

Figure S1. Macroscopic and microscopic characteristics of other strains of cyanobacteria investigated in this study. *Desmonostoc lechangense* CSM 014-B (a–f). (a) Blue-green colony on culture plate. (b–e) Filaments with narrow elongated apical cells (AC) enclosed by thick sheath (S). (e) Less granulated, colorless cells. *Desmonostoc aggregatum* CSM 022-B (g–j). (g) Bright-green colony on culture plate. (h) Aggregated cells in microcolony. (i,j) Vegetative cells mostly spherical with conical-rounded terminal heterocytes (TH). *Dendronalium* sp. CSM 025-N (k–o). (k) Brown colony on culture plate. (l–n) Bent and slightly twisted filaments with spherical to barrel-shaped vegetative cells. (o) Akinetes (A) produced in chain. Scale bar: 10 µm

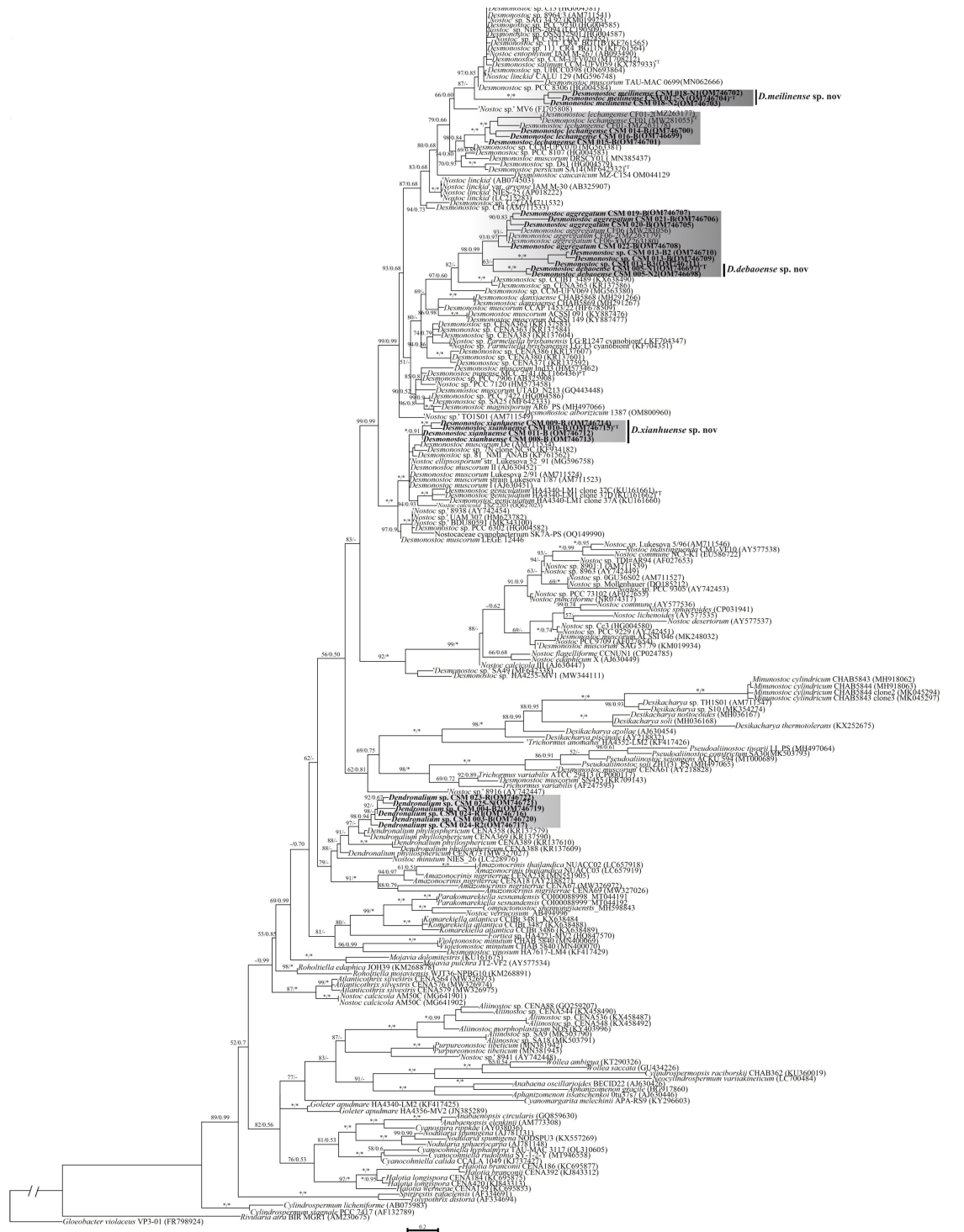


Figure S2. Uncollapsed Maximum Likelihood (ML) tree based on 16S rRNA gene sequences showing the phylogenetic positions of cyanobacterial strains under investigation and closely related taxa (224 16S rRNA gene sequences). Numbers on nodes indicate bootstrap values (>50%) and posterior(>0.50) probability obtained from ML and Bayesian analyses, respectively. An asterisk (*) was used to denote bootstrap values of 100% for ML and posterior probabilities of 1.0 for Bayesian analysis, on the same node. The investigated are in bold font and highlighted in grey. The scale corresponds to substitutions/site.

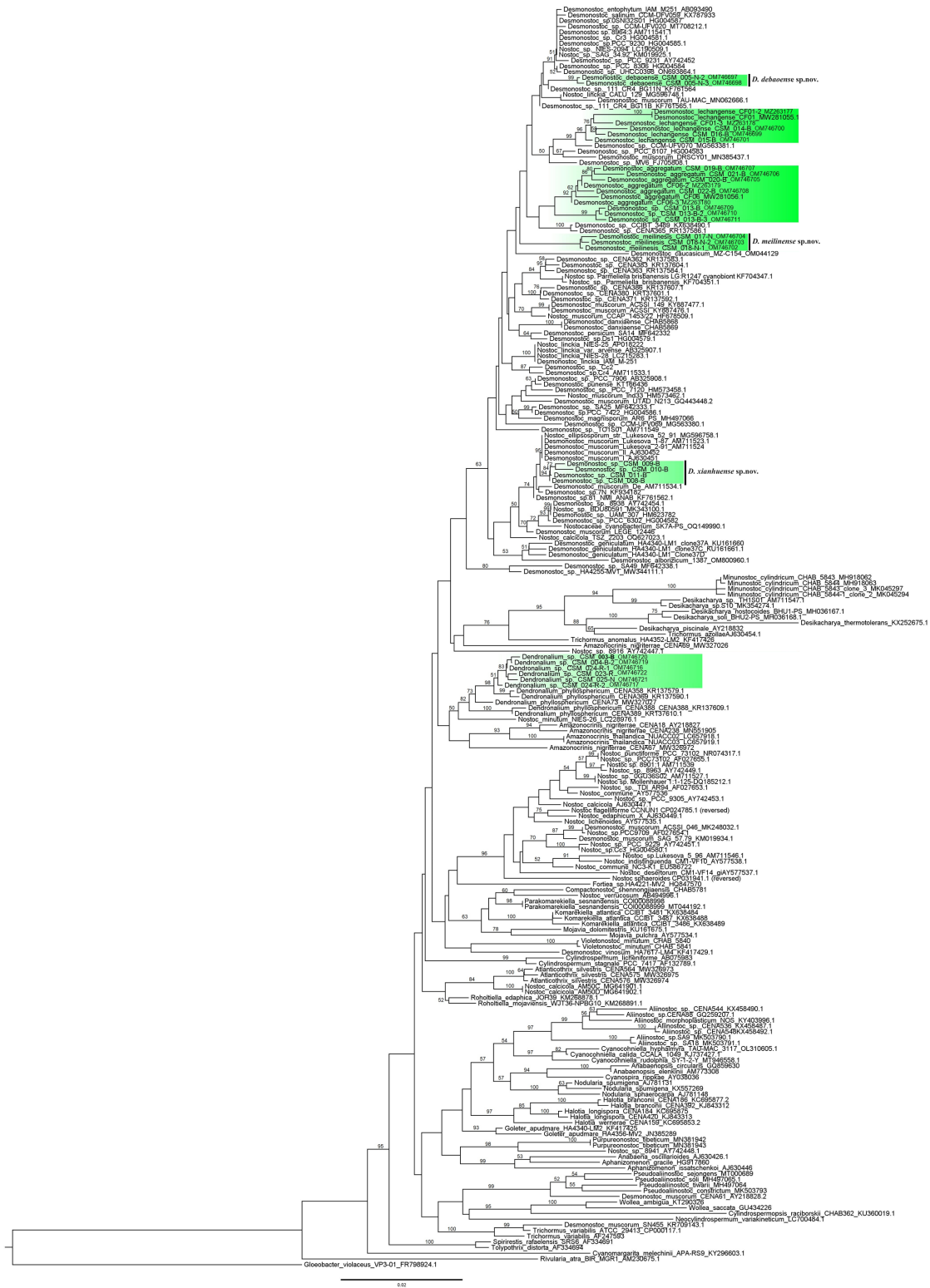


Figure S3. Neighbor-joining (NJ) tree based on 16S rRNA gene sequences showing the phylogenetic positions of the twenty cyanobacterial strains recovered from this study and other cyanobacterial taxa (224 16S rRNA gene sequences). Numbers at nodes indicate bootstrap values (>50%). The investigated strains in this study are highlighted and the novel species are marked with a star beside the name.

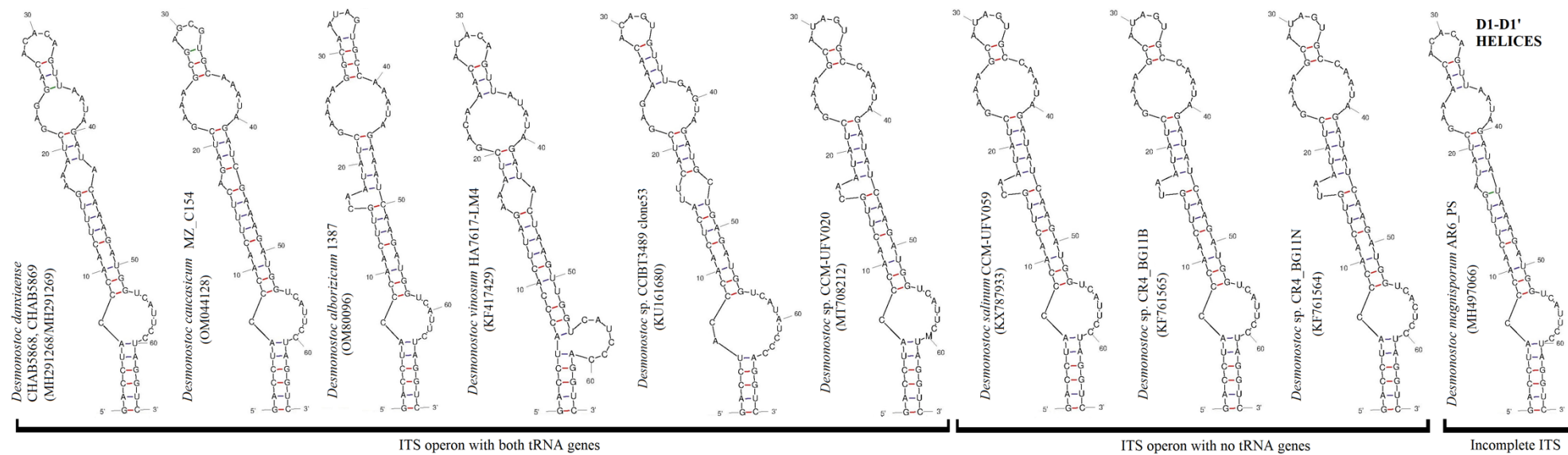


Figure S4. Predicted secondary structures of conserved D1-D1' helices of the 16S-23S ITS region among *Desmonostoc* strains.

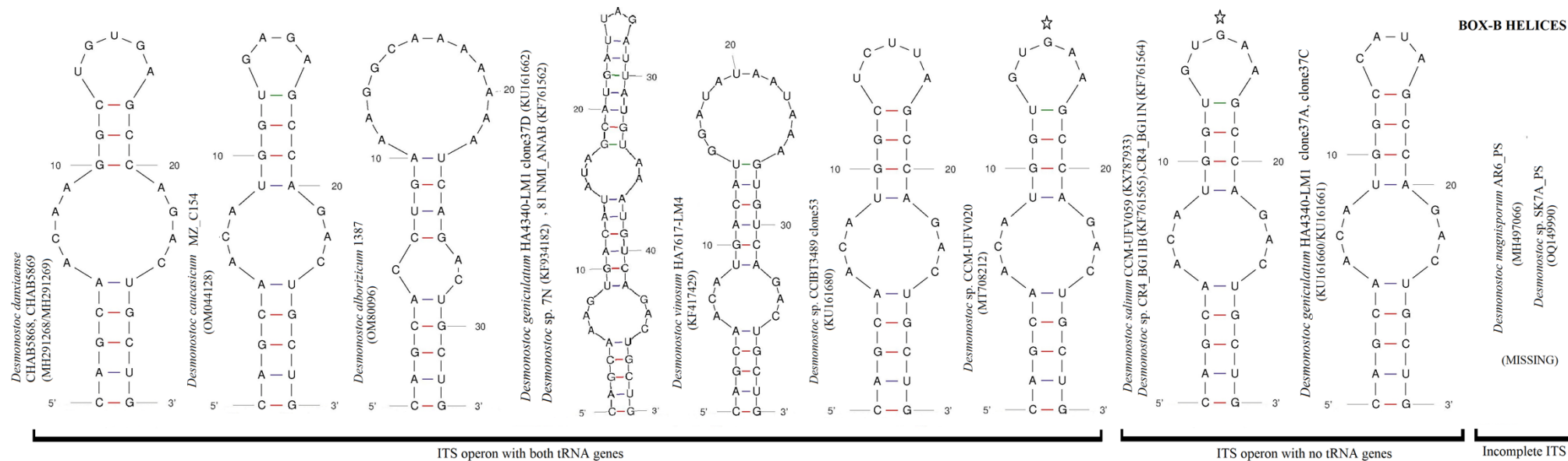


Figure S5. Predicted secondary structures of conserved Box-B helices of the 16S–23S ITS region among *Desmonostoc* strains. A star denotes identical in structure.

V3 HELICES

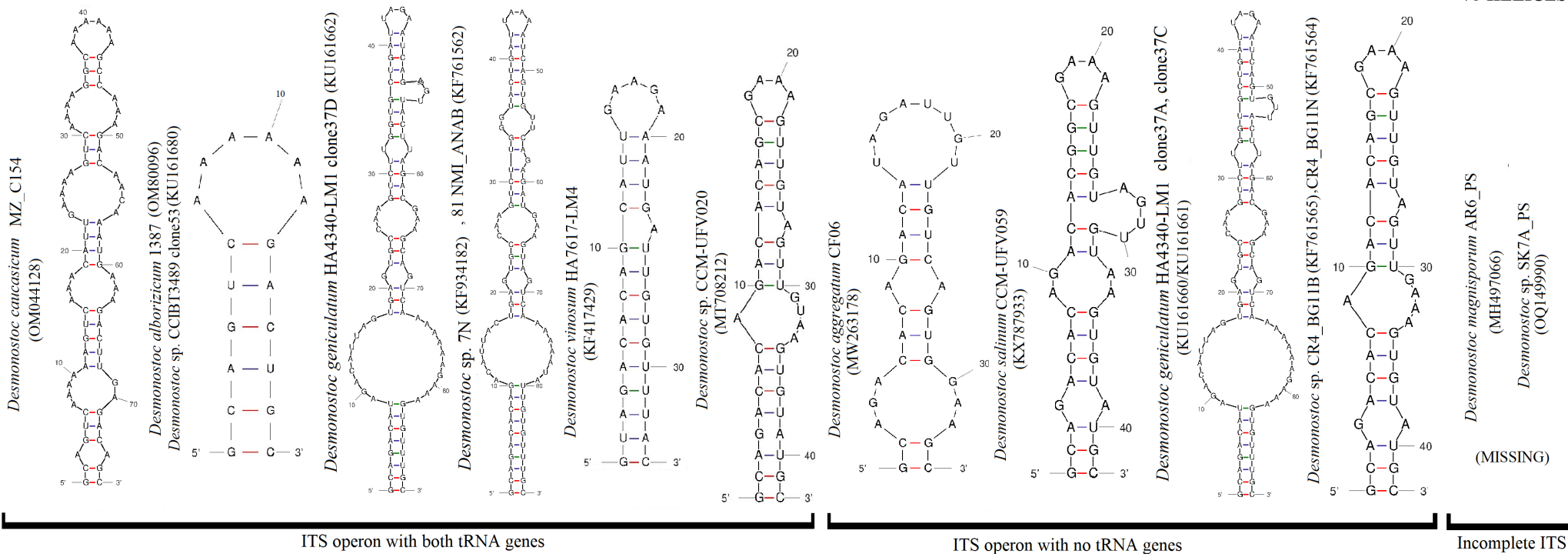


Figure S6. Predicted secondary structures of conserved V3 helices of the 16S–23S ITS region among *Desmonostoc* strains.

Table S1. List of the 20 cyanobacterial strains examined in this study.

No	Strain	Host	¹ Locality	Host habitat type	GenBank Accession number (16S rRNA gene)	Living Collection Code
1	CSM 005-N	<i>Cycas debaoensis</i>	a	natural habitat	OM746697, OM746698	FLBG-CSM 005
2	CSM 014-B	<i>Cycas elongata</i>	d	botanic garden	OM746700	FLBG-CSM 014
3	CSM016-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746699	FLBG-CSM 016
4	CSM015-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746701	FLBG-CSM 015
5	CSM 017-N	<i>Cycas fairylakea</i>	c	natural habitat	OM746704	FLBG-CSM 017
6	CSM 018-N	<i>Cycas fairylakea</i>	c	natural habitat	OM746702, OM746703	FLBG-CSM 018
7	CSM 019-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746707	FLBG-CSM 019
8	CSM 020-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746705	FLBG-CSM 020
9	CSM 021-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746706	FLBG-CSM 021
10	CSM 022-B	<i>Ceratozamia robusta</i>	d	botanic garden	OM746708	FLBG-CSM 022
11	CSM 013-B	<i>Macrozamia moorei</i>	d	botanic garden	OM746709, OM746710, OM746711	FLBG-CSM 013
12	CSM 011-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746712	FLBG-CSM 011
13	CSM 008-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746713	FLBG-CSM 008
14	CSM 009-B	<i>Ceratozamia robusta</i>	d	botanic garden	OM746714	FLBG-CSM 009
15	CSM 010-B	<i>Ceratozamia robusta</i>	d	botanic garden	OM746715	FLBG-CSM 010
16	CSM 003-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746720	FLBG-CSM 003
17	CSM 004-B	<i>Cycas fairylakea</i>	d	botanic garden	OM746718, OM746719	FLBG-CSM 004
18	CSM 023-R	<i>Cycas debaoensis</i>	b	reintroduction site	OM746722	FLBG-CSM 023
19	CSM 024-R	<i>Cycas debaoensis</i>	b	reintroduction site	OM746716, OM746717	FLBG-CSM 024
20	CSM 025-N	<i>Cycas fairylakea</i>	c	natural habitat	OM746721	FLBG-CSM 025

¹Host locality/GPS coordinates: a= Debao County, Baise,Guangxi ; 23°29'29.2" N, 106°12'50.8" E

b= Huanglian mountain nature reserve, Debao County, Baise, Guangxi; 23°33'47.1" N, 106°14'09.8" E

c= Meilin Reservoir Park, Shenzhen, Guangdong; 22°34'38.5" N, 114°00'34.2" E

d= National Cycad Germplasm Conservation Center (NCGCC), Fairy Lake Botanical Garden (FLBG), Shenzhen, Guangdong; 22°34'36.4" N, 114°10'50.6" E

Table S2. Comparison of the 16S rRNA gene sequence similarity between *Desmonostoc* strains under investigation and closely related taxa. The intraspecies variation are highlighted in grey and similarity for the interspecies variation were averaged for species represented by multiple strains.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
1 <i>Desmonostoc debaoense</i>																			
2 <i>Desmonostoc meilinense</i>	97.3	99.4																	
3 <i>Desmonostoc xianhuense</i>	97.9	97.5	99.8																
4 <i>Desmonostoc</i> sp. CSM 013-B	97.8	97.2	97.0																
5 <i>Desmonostoc aggregatum</i>	97.9	97.2	97.2	97.7	99.0														
6 <i>Desmonostoc lechangense</i>	97.3	97.7	96.4	96.8	96.9	98.9													
7 <i>Desmonostoc danxiaense</i> CHAB5868	97.3	98.0	97.0	97.6	96.8	97.3													
8 <i>Desmonostoc muscorum</i> Lukesova 1/87	97.5	96.8	99.6	96.8	96.9	96.2	97.6												
9 <i>Desmonostoc alborizicum</i> 1387	96.4	96.6	96.2	96.0	95.8	95.8	96.8	96.5											
10 <i>Desmonostoc geniculatum</i> HA4340-LM1	97.3	97.1	99.0	96.8	96.9	96.1	97.6	99.2	85.0										
11 <i>Desmonostoc magnisporum</i> AR6_PS	97.5	97.8	97.3	98.0	97.4	97.4	98.5	97.9	95.1	95.5									
12 <i>Desmonostoc persicum</i> SA14	97.4	97.3	97.0	97.7	97.3	97.7	98.6	97.5	94.2	94.8	98.2								
13 <i>Desmonostoc punense</i> MCC 2741	97.4	97.6	97.8	97.3	97.2	96.9	98.3	98.3	97.3	98.1	99.1	98.3							
14 <i>Desmonostoc caucasicum</i> MZ-C154	96.4	96.7	95.9	95.9	95.9	96.4	97.5	96.5	95.3	96.3	97.0	97.6	97.1						
15 <i>Desmonostoc salinum</i> CCM-UFV059	98.3	97.7	97.2	97.4	97.3	97.4	98.0	97.7	88.6	86.4	96.2	96.2	98.0	97.3					
16 <i>Desmonostoc vinosum</i> HA7617-LM4	95.3	95.8	96.3	95.5	94.4	95.0	96.0	96.7	95.8	96.1	96.3	95.9	95.9	95.2	95.9				
17 <i>Desmonostoc</i> sp. 111 CR4_BG11B	98.8	98.3	97.5	97.7	97.5	98.0	98.7	97.9	88.9	88.2	95.8	96.1	98.2	97.8	91.6	96.0			
18 <i>Desmonostoc</i> sp. 7N clone_NC3C	97.6	97.3	99.5	96.9	96.9	96.2	97.8	99.7	84.7	96.7	95.2	94.8	97.7	96.6	86.6	96.6	88.5		
19 <i>Desmonostoc</i> sp. 81-NMI_ANAB	97.7	97.5	99.7	97.1	97.1	96.4	97.9	99.9	84.8	96.8	95.4	95.0	97.9	96.8	86.7	96.6	88.6	99.8	
20 <i>Desmonostoc</i> sp. CCIBT 3489	98.3	97.1	97.4	97.7	97.8	96.9	98.0	97.9	94.8	84.4	95.8	95.8	98.2	97.0	88.1	95.9	87.6	84.5	84.6

Table S3. Comparison of the 16S rRNA gene sequence similarity between *Desmonostoc* strains under investigation and species of other genera in Nostocaceae.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
1 <i>Desmonostoc debaoense</i>															
2 <i>Desmonostoc meilinense</i>	97.3														
3 <i>Desmonostoc xianhuense</i>	97.9	97.5													
4 <i>Desmonostoc</i> sp. CSM 013-B	97.8	97.2	97.0												
5 <i>Desmonostoc aggregatum</i>	97.9	97.2	97.2	97.7											
6 <i>Desmonostoc lechangense</i>	97.3	97.7	96.4	96.8	96.9										
7 <i>Dendronalium phyllosphericum</i> CENA369	96.7	95.7	96.1	95.8	95.6	95.7									
8 <i>Nostoc commune</i> EV1-KK1	94.8	94.8	95.4	95.2	94.8	94.7	95.3								
9 <i>Nostoc punctiforme</i> PCC73102	94.7	94.1	94.5	94.8	94.4	93.8	95.4	99.1							
10 <i>Mojavia pulchra</i> JT2-VF2	94.6	94.5	94.9	94.6	94.4	94.4	96.2	95.5	95.8						
11 <i>Desikacharya soli</i> BHU2-PS	93.8	93.5	94.1	94.0	93.4	94.0	94.8	94.9	94.4	93.2					
12 <i>Komarekiella atlantica</i> CCIBt 3483	94.7	94.5	95.3	94.8	94.3	94.9	96.5	95.7	94.8	96.6	82.9				
13 <i>Aliinostoc morphoplasticum</i> NOS	93.4	93.0	93.3	93.3	92.5	93.9	94.3	93.1	93.0	93.7	88.3	85.2			
14 <i>Nostoc minutum</i> NIES-26	96.3	95.7	95.9	95.9	96.0	95.7	98.3	96.0	95.9	96.0	95.0	95.9	94.8		
15 <i>Violetonostoc minutum</i> CHAB5841	94.7	94.4	94.6	94.8	94.1	95.0	96.4	95.0	94.6	95.0	94.6	96.6	93.3	96.0	
16 <i>Amazonocrinis nigriterrae</i> CENA18	95.1	95.1	95.3	95.7	95.1	95.1	97.2	95.7	95.7	96.1	83.7	89.0	84.6	97.5	95.9

Table S4. Comparison of the 16S rRNA gene sequence similarity between *Dendronalium* strains under investigation and closely related taxa.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
1 <i>Dendronalium</i> sp. CSM 003-B																		
2 <i>Dendronalium</i> sp. CSM 004-B1	99.0																	
3 <i>Dendronalium</i> sp. CSM 023-R	99.2	98.8																
4 <i>Dendronalium</i> sp. CSM 024-R1	99.9	99.9	99.9															
5 <i>Dendronalium</i> sp. CSM 025-N	99.1	98.7	99.6	99.8														
6 <i>D. phyllosphericum</i> CENA369	99.0	98.8	98.9	99.6	98.7													
7 <i>D. phyllosphericum</i> CENA73	98.6	98.4	98.6	99.2	98.3	99.1												
8 <i>Dendronalium</i> sp. NP-KLS-5C-PS	99.0	98.7	99.2	99.4	98.9	99.5	99.2											
9 <i>D. phyllosphericum</i> CENA389	98.0	98.0	97.9	98.8	97.8	98.4	98.7	98.2										
10 <i>D. phyllosphericum</i> CENA358	99.1	98.8	99.0	99.7	98.8	99.8	99.2	99.6	98.5									
11 <i>Nostoc minutum</i> NIES-26	97.6	97.4	97.5	98.3	97.4	98.3	98.5	98.4	97.9	98.4								
12 <i>Nostoc commune</i> EV1-KK1	94.9	95.0	95.2	95.7	95.2	95.3	95.6	95.5	95.7	95.4	96.0							
13 <i>Desmonostoc debaoense</i>	97.1	96.9	97.2	97.7	96.9	96.9	97.0	96.6	96.8	97.0	96.5	94.8						
14 <i>Mojavia pulchra</i> JT2-VF2	96.2	96.2	96.4	96.8	96.4	96.2	96.7	96.4	96.5	96.3	96.0	95.5	94.9					
15 <i>Desikacharya soli</i> BHU2-PS	93.9	93.8	93.8	94.6	93.6	94.8	94.8	83.8	95.0	94.8	95.1	95.0	94.4	93.2				
16 <i>Komarekiella atlantica</i> CCIBt	96.0	96.0	96.0	97.1	95.9	96.5	97.0	80.4	96.2	96.5	95.9	95.7	95.5	96.6	85.4			
17 <i>Aliinostoc morphoplasticum</i>	93.8	94.0	94.0	94.9	93.8	94.3	94.5	83.7	94.8	94.4	94.9	93.2	93.6	93.7	88.4	86.8		
18 <i>Violetonostoc minutum</i>	95.6	95.7	95.5	96.4	95.4	96.4	96.5	96.9	96.2	96.5	95.8	95.0	94.7	94.9	94.7	96.6	94.1	
20 <i>Amazonocrinis nigriterrae</i> CENA18	96.7	96.8	96.7	97.7	96.6	97.2	97.7	79.3	97.3	97.3	97.5	95.7	95.5	96.1	84.5	87.4	84.9	95.8

Table S5. Percent dissimilarity for the 16S-23S ITS region for *Dendronalium* strains.

	1	2	3	4	5
1 <i>Dendronalium</i> sp. CSM 003-B					
2 <i>Dendronalium</i> sp. CSM 004-B	6.6				
3 <i>Dendronalium</i> sp. CSM 023-R	1.0	6.9			
4 <i>Dendronalium</i> sp. CSM 024-R	1.0	6.6	0.3		
5 <i>Dendronalium</i> sp. CSM 025-N	1.4	6.6	0.2	0.2	
5 <i>Dendronalium phyllosphericum</i> CENA369	10.6	4.3	10.6	10.6	10.6

Table S6. Analyses of the 16S-23S ITS region among *Desmonostoc* strains for which ITS sequences are complete. The strains under investigation are in bold font. Cyanobacterial taxa with ITS operon which does not contain tRNA genes are highlighted in yellow. The proposed three novel species are marked with an asterisk (*) beside the name.

Strains	ITS Total length (nt)	Leader	D1-D1' Helix	Spacer + D2	Spacer + D3 + Spacer	tRNAIle	Spacer + V2 Helix	tRNAAla	Post tRNA Spacer	Box-B Helix	Spacer + Box A	D4 + Spacer	V3 Helix	D5 (end region of ITS)	Accession No.
<i>D. debaoense</i> CSM 005-N *	530	9	65	34	16	74	93	73	36	29	28	21	31	21	OM746734/ OM746735
<i>D. debaoense</i> CSM 005-N clone1/ clone2 *	276	9	65	31	41	-	-	-	-	29	28	21	31	21	OR723793/ OR723794
<i>D. meilinense</i> CSM 017-N/ CSM 018-N *	508	9	65	33	14	74	81	73	37	29	28	21	34	10	OM746739/ OM746738
<i>D. meilinense</i> CSM 017-N clone1/clone2 *	258	8	65	31	29	-	-	-	-	33	28	21	31	12	OR723795/ OR723796
<i>D. xianhuense</i> CSM 008-B/009-B/010-B *	608/609	9	65	35	16	74	79	73	38	54	28	25	90/91	22	OM746747/ OM746748/ OM746749
<i>D. xianhuense</i> CSM 010-B clone1/clone2 *	335	9	65	31	37	-	-	-	-	28	28	25	90	22	OR723797/ OR723798
<i>Desmonostoc</i> sp. CSM 013-B	508	9	65	33	15	74	85	73	37	32	26	21	34	4	OM746744
<i>Desmonostoc</i> sp. CSM 013-B clone 1/clone2	254	8	65	31	33	-	-	-	-	32	26	21	34	4	OR723799/ OR723800
<i>D. aggregatum</i> CSM 019B/022B/021B	510	9	65	33	14	74	80	73	39	29	28	21	34	11	OM746742 / OM746743/ OM746741
<i>D. aggregatum</i> CSM 020-B/CF06	262	9	65	31	34	-	-	-	-	29	28	21	34	11	OM746740/ MW263178

Table S6. Cont.

Strains	ITS Total length (nt)	Leader	D1-D1' Helix	Spacer + D2	Spacer + D3 + Spacer	tRNAIle	Spacer + V2 Helix	tRNAAla	Post tRNA Spacer	Box-B Helix	Spacer + Box A	D4 + Spacer	V3 Helix	D5 (end region of ITS)	Accession No.
<i>D. lechangense</i> CSM 014-B/ CF01	520	9	65	33	14	74	85	73	46	29	28	21	31	12	OM746737/ MW263177
<i>D. lechangense</i> CSM 016-B	263	9	65	31	36	-	-	-	-	29	29	21	31	12	OM746736
<i>D. danxiaense</i> CHAB5868/CHAB5869	514	7	65	33	16	74	85	73	38	29	29	21	34	10	MH291268/ MH291269
<i>D. alborizicum</i> 1387	527	9	69	32	15	74	83	73	36	33	29	25	18	31	OM800960
<i>D. salinum</i> CCM-UFV059	275	9	66	32	37	-	-	-	-	29	28	21	42	11	KX787933
<i>D. caucasicum</i> MZ-C154	571	9	66	34	15	74	82	71	40	28	28	25	76	23	OM044128
<i>D. geniculatum</i> HA4340-LM1 clone37D	605	9	65	35	16	74	81	73	37	50	28	25	90	22	KU161662
<i>D. geniculatum</i> HA4340-LM1 clone 37A/37C	344	9	65	31	46	-	-	-	-	28	28	25	90	22	KU161660/ KU161661
<i>Desmonostoc</i> sp. CCIBT3489 clone53	525	9	69	34	15	74	83	73	36	29	29	25	18	31	KU161680
<i>D. vinosum</i> HA7617-LM4	577	9	65	36	16	74	94	73	16	40	29	27	34	64	KF417429
<i>Desmonostoc</i> sp. 7N/NMI_ANAB	603	9	65	35	16	74	79	73	37	50	28	25	89	23	KF934182/ KF761562
<i>Desmonostoc</i> sp. 111 CR4 BG11B/BG11N	274	9	66	31	37	-	-	-	-	29	28	21	42	11	
<i>Desmonostoc</i> sp. CCSM-UFV020	526	9	66	34	14	74	85	73	40	29	28	21	42	11	MT708212

Table S7. Analyses of the 16S-23S ITS region among *Dendronalium* strains. The strains under investigation are in bold font.

Strains	ITS Total length (nt)	Leader	D1-D1' Helix	Spacer + D2	Spacer + D3 + Spacer	tRNAIle	Spacer + V2 Helix	tRNAAla	Post tRNA Spacer	Box-B Helix	Spacer + Box A	D4 + Spacer	V3 Helix	D5 (end region of ITS)	Accession No.
<i>Dendronalium</i> sp. CSM 003-B	534	8	64	34	14	74	80	73	28	46	29	25	36	23	OM746754/OM746755
<i>Dendronalium</i> sp. CSM 004-B	540	8	64	34	14	74	86	73	28	46	29	25	36	23	OM746752/OM746753
<i>Dendronalium</i> sp. CSM 023-R	534	8	64	34	14	74	80	73	28	46	29	25	36	23	OM746750/OM746751
<i>Dendronalium</i> sp. CSM 024-R	534	8	64	34	14	74	80	73	28	46	29	25	36	23	OM746757
<i>Dendronalium</i> sp. CSM 025-N	534	8	64	34	14	74	80	73	28	46	29	25	36	23	OM746756
<i>D. phyllosphericum</i> CENA369	540	8	65	34	14	74	86	73	28	35	29	25	36	23	NZ_JAECZA000000000.1

