Corrigendum

A cyclic GMP-dependent signalling pathway regulates bacterial phytopathogenesis

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Since the publication of this article, the authors have noticed several errors that in any case do not affect the original conclusions presented. The authors apologize for any inconvenience caused.

The gel presented in panel A of Figure 2 suggests a slightly incorrect size for the purified CYC domain of XC_250. Panel E in the same figure incorrectly describes D28 as D41 (see below). The correct figure and legend are shown below. Source data for this figure is now available on the online supplementary information page.

The predicted critical amino acid sites of XC_0250 were reported incorrectly in the section ‘The cyclase domain of XC_0250 is active in cyclic GMP synthesis’, with D28 described as D41. The text should have read:

‘The two critical metal-ion binding aspartates are conserved (D28 and D71) as well as an alanine (A150) residue that occupies a substrate-specifying position. However, the transition state-stabilizing asparagine and arginine residues are substituted by leucine (L157) and alanine (A161). The importance of both conserved and altered residues (D28, D41, D71, L73, A150, L157, A161) for the enzymatic activity of this domain was examined by assessing the effects of alanine or serine substitutions’.

And at a later point in the same section: ‘Several of these residues (D28, D71, A150, L157) are conserved in the *R. centenum* guanylyl cyclase (Supplementary Fig S1).’

Consistent with this, the alignment in Supplementary Fig S1 panel A incorrectly shows the position of D28. The correct figure and legend are available below.

Figure 2 The isolated CYC domain of XC_0250 possesses guanylyl cyclase activity. (A) SDS-PAGE of the CYCHis6 protein purified by nickel affinity chromatography showed a single band of the expected size. Image shows spliced lanes from the SDS/PAGE gel. Lane 1: protein marker; lanes 2, 3: total protein; lanes 4, 5: samples purified by the Ni column method. The purified CYC domain from XC_0250 had enzymatic activity against GTP. (B) Reverse-phase HPLC separation of the reaction mixtures and guanine nucleotide standards GMP, cyclic GMP and GTP showed the synthesis of a compound with the same mobility as the cyclic GMP standard. (C and D). The CYC domain with mutation D28 or D71 variations lost enzymatic activity (E and F). All enzymatic reactions contained 0.5 mM GTP, 10 mM MnCl_2 and $0.5 \mu\text{g} \mu\text{l}^{-1}$ (0.025 nM) purified protein. Aliquots of reaction mixtures were boiled immediately after addition of the enzyme (time, 0 min) and after 60 min of incubation (time, 60 min). The identity of the product was confirmed by mass spectrometry. Source data for the gel shown in panel A is now available on the online supplementary information page.

Supplementary Figure 1. (A). Amino acid sequence alignment of the cyclase domain of XC_0250 with domains of known cyclases. Key: Rv0891C, Rv1264, Rv1625c, Rv1900c and Rv2488c from *Mycobacterium tuberculosis*, 1WC0_A: *Streptomyces platensis*; 1CUL_A: Mammalian (dog); 1FX2_A: *Trypanosoma brucei*; 3R5G_B: *Pseudomonas aeruginosa* are adenyl cyclases. Guanylylcyclases are 2W01_A: *Synechocystis* sp. PCC 6803 Cya2 (NP_440289); 3UVJ_A: Homo sapiens 3UVJ_A (NP_004954); 2WZ1_A: Homo sapiens 2WZ1_A (NP_004954); 3ET6_A: *Chlamydomonas reinhardtii* CYG12 (XP_001700847); RC1-3783: *Rhodospirillum rubrum* (YP_002299938). XC_0250: *Xanthomonas campestris*. Amino acids predicted to be involved in the formation of the active-site or metal binding are boxed in red while residues that were mutated in XC_0250 are indicated by arrows (D28, D41, D71, L73, A150, R151, L157). The alignment was done using MEGA v 4.0 (B). Contribution of XC_0250 to cyclic GMP levels in Xcc. Deletion of XC_0250 leads to a reduced level of cyclic GMP in Xcc that can be restored to wild-type levels by complementation with a clone (pXC_0250) expressing the full-length XC_0250 protein. Alanine or serine substitutions in residues predicted to be critical for enzymatic activity and other residues of XC_0250 abolish restoration (D28A, D41A, D71A, L73A, A150S, R151A, L157A). Values given are the mean and standard deviation of triplicate measurements (three biological and three technical replicates). (C) The guanylatecyclase activity of XC_0250 is required for biofilm formation. Variants in the CYC domain of XC_0250 that can no longer function as a guanylatecyclase (D28A, D71A) are unable to restore biofilm formation to the XC_0250 mutant. Values given are the mean and standard deviation of triplicate measurements (three biological and three technical replicates).