

1 *Supplementary Information*

2 **Chitosan/Cyclodextrin/TPP nanoparticles loaded**
 3 **with quercetin as novel bacterial quorum sensing**
 4 **inhibitors**

5 **Hao Thanh Nguyen^{1,3}, Francisco M. Goycoolea^{1,2,*}**

6 ¹ Institute for Biology and Biotechnology of Plants, University of Münster, Münster 48143, Germany;
 7 t_nguy32@uni-muenster.de

8 ² School of Food Science and Nutrition, University of Leeds, LS2 9JT United Kingdom;
 9 F.M.Goycoolea@leeds.ac.uk

10 ³ Faculty of Biotechnology, Vietnam National University of Agriculture, Vietnam; nthao.cnsh@vnua.edu.vn

11 *Correspondence: F.M.Goycoolea@leeds.ac.uk; Tel.: +44(0)-113-343-1412

12 **Table S1.** Physicochemical properties of unexpected CS_{70/5} NPs. (mean ± S.D., *n* = 3)

Mass ratio CS/CD/TPP (w/w/w)	Charge ratio (+/-)	Size (nm)	PDI	ζ potential (mV)	Production yield (%)
4/1/0	5.20	a	-	-	-
4/2/0	2.60	a	-	-	-
4/3/0	1.73	b	-	-	-
4/4/0	1.30	b	-	-	-
4/5/0	1.04	b	-	-	-
4/6/0	0.87	b	-	-	-
4/1.5/1	1.22	b	-	-	-
4/2/0.75	1.27	b	-	-	-
4/3/0.5	1.19	b	-	-	-
4/4/0.25	1.11	b	-	-	-
4/0/1	1.88	Non- resuspendable	-	-	-

13 PDI: Polydispersity index; ^a A clear solution was obtained; ^b Precipitation or gel-formation was
 14 observed.

15 **Table S2.** Physicochemical properties of unexpected CS_{70/20} NPs (mean ± S.D., *n* = 3)

Mass ratio CS/CD/TPP (w/w/w)	Charge ratio (+/-)	Size (nm)	PDI	ζ potential (mV)	Production yield (%)
4/1/0	6.25	a	-	-	-
4/2/0	3.12	a	-	-	-
4/5/0	1.25	b	-	-	-
4/1.5/1	1.46	b	-	-	-
4/2/0.75	1.53	b	-	-	-
4/3/0.5	1.42	b	-	-	-
4/4/0.25	1.33	b	-	-	-
4/0/1.25	1.80	b	-	-	-
4/0/1.5	1.50	b	-	-	-
4/0/2	1.13	b	-	-	-
4/1/1	1.66	b	-	-	-
4/1/0.5	2.62	a	-	-	-
4/2/0.25	2.32	Non- resuspendable	-	-	-

16 PDI: Polydispersity index; ^a A clear solution was obtained; ^b Precipitation or gel-formation was
17 observed.

18 **Table S3.** The effect of Captisol® concentration on the solubility of quercetin, encapsulation efficiency
19 (EE) and the final loading of quercetin into CS_{70/5} nanoparticles (mean ± SD, *n* =3). Control values:
20 4/0/0.75 NPs

Mass ratio CS/CD/TPP (w/w/w)	Charge ratio (+/-)	Captisol® conc. (mg/mL)	Quer. Solubility conc. (mg/mL)	Association efficiency (%)	Loading efficiency (%)
4/0/0.75	2.5	0	0.0095 ± 0.0004	95.38 ± 0.67	0.50 ± 0.0335
4/0/0.5	3.75	0	0.0095 ± 0.0004	96.80 ± 0.76	0.55 ± 0.0236
4/1/0.5	2.18	0.375	0.0219 ± 0.0013	93.20 ± 1.36	0.98 ± 0.0624
4/2/0.25	1.93	0.75	0.0394 ± 0.0009	90.45 ± 1.30	1.49 ± 0.0252

21 **Table S4.** The effect of Captisol® concentration on the solubility of quercetin, encapsulation efficiency
22 (EE) and the final loading of quercetin into CS_{70/20} nanoparticles (mean ± SD, *n* =3). Control values:
23 4/0/1 NPs

Mass ratio CS/CD/TPP (w/w/w)	Charge ratio (+/-)	Captisol® conc. (mg/mL)	Quer. Solubility conc. (mg/mL)	Association efficiency (%)	Loading efficiency (%)
4/0/1	2.25	0	0.0083 ± 0.0004	67.29 ± 3.90	0.30 ± 0.0159
4/0/0.75	3	0	0.0083 ± 0.0004	71.43 ± 2.43	0.33 ± 0.0112
4/3/0	2.08	1.125	0.0646 ± 0.0051	91.82 ± 2.07	2.20 ± 0.1078
4/4/0	1.56	1.5	0.0787 ± 0.0018	94.69 ± 0.33	2.43 ± 0.0084
4/1/0.75	2.03	0.375	0.0219 ± 0.0013	85.42 ± 2.28	0.86 ± 0.0230
4/2/0.5	1.85	0.75	0.0394 ± 0.0009	85.17 ± 3.25	1.35 ± 0.0404

24 **Table S5.** Influences of different formulations on the reduction of bacterial growth (OD₆₀₀) and
25 relative QS activities as compared to the positive control

Free components	Captiso 10.1875 mg/mL	Captiso 10.375 mg/mL	Captiso 10.75 mg/mL	CS _{70/5} 0.75 mg/mL	CS _{70/20} 0.75 mg/mL	Querceti n 0.0125 mg/mL	Quercetin 0.025 mg/mL	Quercetin 0.0375 mg/mL
OD reduction	-2.09%	0.48%	1.18%	21.08 %	88.67 %	25.38%	29.06%	33.34%
QS reduction	8.43%	17.53%	20.86%	27.08 %	65.79 %	-54.95%	-35.57%	-20.97%
CS _{70/20} NPs	U 4/0/1	U 4/2/0.5	U 4/3/0	U 4/4/0	L 4/0/1	L 4/2/0.5	L 4/3/0	L 4/4/0
OD reduction	30.10%	71.33%	44.64%	63.34 %	39.48 %	81.96%	60.03%	88.32%
QS reduction	43.36%	69.27%	64.08%	78.57 %	51.46 %	72.93%	78.43%	100.00%
CS _{70/5} NPs	U 4/0/0.75	U 4/1/0.5	U 4/2/0.25	L 4/0/0.75	L 4/1/0.5	L 4/2/0.25	U: unloaded L: loaded NPs: nanoparticles	
OD reduction	21.80%	22.72%	21.14%	25.18 %	30.33 %	34.71%		
QS reduction	30.90%	38.45%	38.07%	38.18 %	52.32 %	61.12%		



27

4/0/0.5

4/0/0.75

4/1/0.5

4/2/0.25

28

Figure S1. Unloaded CS_{70/5} nanoparticles

29



30

4/3/0

4/4/0

4/0/1

4/2/0.5

4/1/0.75

31

Figure S2. Unloaded CS_{70/20} nanoparticles

32