Supplementary material

β-glucan-producing *Pediococcus parvulus* 2.6: test of probiotic and immunomodulatory properties in zebrafish models

Pérez-Ramos A.¹, Mohedano M.L.¹, Pardo M.A.², López P.^{1*}

¹Laboratory of Molecular Biology of Gram-positive Bacteria. Department of Microorganisms and Plant Biotechnology. Biological Research Center (CIB), CSIC, Madrid, Spain ²Food Research Division, AZTI-Tecnalia, Bizkaia, Spain

* Correspondence:

Paloma López plg@cib.csic.es

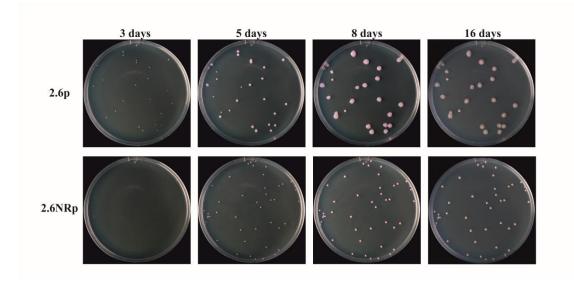


Figure S1. Detection of mCherry production by *P. parvulus* **2.6**[pRCR12] (2.6p) **and** *P. parvulus* **2.6NR**[pRCR12] (2.6NRp) **on solid media.** Bacterial colonies were grown in MRS plates during 16 days. Images of plates at 3 days, 5 days, 8 days and 16 days are depicted.

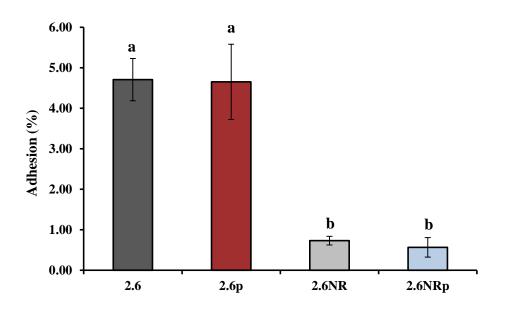


Figure S2. *P. parvulus* adhesion to a monolayer of Caco-2 cells. Enterocytes were exposed independently to four bacterial solution containing 2.6, 2.6[pRCR12] (2.6p). 2.6NR or 2.6NR[pRCR12] (2.6NRp) strains. Statistical significances are represented by different letters that mean a $P \le 0.01$.

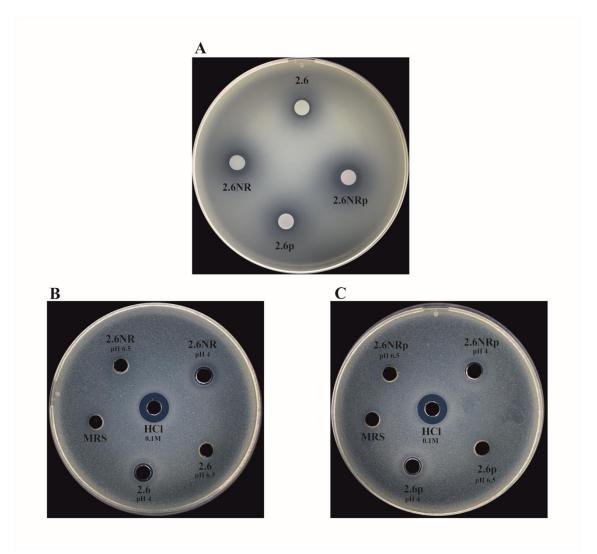


Figure S3. Antimicrobial activity of *P. parvulus* **strains against** *V. anguillarum*. The activity of spots of the four indicated strains was tested (A). A diffusion test was performed with the supernatants of stationary phase cultures of either 2.6 or 2.6NR (B) and either 2.6p or 2.6NRp (C). Samples of supernatants with pH 4.0 were neutralized to pH 6.5, and both samples were tested. 0.1 M HCl, and MRS medium were used as a positive and a negative controls.

Antibiotic	Concentrations tested (mg L ⁻¹)	2.6	2.6NR
Ampicillin	1	+	+
	2	+	+
	4*	_**	_**
	8	-	-
Gentamycin	4	_**	_**
	8	-	-
	16*	-	-
	32	-	-
Kanamycin	16	+	+
	32	_**	_**
	64*	-	-
	128	-	-
Streptomycin	16	_**	_**
	32	-	-
	64*	-	-
	128	-	-
Erythromycin	0.25	_**	_**
	0.5	-	-
	1*	-	-
	2	-	-
Clindamycin	0.25	_**	_**
	0.5	-	-
	1*	-	-
	2	-	-
Tetracycline	2	+	+
	4	_**	_**
	8*	-	-
	16	-	-
Chloramphenicol	1	+	+
	2	_**	_**
	4*	-	-
	8	-	-

Table S1. Antibiotic resistant profile of <i>P. parvulus</i> 2.6 and
--

*Microbiological cut-off values according to EFSA (2012). **MIC: minimal inhibitory concentration.