Strains/plasmid	**Characteristics**	**Reference**
E. coli FT1/pLysS	BL21(DE3) Δ (*ptsHIcrr*)/pLys, KmR, Cm^R	Parche *et al*., 1999
Plasmids		
p4813	Amp^R, *ptsK*	Reizer *et al*., 1998
pWH466	pET3c, *ptsH* (*B. subtilis*)	Seidel *et al*., 2005
pWH653	pET3c*, ccpA(his)$_6$* (*B. subtilis)*	Seidel *et al*., 2005

Oligonucleotide	**Sequence (5' → 3')**	**Modification**	**Reference**
ackA_cre2_fw	TTCTTATTGTAAGCGTTATCAATACG	5' biotinylated	Schumacher *et al*., 2010
ackA_cre2_rev	CGTATTGATAACGCTTACAATAAGAA		Schumacher *et al.*, 2010
gntR_cre_fw	GTCTGATTGAAAGCGGTACCATTTTA	5' biotinylated	Schumacher *et al.*, 2010
gntR_cre_rev	TAAAATGGTACCGCTTTCAATCAGAC		Schumacher *et al.*, 2010
syn_cre_fw	TTCTTACTGTTAGCGCTTTCAGTACG	5' biotinylated	Schumacher *et al.*, 2010
syn_cre_rev	CGTACTGAAAGCGCTAACAGTAAGAA		Schumacher *et al.*, 2010
non-specific DNAfw	AATCATTTATGGCATAGGCAACAAGT	5' biotinylated	Seidel *et al.*, 2005
non-specific DNArev	ACTTGTTGCCTATGCCATAAATGATT		Seidel *et al.*, 2005

References:

Reizer,J., Hoischen,C., Titgemeyer,F., Rivolta,C., Rabus,R., Stülke,J., Karamata,D., Saier,M.H. Jr and Hillen,W. (1998) A novel protein kinase that controls carbon catabolite repression in bacteria. *Mol. Microbiol.,* **27**, 1157–1169.

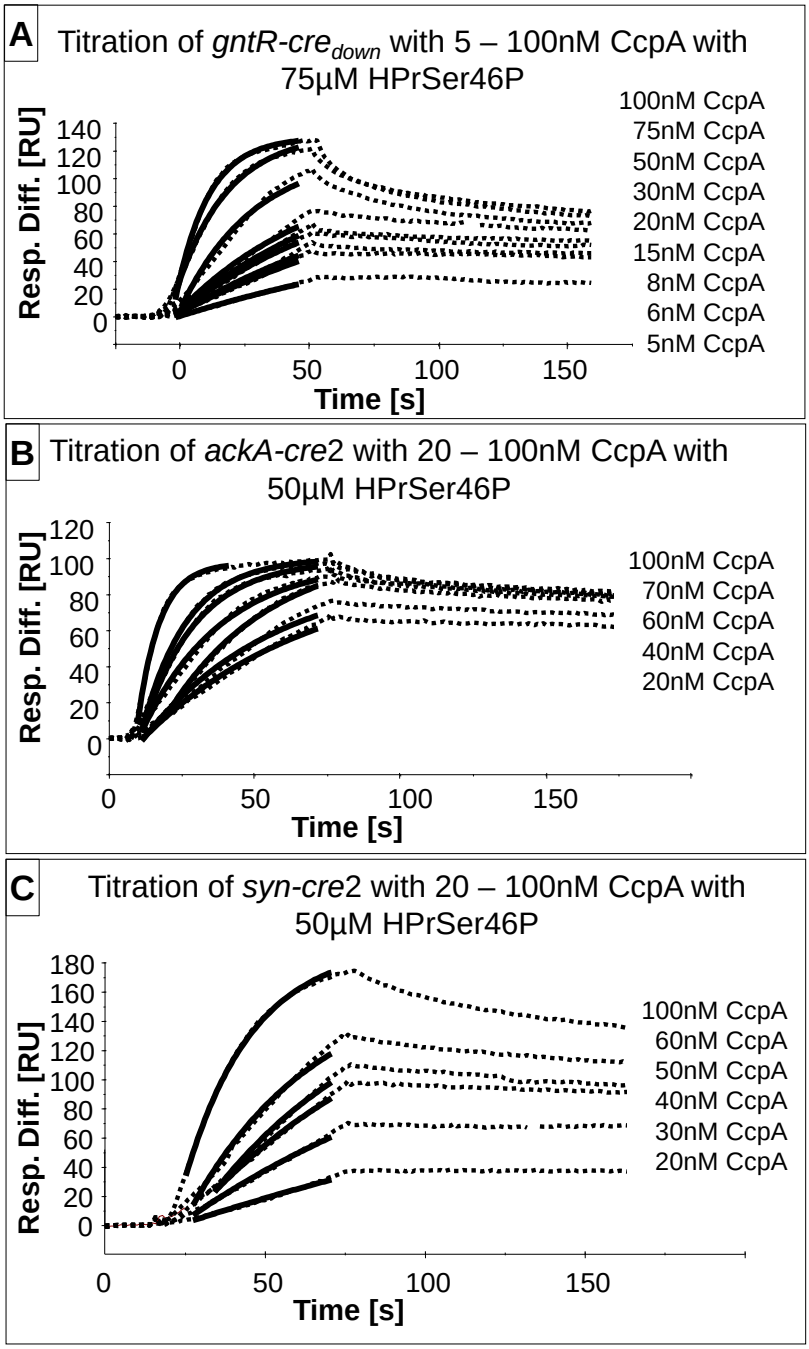
Parche,S., Schmid,R. and Titgemeyer,F. (1999) The phosphotransferase system (PTS) of *Streptomyces coelicolor* identification and biochemical analysis of a histidine phosphocarrier protein HPr encoded by the gene *ptsH.Eur. J. Biochem.,* **265**, 308–317.

Schumacher, M.A., Sprehe, M., Bartholomae M., Hillen, W., Brennan, R.G., Structures of carbon catabolite protein A–(HPr-Ser46-P) bound to diverse catabolite response element sites reveal the basis for high-affinity binding to degenerate DNA operators *Advance access publishes November 23, 2010 doi:10.1093/nar/gkq1177*

Seidel,G., Diel,M., Fuchsbauer,N. and Willen,W. (2005) Quantitative interdependence of coeffectors, CcpA and *cre* incarbon catabolite regulation of *Bacillus subtilis. FEBS J*., **272**,2566–2577.

Data:

Kinetic analysis of CcpA-HPrSer46P binding to *cre*-elements by SPR. The titrations were carried out with 5-100nM CcpA $(His)_6$. The running buffer was supplemented with 75µM HPrSer46P in the case of *gntR-cre*$_{down}$ (A) and with 50µM HPrSer46P in the case of *ackA-cre*2 and *syn-cre* (B and C) because under these conditions CcpA is completely bound by HPrSer46P (Seidel et al, 2005). Dashed lines represent the measured data, bold lines the best fits for the association reactions. The Table shows association and dissociation rate constants calculated from the sensorgramms.



	k_a [1/Ms]	k_d [1/M]	K_D [M]	χ^2
ackA cre2	$6.0 \pm 1.9 \times 10^{5}$	$9.5 \pm 1.0 \times 10^{-4}$	$1.6 \pm 0.4 \times 10^{-9}$	3.5-4.4
gntR cre$_{down}$	$6.0 \pm 1.7 \times 10^{5}$	$1.8 \pm 0.3 \times 10^{-3}$	$3.0 \pm 0.4 \times 10^{-9}$	3.4-5.2
syn *cre*	$3.2 \pm 0.6 \times 10^{5}$	$1.2 \pm 0.2 \times 10^{-3}$	$3.8 \pm 0.1 \times 10^{-9}$	5.8