>rin</i> A	This study
JP5293	JP4717 (SaPlbov1 <i>tst::tet</i> M)	This study
JP5294	JP4717 (SaPI1 <i>tst::tet</i> M)	This study
JP5418	JP4717 (pJP740)	This study
JP5419	JP5293 (pJP740)	This study
JP5420	JP5294 (pJP740)	This study
JP5011	RN4220 lysogenic for <sub>\$LT</sub> pvl::tetM	This study
JP6895	JP5011 Δ <i>rin</i> A	This study
JP6391	JP6895 (pJP838)	This study
JP4223	RN451 (pJP742)	This study
JP4222	JP4028 (pJP742)	This study
JP4216	RN4220 (pJP743)	This study
JP5963	RN4220 (pJP744)	This study
JP5619	RN4220 (pJP746)	This study
JP5620	RN4220 (pJP747)	This study
JP5925	RN4220 (pJP748)	This study
JP5926	RN4220 (pJP749)	This study
JP7188	RN451 chimera $\phi$ 11- $\phi$ 69 ( $\phi$ 11 carrying P <i>ter</i> from $\phi$ 69)	This study
JP7242	JP7188 (SaPIbov1 <i>tst::tet</i> M)	This study
JP7218	JP7188 (pJP741)	This study
JP7243	JP7242 (pJP741)	This study
JP3960	Enterococcus faecalis V583	Lab strain
JP5093	Streptococcus pyogenes NZ131	Lab strain
JP3345	Lactococcus lactis IL1403	Lab strain
JP3762	RN4220 (pJP535)	This study
JP3763	RN4220 (pJP537)	This study
JP3766	RN4220 (pJP543)	This study
JP3767	RN4220 (pJP544)	This study
JP3779	RN4220 (pJP564)	This study
JP3780	RN4220 (pJP565)	This study

Table S1. Strains used in this study

Plasmids	Description	Reference
pMAD	Vector for efficient allelic replacement	(6)
pJP508	pMAD derivative. Deletion of $\textit{rin}A$ from $\phi 11$ and $80\alpha$	This study
pJP835	pMAD derivative. Deletion of <i>rin</i> A from $\phi$ SLT	This study
pJP956	pMAD derivative. Generation of the chimera $\phi$ 11- $\phi$ 69 ( $\phi$ 11 carrying Pter from $\phi$ 69)	This study
pCN51	Expression vector	(7)
pCN41	Used in transcriptional fusions to the staphylococcal $\beta$ -lactamase $\textit{blaZ}$	(7)
pCN42	Used in transcriptional fusions to the staphylococcal $\beta$ -lactamase <i>bla</i> Z. Conatins the P <i>cad</i> promoter	(7)
pJP740	Expression of $\phi$ 11 RinA, pCN51 derivative	This study
pJP741	Expression of $\phi$ 69 RinA, pCN51 derivative	This study
pJP838	Expression of $\phi$ SLT RinA, pCN51 derivative	This study
pJP742	Transcriptional analysis of $\phi$ 11 <i>ter</i> S, pCN41 derivative	This study
pJP743	Transcriptional analysis of $\phi 11~\mbox{terS}$ in presence of $\phi 11$ RinA, pCN42 derivative	This study
pJP744	Transcriptional analysis of $\phi 11 \ \textit{ter}S$ in absence of RinA, pCN42 derivative	This study
pJP746	Transcriptional analysis of $\phi$ 69 $\textit{terS}$ in presence of $\phi$ 69 RinA, pCN42 derivative	This study
pJP747	Transcriptional analysis of $\varphi 69~\mbox{ter}S$ in absence of RinA, pCN42 derivative	This study
pJP748	Transcriptional analysis of $\phi 69$ terS in presence of $\phi 11$ RinA, pCN42 derivative	This study
pJP749	Transcriptional analysis of $\phi 11 \ ter S$ in presence of $\phi 69$ RinA, pCN42 derivative	This study
pJP750	Expression in <i>E. coli</i> of $\phi$ 11 RinA, pGEX-4T-1 derivative	This study
pJP535	Transcriptional analysis of <i>ter</i> S from <i>E. faecalis</i> phage, in presence of the phage-encoded homologous RinA, pCN42 derivative	This study
pJP537	rinA mutant of pJP535	This study
pJP543	Transcriptional analysis of <i>ter</i> S from <i>S. pyogenes</i> phage, in presence of the phage-encoded homologous RinA, pCN42 derivative	This study
pJP544	rinA mutant of pJP543	This study
pJP564	Transcriptional analysis of <i>ter</i> S from <i>L. lactis</i> phage, in presence of the phage-encoded homologous RinA, pCN42 derivative	This study
pJP565	<i>rin</i> A mutant of pJP564	This study

## Table S2. Plasmids used in this study

Table S3. Oligonucleotides	used in	this	study
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Plasmid	Oligonucleotides	Sequence (5'-3')
pJP508	orf28phi11-1mB	CGC <u>GGATCC</u> GTGATAAGAAGTGACGC
	orf28phi11-2c	TTTAGTCATGAATACCCTCCG
	orf28phi11-3m	CGGAGGGTATTCATGACTAAAAGATTGGCGGAAGAGTTAGGG
	orf28phi11-4cE	CCG <u>GAATTC</u> TGTACTTCAGTGAAGTTATCG
pJP835	phiSLTp36-1mB	CGC <u>GGATCC</u> ATCAGAGCTGAAGTTTCATGG
	phiSLTp36-2c	GATATCATATATTGTGTTCCC
	phiSLTp36-3m	GGAACACAATATATGATATCAACTTTGTTAAAGCGGTAGCG
	phiSLTp36-4cE	CCG <u>GAATTC</u> AAACATTTTAAGCCGATGGGC
pJP956	orf29phi11-11mB	CGC <u>GGATCC</u> ATCAGTCTGATTTGATGAGGGC
	orf28phi11-15c	GTCTTCCCCTAACTCTTCCG
	orf30phi69-18m	CGGAAGAGTTAGGGGAAGACTAAGTTTGGAAAAAGTCTGG
	orf25phi69-17cS	ACGC <u>GTCGAC</u> TAATGTACTTCAGTGAAGTTATCG
pJP740	orf28phi11-6mB	CGC <u>GGATCC</u> GGTGATGTAAAAGTTAAAGAGCG
	orf28phi11-5cK	CGG <u>GGTACC</u> ACTGCCAATTTCAGTCTTCCC
pJP741	orf30phi69-1mB	CGCGGATCCGTTTGGTGTGATGTTGCAAGC
·	orf30phi69-6cK	CGG <u>GGTACC</u> ATGTATTATACCTAATTCATCTGC
pJP838	phiSLTp36-5mB	CGCGGATCCTTGCAGAATAAAGAACTAACG
·	phiSLTp36-6cE	CCG <u>GAATTC</u> GCCTTTTGCACAATCTTTGC
pJP742	orf28phi11-5mB	CGCGGATCCATCAAGCAGGAAAGAACGC
·	orf29phi11-3cK	
pJP743	orf28phi11-6mB	CGCGGATCCGGTGATGTAAAAGTTAAAGAGCG
	orf29phi11-3cK	CGG <u>GGTACC</u> TAACCTGCTGTAATTGCTGC
pJP744	orf28phi11-6mB	CGCGGATCCGGTGATGTAAAAGTTAAAGAGCG
	orf29phi11-3cK	CGG <u>GGTACC</u> TAACCTGCTGTAATTGCTGC
pJP746	orf30phi69-1mB	CGCGGATCCGTTTGGTGTGATGTTGCAAGC
	orf25phi69-2cK	CGG <u>GGTACC</u> TCATTTACCACCAACTCTCGC
pJP747	orf30phi69-1mB	CGC <u>GGATCC</u> GTTTGGTGTGATGTTGCAAGC
	orf30phi69-3c	CCTCATATCATAAGCAGTAGACATTGCTTTGCACCCTATCTCC
	orf30phi69-4m	GTCTACTGCTTATGATATGAGG
	orf25phi69-2cK	CGG <u>GGTACC</u> TCATTTACCACCAACTCTCGC
pJP748	orf28phi11-6mB	CGC <u>GGATCC</u> GGTGATGTAAAAGTTAAAGAGCG
	orf28phi11-7cK	CGG <u>GGTACC</u> GTCTTCCCCTAACTCTTCCG
	orf30phi69-5mK	CGG <u>GGTACC</u> ATTAAGTTTGGAAAAAGTCTGG
	orf25phi69-2cE	CCG <u>GAATTC</u> CAAATTAATTAATATGCCATCAG
pJP749	orf30phi69-1mB	CGC <u>GGATCC</u> GTTTGGTGTGATGTTGCAAGC
	orf30phi69-6cK	CGG <u>GGTACC</u> ATGTATTATACCTAATTCATCTGC
	orf28phi11-8mK	CGG <u>GGTACC</u> CTGAAATTGGCAGTAAAGTGGC
	orf29phi11-3cE	CCG <u>GAATTC</u> TAACCTGCTGTAATTGCTGC
pJP750	orf28phi11-5cX	CCG <u>CTCGAG</u> ACTGCCAATTTCAGTCTTCCC
	orf28phi11-7mB/b	CGC <u>GGATCC</u> ATGACTAAAAAGAAATACGG
pJP535	EF_V583-Pla-1mS	ACGC <u>GTCGAC</u> AATCCATCAAGGCCGCATCG
	EF_V583-Pla-4cB	CGC <u>GGATCC</u> TGTCGGCAGTTCCTCTAGC
pJP537	EF_V583-Pla-1mS	ACGC <u>GTCGAC</u> AATCCATCAAGGCCGCATCG
	EF_V583-Pla-2c	TAATTCTTCTTCCCGTTGACG
	EF_V583-Pla-3m	TCAACGGGAAGAAGAATTACATTAATTGGACTTGCCCAGC
	EF_V583-Pla-4cB	CGC <u>GGATCC</u> TGTCGGCAGTTCCTCTAGC
pJP543	Spy_NZ131-Pla-1mB	CGC <u>GGATCC</u> CGTGCTAGTGATGATAGAGC
	Spy_NZ131-Pla-4cE	CCG <u>GAATTC</u> TTTAATAGCCGCTGCTGTCGC

Plasmid	Oligonucleotides	Sequence (5'-3')
pJP544	Spy_NZ131-Pla-1mB	CGC <u>GGATCC</u> CGTGCTAGTGATGATAGAGC
	Spy_NZ131-Pla-2c	AAATTTGCTCTTCCTCCACCG
	Spy_NZ131-Pla-3m	GTGGAGGAAGAGCAAATTTGCCTATACAAGACATCTGTCG
	Spy_NZ131-Pla-4cE	CCG <u>GAATTC</u> TTTAATAGCCGCTGCTGTCGC
pJP564	bIL309-Alt-1mB	CGC <u>GGATCC</u> ACACTTGGTCAGTAGTATGG
	bIL309-Alt-5cE	CCG <u>GAATTC</u> TATAAATTTCTTGAACCTCACC
pJP565	bIL309-Alt-1mB	CGC <u>GGATCC</u> ACACTTGGTCAGTAGTATGG
	bIL309-Alt-2c	AGAATATGCTGTACGAATGCC
	bIL309-Alt-3m	CATTCGTACAGCATATTCTGGTTACGGTGGTTTTACATGG
	bIL309-Alt-5cE	CCG <u>GAATTC</u> TATAAATTTCTTGAACCTCACC

\* Sequences recognized by the restriction enzymes used in cloning are underlined.

Oligonucleotide	Sequence (5'-3')	Description
gyr-L	CACCATGTAAACCACCAGATA	qPCR. Expression of gyrB gene
gyr-U	TTATGGTGCTGGGCAAATACA	
ORF29-phi11-4m	CAGCAGAGTCTTTAGCAAGTCG	qPCR. Expression of $\phi$ 11 <i>terS</i> gene
ORF30-phi11-2c	CTTACCGCTCGAACCTCCACC	
ORF32-phi11-7m	GATGAGTTCGATGTAAAAGCG	qPCR. Expression of $\phi$ 11 ORF32
ORF32-phi11-8c	TGAACACGTGCCGATTCCG	
ORF34-phi11-4m	ATTAGGTAAGTACGAACCAATGG	qPCR. Expression of $\phi$ 11 ORF34
ORF34-phi11-5c	GTCTAACGAATCACTGTTACGG	
ORF37-phi11-1mB	CGCGGATCCAGTTGAACAAATACCGGAA AGG	qPCR. Expression of $\phi$ 11 ORF36
ORF37-phi11-7c	GTACCAAATTCTAAGAAACCAC	
ORF39-phi11-1m	TACAGAGGATACAATGGACGG	qPCR. Expression of $\phi$ 11 ORF39
ORF39-phi11-2c	GTGGAATCTGTATCCAGTCGC	
ORF45-phi11-2m	AACGGTCGATTTAGCAACATGG	qPCR. Expression of $\phi$ 11 ORF45
ORF45-phi11-3c	TCTCCATCAATGTATCTATACGC	
ORF49-phi11-6m	TTTTGTACCACAACCAGGCG	qPCR. Expression of $\phi$ 11 ORF49
ORF48-phi11-4cE	CCGGAATTCGTTTTTCGAGTCGTCGATTA CC	
ORF53-phi11-7m	AGTGGTTGAAAACTTCTGAGGG	qPCR. Expression of $\phi$ 11 ORF53
ORF53-phi11-8c	TACCTTTAGGGTTACTACCACG	
ORF1-phi11-1m	AGCGTACGAGTGTCTTAAATGG	qPCR. Expression of $\phi$ 11 <i>int</i>
ORF1-phi11-2c	CTGTGTGGGCAGGTTTTAAAGC	
ORF29-phi11-sp1c	TGGTCAATAGATCTCTGACGC	5' RACE φ11 <i>ter</i> S
ORF29-phi11-sp2c	AACCTCTTTTTCCACTTCATCG	
ORF29-phi11-sp3c	TAGAAGCAGATAACGCTAAAGC	
MH22	CCTCTTTTTCCACTTCATCG	5' RACE 80α <i>ter</i> S
MH23	GTCGATATACGCACCATGAAC	
UpTerSau-1m	AAAAATCATATCTAGATTGGCGG	EMSA
DwTerSau-2c	TCATTATATATTCATCTGCGAATCTC	

Figure S1. Comparison of the RinA protein sequences of  $\phi$ 11, 80 $\alpha$ ,  $\phi$ SLT and  $\phi$ 69. Colours indicate relative sequence conservation at each position, with red being most conserved and violet being least (adapted from alignment generated by PRALINE (8)). The scoring scheme works from 0 for the least conserved alignment position, up to 10 (\*) for the most conserved alignment position.

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69	EL	Y	Y	F	ĸ	KI	? F	т	L	N	L	т	G	v	A	QB	2 3		N	v	s	ĸ	s 1	C A	Y	D	М	F	١K	D	I	Ľ	V	R I	A	D	I	ΞI	G	I	I	н				
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Figure S2. Sequence features of the intergenic region containing the RinA binding site. The sequence shown corresponds to the longest fragment used as a probe in the electrophoretic mobility shift assays. The upper line shows the  $\phi$ 11 DNA sequence; the lower line shows an alignment with the same region from  $\phi$ 69. Asterisks designate nucleotides conserved between the two. Sequences corresponding to the end of *rin*A and the start of *ter*S are shown in red and blue, respectively. The first nucleotide, +1, in the  $\phi$ 11 transcript is shown in pink. Arrows indicate the inverted repeat in the intergenic region that is a potential terminator. Shaded boxes (green and yellow) correspond to the forward primers used to generate smaller DNA fragments of the promoter region used in mobility shift experiments. The heavy blue lines mark direct repeats of different sequences present at similar locations in this region of the  $\phi$ 11 and  $\phi$ 69 genomes.

<i>rin</i> A
AAAAATCATATCTAGATTGGCGGAAGAGTTAGGGGAAGACTGA
<b>AGACATACTAGTTAGATTAGCAGATGAATTAGGTATAATACATTAA</b> GTTTGGAAAAAGTCTGGAAAAA
* * ** ** ***** ** ** ** ** ** * * * * *
TT-TGAT ACCTTAAATGAGATATT   TAACGTCACTTTCGGTGTTAATATGATAGCGTAAGATATTGACTATCTTACTGCGTTTCCCTTATC   * **   * **   * **   * **   * **   * **   * **   * **   ** **
$\longrightarrow$ $\longleftarrow$
GCAATTAGGAATAAAGGATCTATGTGGGTTGGCTGATTATAGCCAATCC-TTTTTTAAAAAAG
GCAATTAGGAATAAAGGATCTATGTGGGTTGGCTGATTATAGCCAATCCCTTTTTTAATTTTAAAAAG
+1 terS
CGTATAGCGCGAGAGTTGGTGGTAAATGAAATGAAAACGAAAAAGAGATTCGCAGATGAATATAT
CGTATAGCGCGAGAGTTGGTGGTAAATGAA <b>ATGAACGAAAAACAAAAGAGATTCGCAGATGAATATAT</b>
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AATGA

## AATGA

**Figure S3. RinA does not control expression of \phi11** *int***. Real time quantification of the expression of** *int* **in wild-type \phi11 and a** *rin***A mutant at different times (0, 30 or 60 min) after MC induction of the phage lytic cycle. Expression was normalized to** *gyr***B, as described in Materials and Methods.** 



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