Corrigendum

Structural analysis of a non-ribosomal halogenated cyclic peptide and its putative operon from *Microcystis*: implication for evolution of cyanopeptolins

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Due to a mistake during data processing, the putative substrate-binding pocket sequences of two of the adenylation domains were switched. Consequently, the structure of the peptide (cyanopeptolin-984) produced by *Microcystis* NIVA-CYA 172/5 (N-C 172/5) shown in Fig. 1 is incorrect in the positioning of two amino acid residues. The underlying experimental data and the molecular mass of cyanopeptolin-984 were, however, correct. Fig. 1 showing the corrected peptide structure and corrected Tables 1 and 2 are shown below. It should be noted that the phylogenetic tree of adenylation domain sequences (Fig. 3) is correct, but the designations for McnC1-Ile/Thr and McnE-Gln/Ile should have read McnC1-Gln/Thr and McnE-Ile, respectively. This correction makes interpretation of the phylogenetic tree more straightforward, and has no impact on the major conclusions of the paper.

Fig. 1. (a) Molecular structure of cyanopeptolin-984 as deduced from LC/MS/MS fragmentation patterns. (b) Predicted structure of the peptide synthesized by the peptide synthetase from *Microcystis* N-C 172/5. R positions according to Namikoshi & Rinehart (1996).

References

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Table 1. LC/MS/MS fragmentation pattern of cyanopeptolin-984 produced by *Microcystis* N-C 172/5

Collision energy	Fragment mass (Da)	Fragment(s)
30 eV	984.4	$M + H^+$
	966.4	$M + H^+ - H_2O$
	797.3	$M + H^+ - \text{side chain [Ile-ClMeTyr-Phe-Ahp-Gln-Thr-NH}_3 + H^+]$
	779.3	$M + H^+ - \text{side chain} - H_2O$ [Ile-ClMeTyr-Phe-Ahp-Gln-Thr-NH ₃ -H ₂ O + H ⁺]
	454.1	Ahp[ring]-Phe-ClMeTyr-NH ₂
40 eV	254.1	Ac-Gln-NH-C ₃ (⁺)H ₄ =C=O [Ac-Gln-Thr-H ₂ O + H ⁺]
	243.1	Ac-Gln-NH-C(⁺)H-CONH ₂
	198.0	NH ₂ -ClMeTyr-CH ₃ NH
	159.1	OH-C(⁺)H-Ahp
50 eV	—	No additional fragments
60 eV	342.1	ClMeTyr-Phe + H ⁺ - NH ₃
	326.2	Ile-Thr-Gln-NH ₂ + H ⁺ - 2NH ₃
	195.1	NH ₂ CO-CH ₂ -CH=CH-CO-NH-C ₃ (⁺)H ₄ =C=O [from Gln-Thr]
	183.1	Ahp[ring]-C(⁺)H-CO-NH-CH ₃
	127.0	ClTyr ring
	114.1	Ahp ring
	101.1	N(⁺)H=CH-CH ₂ -CH ₂ -CONH ₂ [Gln-CO + H ⁺]
	97.1	CH ₃ -CH ₂ -CHCH ₃ -C(⁺)=C=O [from Ile]
	84.1	CH ₃ -C(⁺)H-CN ₂ =C=O [from Thr]
	72.0	Ac-NH-C(⁺)H ₂
	70.0	Ac-N(⁺)CH
30 eV	984.4	$M + H^+$
	966.4	$M + H^+ - H_2O$
	797.3	$M + H^+ - \text{side chain [Ile-ClMeTyr-Phe-Ahp-Gln-Thr-NH}_3 + H^+]$
	779.3	$M + H^+ - \text{side chain} - H_2O$ [Ile-ClMeTyr-Phe-Ahp-Gln-Thr-NH ₃ -H ₂ O + H ⁺]
	454.1	Ahp[ring]-Phe-ClMeTyr-NH ₂
40 eV	254.1	Ac-Gln-NH-C ₃ (⁺)H ₄ =C=O [Ac-Gln-Thr-H ₂ O + H ⁺]
	243.1	Ac-Gln-NH-C(⁺)H-CONH ₂
	198.0	NH ₂ -ClMeTyr-CH ₃ NH
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50 eV	—	No additional fragments
60 eV	342.1	ClMeTyr-Phe + H ⁺ - NH ₃
	326.2	Ile-Thr-Gln-NH ₂ + H ⁺ - 2NH ₃
	195.1	NH ₂ CO-CH ₂ -CH=CH-CO-NH-C ₃ (⁺)H ₄ =C=O

Table 2. Deduced functions of encoded proteins in the *mcn* gene cluster of *Microcystis* N-C 172/5

Protein	Length (aa)	Proposed domains*	Putative substrate-binding pocket†	Putative substrate‡	Putative substrate§
McxA	1098				
Module 1		C, A _{Gln} , T	DAWQTGLIDK	Gln	Gln
McxB	1105/1101				
Module 2		C, A _{Thr} , T	DFWNIGMVHK	Thr	Thr
McxC	4721				
Module 3		C, A _{Gln} , T	DAQDLGVVDK	Gln	Gln/Thr
Module 4		C, A _{Ahp} , T	DVENAGVVTK	?	Ahp precursor
Module 5		C, A _{Phe} , T	DAWTIAAVCK	Phe	Phe
Module 6		C, A _{Tyr} , M, T	DASTIAGVCK	Tyr	Tyr
McxD	625	Halogenase			
McxE	1418/1387				
Module 7		C, A _{Ile} , T, TE	DAFFLGVTFK	Ile	Ile

*The domains encoded by the genes are indicated as follows: C, condensation; A, adenylation; T, thiolation (peptidyl carrier); M, *N*-methyltransferase; TE, thioesterase.

†The binding pocket sequences were identified by aligning the A-domains of Mcn with the Phe activating A-domain of GrsA (Conti *et al.*, 1997).

‡Substrate as predicted from the specificity-inferring code of adenylation domains (Challis *et al.*, 2000; Stachelhaus *et al.*, 1999). The Ahp precursor was deduced from extensive similarity with the Ahp precursor activating domain described by Rouhiainen *et al.* (2000), including the identical binding pocket residues.

§Substrate as predicted by sequence alignments and phylogenetic analyses.