Table S1.- Bacterial strains and plasmids used in this work.

Item	Genotype or phenotype	Source or reference
Strains		
Bifidobacterium breve UCC2003	Isolate from nursling stool	(27)
Bifidobacterium breve UCC2003-phoP-481	pORI19-*tetW-phoP* insertion mutant of UCC2003	This study
Escherichia coli TOP10	F$^-$*mcrA* Δ(*mrr-hsd*RMS-*mcr*BC) F80*lacZ*ΔM15 Δ*lacX*74 *rec*A1 *deo*R *ara*D139 Δ(*ara-leu*)7697 *gal*U *gal*K *rsp*L (Str^r) *end*A1*nup*G	Invitrogen, Carlsbad, CA
Escherichia coli EC1010	*E. coli* JM101 with *repA* from pWV01 integrated in chromosome, km^r	(21)
Escherichia coli XL1-blue	Δ(*mcrA*)*183* Δ(*mcrCB-hsdSMR-mrr*)*173 endA1 supE44 thi-1 recA1* gyrA96 relA1 lac *[F'* proAB lacI^{q}ZΔM15 *Tn*10 *(*Tet^r*)]*	Stratagen
Escherichia coli M15	*lac,ara,gal,mtl,recA$^+$,uvr$^+$*[pREP4,*lacI,*km^r*]*	Quiagen
Lactococcus lactis NZ9000	MG1363, nisin-inducible overexpression host; *pepN*::*nisRK*	(6)
Plasmids		
pAM5	pBC1-pUC19 Tet^r [*tetW*]	(2)
pORI19	Em^r*, repA$^-$, ori+,* cloning vector	(21)
pORI19-phoP	Internal 481 bp fragment of *phoP* cloned in pORI19	This work
pORI19-tet-phoP	Internal 481 bp fragment of *phoP* and *tetW* cloned in pORI19	This work
pCR4-TOPO	Ap^r, *lacZ*α; TA cloning vector	Invitrogen

pCR4-TOPO-phoP	Ap^r, pCR4-TOPO containing *phoP* gene of *B. breve* UCC2003	This study
pCR4-TOPO-phoPR-Pro	Ap^r, pCR4-TOPO derivative carrying the *phoPR* promoter	This study
pCR4-TOPO-pstS-Pro	Ap^r, pCR4-TOPO derivative carrying the *pstS* promoter	This study
pCR4-TOPO-pit-Pro	Ap^r, pCR4-TOPO derivative carrying the *pit* promoter	This study
pNZ8048	Cm^r, nisin-inducible translational fusion vector	(6)
pNZ8048:phoP	Cm^r; pNZ8048 derivative containing translational fusion of *phoP* encoding DNA fragment to nisin-inducible promoter.	This study
pNZ44	Cm^r, pNZ8048 containing constitutive P44 promoter from *L. lactis* chromosome	(28)
pNZ44:phoP	Cm^r; pNZ44 derivative containing translational fusion of *phoP* encoding DNA fragment to p44 promoter.	This study
pNZ272	Cm^r, pSH71 derivative containing promoterless glucuronidase gene for promoter screening.	(35)
pNZ272:pho	pNZ272 derivative carrying the *phoPR* promoter	This study
pNZ272:pst	pNZ272 derivative carrying the *pstS* promoter	This study
pNZ272:pit	pNZ272 derivative carrying the *pit* promoter	This study

Ap^r, Em^r,Km^r Str^r and Tet^r, resistance to ampicillin, erythromycin, kanamycin , streptomycin and tetracycline, respectively.

Table S2. Oligonucleotides used in this work

Purpose	Primer	Sequence (5'→3')
Cloning of internal fragment of *phoP* in pORI19	phoPKOF	ctagtgctgcagggtatgacgtgtccgctgctg
	phoPKOR	gtcagttctagacggtcaatcagctgatgccg
Cloning *phoP* gene in pNZ44	RrFoefor	aaccatggcaggaaacaacagggtaatggactc
	RrFoerev	aatctagaggtcgcctttcatcatccggcaaacg
Clonig promoter region of phoPR in pNZ272	p2CSFfor	ctgcagatgatctgccagatatccat
	p2CSFrev	ggatccggaaagcaaggacaaggacta
Clonig promoter region of *pstS* in pNZ272	pPstfor	ggctgcagggaagcggaatcagtggaggagctgtcggtcgtc
	pPstRev	ggaggatccggaattcgagctgctcgaatacctgatgcagaa
Clonig promoter region of *pit* in pNZ272	pPitfor	ggctgcagccaagtcacgggcatattc
	pPitrev	ggaggatccacgccgaggaagttaaagatgg
Cloning *phoP* gene in pNZ8048	Bbr1683fNco1	tgcacgccatggcccatcaccatcaccatcaccatcaccatca cacgcgcattctcatcgtcgaag
	Bbr1683rXba1	tgcgcatctagattactccaccggggcgtcgatc
tetW gene amplification	tetWf	tcagctgtcgacatgctcatgtacggtaag
	tetWr	gcgacggtcgaccattaccttctgaaacat
qRT-PCR primers	psTS-7F	atggcaccatcggttacg
	psTS-7R	taggagtcgccgaccttg

	1573-31F	gtgaagctgctcgccaag
	1573-31R	gaatgtggaggccagacg
	rnpA-59L	GCATCGTTCTCATCGTTGG
	rnpA-59R	cgccttacgagccacttt
Primers for *pstS* upstream region amplification	ptsSird1f	tggctacaagatcgacgc
	ptsSird1r	caatggagcgaatgaggatg
	ptsSird2f	catgatggcctctttcccgcg
	ptsSird2r	ggaggagctgtcggtcgtg
	ptsSird3f	cttctgtttactgatttc
	ptsSird3r	gcttcgacggcggactgc
Primer for *pstC* upstream region amplification	ptsCirdf	caaggccatcaagaccaag
	ptsCirdr	gccgttttgtcttgcgaactc
Primers for *phoU* upstream region amplification	Bbr1573ird1f	gaaacggcatcgatgtcatac
	Bbr1573ird1r	taacgcgcatttctccagcc
	Bbr1573ird2f	gcacgccgataacatgaatttc
	Bbr1573ird2r	catgcggtcgaggtcatc
	Bbr1573ird3f	ccattatctacattgaag
	Bbr1573ird3r	cacggtttgagcggcttc
	Bbr1573ird1f	gaaacggcatcgatgtcatac
Primers for *phoR* upstream region amplification	Bbr1684ird1f	gcaaggacaaggactacg

	Bbr1684ird1r	gcaagcaccacaaacacg
	Bbr1684ird2f	gatattgtgctcccgctg
	Bbr1684ird2r	gacacacgcccggaaacac
	Bbr1684ird3f	ctttgctatattcgccctg
Others putatives target genes promoter regions	p1811for	tggcagcacccatttgaggttgcttg
	p1811rev	cgccactttcccgggaacgtgtcgctg
	p0824for	gactgacctgtcggcatggaacagccttg
	p0824rev	gattcggacgtaagaccttatggtg
	p1389for	ggtcgacacgccaagtaccgcggacagct
	p1390rev	aggcgatcacttcattgaacggtatattca
	patpBfor	tcgttcgcacagtggactgtaatttgttc
	patpBrev	gttgactgagggcagatgagcgccctcgt
	p1506for	aagtgctctccttttgagctcgttcattaacc
	p1506rev	caggagtggcgggatcgagaaataggga
	pczcDfor	tttctctcgcagccgattcgctcacatcg
	pczcDrev	agggatccggtaagcacggcggatatgactt
	p1771for	ttccactcgatgctcccgttggcgaaagctgac
	p1771rev	ctgccgtattcgtggcatgcgcggcgttcgt
	p1894for	atacagcatgccgttcggttgaaaa
	p1894rev	ccgaacgcaaggccgcccgcaacggc
*pst*S promoter primer extension analysis	ptsSird1r	caatggagcgaatgaggatg
	ptsSird2r	ggaggagctgtcggtcgtg
*pho*R promoter primer extension analysis	Bbr1684ird1r	gcaagcaccacaaacacg

	Bbr1684ird2r	gacacacgcccggaaacac
Amplification of DNA for sequencing ladders	p2CSFfor	ctgcagatgatctgccagatatccat
	p2CSFrev	ggatccggaaagcaaggacaaggacta
	pPstfor	ggctgcagggaagcggaatcagtggaggagctgtcggtcgtc
	pPstRev	ggaggatccggaattcgagctgctcgaatacctgatgcagaa

[a] Underlined oligonucleotide sequences show artificial restriction enzyme sites introduced for cloning.

Table S3

Basal Medium –Dissolve in 850 ml dH20

Chemically defined medium		**Phosphate Starvation Medium**	
β-glycerophosphate	19 g	MOPS	10 g
K_2HPO_4	3 g		
KH_2PO_4	3 g		
Sodium acetate	1 g	Sodium acetate	1 g
tri-ammonium citrate	0.6 g	tri-ammonium citrate	0.6 g
ascorbic acid	0.5 g	ascorbic acid	0.5 g
tyrosine	0.25 g	tyrosine	0.25 g
Tween 80	0.1%	Tween 80	0.1%
Glycerol	0.1%	Glycerol	0.1%

Trace metals per litre:

50 mg $MgSO_4.7H_2O$
10 mg $MnSO_4.4H_2O$ (pink)
5 mg $FeSO_4.7H_2O$ (green)
5 mg $CuSO_4.5H_2O$
0.5 mg $CoCl_2$
0.15 mg $ZnSO_4.7H_2O$

Vitamins per litre:

10 mg p-aminobenzoic acid
5 mg inosine
5 mg thymidine
2.5 mg 6,8-thioctic acid
2.5 mg biotin
2.0 mg pyridoxine-HCl
1.0 mg folic acid
1.0 mg nicotinic acid
1.0 mg Ca-(D^+)-pantothenate
1.0 mg riboflavin
1.0 mg thiamine-HCl
1.0 mg cobalamin

Bases (in 0.1N NaOH) per litre:

0.1 mg adenine
0.1 mg guanine
0.1 mg uracil
0.1 mg xanthine

Casamino acids-5 g/L

Sugar source (10% or 20% stock)
Add 100 ml or 50 ml per litre (1 % or 0.5% final concentration)

Cys-HCl (0.05 % final conc.)
Add ~8.3 ml (6 % stock) per litre of medium

Fatty acids (100 % stock)
Add at final conc. of 10 µg/ml

pH 6.8 and filter sterilise

For growth experiments the phosphate starvation medium was supplemented with 20 mM, 10 mM, 1 mM and 0.1 mM phosphate from a 100 mM potassium phosphate stock solution (pH7.0)

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