

Supplementary Materials

Table S1. Primers used in this study.

Primer	Sequence (5' to 3')
RNC1F	GGCTGCACATATCAGATCCTTG
RNC1R	ACTTCTATGTCTTCAAACCCTCTG
RNC2F	AGGCGATTATGTCTTACCTGATT
RNC2R	TAAACTTTGTTGAGGCTGTCTACC
GTC1F	GAATTTGAATTGCGTAATAGTGC
GTC1R	AACTTAAACGACGGTTAACGC
GTC2F	GAAAAACAAGGCGTTATCGGT
GTC2R	CCGTGAGGTAATTCCTGCAT
AFSP1	CACTCGTGACCACCAGATTGCCA
AFSP2	GGAGAACATTACACGCTCTACTGG
AFSP3	ACGGGAGTTGAGTAGTTCGTTAGTG
ARSP1	GTTTGATGTTGCTGACGTGGCTTTA
ARSP2	TGGTAATAACCGCGACTTGGCAG
ARSP3	CGACTGTCAGCATTTATGCCTCTAG
GRSP1	ACAAAGCATTGACATGGGTGGAC
GRSP2	TTCTATTCTCTTGCTCCGCACCG
GRSP3	TTATGGGTATCAGTGAAAAGGCGA
HRSP1	AGATGAACTGCTGACTCGCCGTA
HRSP2	TCCTCGCACTATTGCCGCTTTTA
HRSP3	CCTGGTTTATCCGTCCGATTTATG

Note: Primers AFSP1-3, ARSP1-3, GRSP1-3 and HRSP1-3 were used in the genome walking experiments.

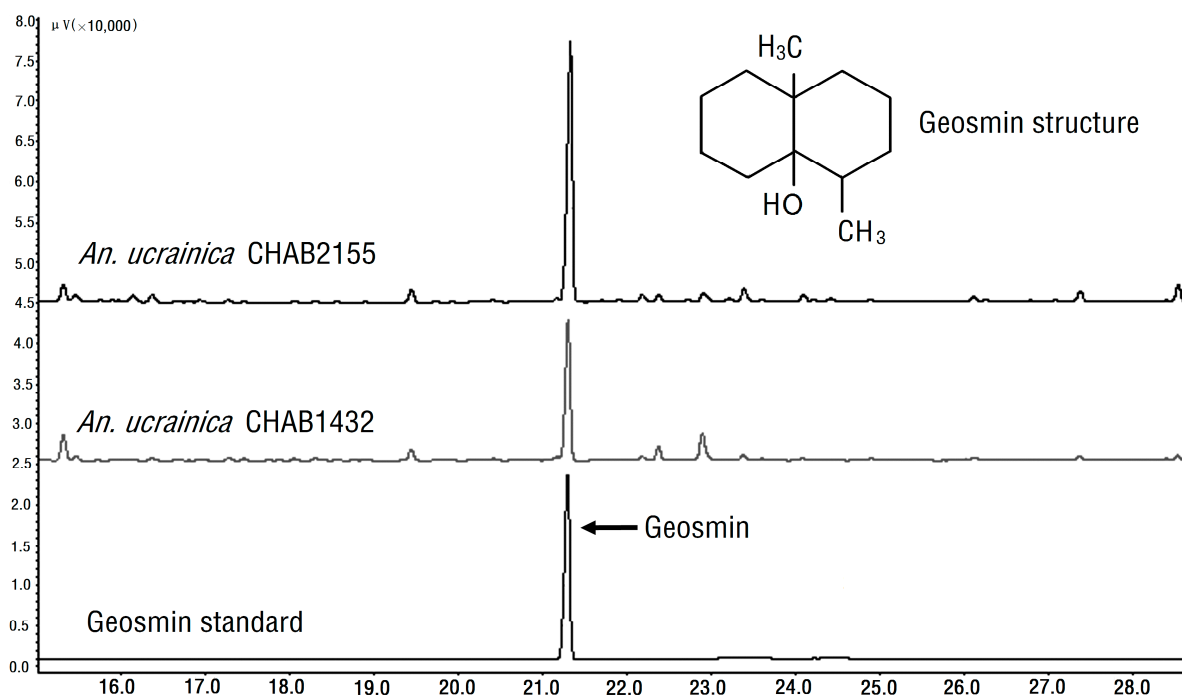


Figure S1. Geosmin detected chromatograms for cultures of *Anabaena ucrainica* CHAB 1432 and 2155. The x and y axes are retention time (min) and abundance (μV) respectively.

					Primer GSMif	
Geo1	1	--MQPFKLPAYMPWPARLNPNEAARVHSHKAWAYEMGILGSKEESQGEPIWDERKFDADHYALLCSYTHPDTPSTELNVLVDWYVWVFFDDHFLIYK	98			
Geo2	1	--MQPFKLPAYMPWPARLNPNEAARVHSHKAWAYEMGILGSKEESQGEPIWDERKFDADHYALLCSYTHPDTPSTELNVLVDWYVWVFFDDHFLIYK	98			
GeoA1	1	--MQPFKLPDFYMPWPARLNPNEAARVHSHKAWAYEMGILGSKEEANGEPIDWDERKFDADHYALLCAYTHPDAPATELDTITDQYVWVFFDDHFLIYK	98			
GeoA2	1	--MQPFKLPDFYMPWPARLNPNEAARVHSHKAWAYDMGILGSKEEANGEPIDWDERKFDADHYALLCAYTHPDTPGTELDLITDQYVWVFFDDHFLIYK	98			
NPUNMOD	1	--MQPFELPEFYMPWPARLNPNEAARSHSKAWAYQMGILGSKEEASSVWDERTFDAHDYALLCSYTHPDAPGTELDLVDWYVWVFFDDHFLIYK	98			
Sav2163	1	--MTQPFQLPHFYMPYPARLNPHEAARSHSTRWARGMGL-----GSGIWEQSDLAHDYGLLCAYTHPDGDPALSLITDQYVWVFFDDHFLIYK	93			
Sco6073	1	MTQPFQLPHFYLPARLNPHEAARSHSTRWARGMGL-----GSGVWEQSDLEAHDYGLLCAYTHPDGDPALSLITDQYVWVFFDDHFLIYK	94			
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Geo1	99	RSQDLIGAKEYLDRLPAFMPIYQDNLFPFNPVERGLADLWSRTAFIKSVWRQRFESTKNNLDESMWELANINQNRANPIEYIEMRRKVGAPWSA	198			
Geo2	99	RSQDLIGAKEYLDRLPAFMPIYQDNLFPFNPVERGLADLWSRTAFIKSVWRQRFESTKNNLDESMWELANINQNRANPIEYIEMRRKVGAPWSA	198			
GeoA1	99	RSQDMIGAKAYLDRLPAFMPIYQDNLFPFNPVERGLADLWSRTAFIKSVWRQRFESTKNNLDESMWELANINQNRANPIEYIEMRRKVGAPWSA	198			
GeoA2	99	RSQDMIGAKAYLDRLPAFMPIYQDNLFPFNPVERGLADLWSRTAFIKSVWRQRFESTKNNLDESMWELANINQNRANPIEYIEMRRKVGAPWSA	198			
NPUNMOD	99	RTQDMAGAKAYLDRLPAFMPIYQDNLFPFNPVERGLADLWSRTAFIKSVWRQRFESTKNNLDESMWELANINQNRANPIEYIEMRRKVGAPWSA	198			
Sav2163	94	RTQDREGGKAYLDRLPAFMPIYQDNLFPFNPVERGLADLWSRTAFIKSVWRQRFESTKNNLDESMWELANINQNRANPIEYIEMRRKVGAPWSA	193			
Sco6073	95	RSQDLIGAKEYLDRLPAFMPIYQDNLFPFNPVERGLADLWSRTAFIKSVWRQRFESTKNNLDESMWELANINQNRANPIEYIEMRRKVGAPWSA	194			
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Geo1	199	DLVEHAFAVEVPKAAATRPMPVLKDTFADGVHLRNDLFSYQREVEEENGNSNCVLVVERFLNVSTQEAANLTNELLNSRLYQFDMTAVTELPFLFEEYG	298			
Geo2	199	DLVEHAFAVEVPKAAATRPMPVLKDTFADGVHLRNDLFSYQREVEEENGNSNCVLVVERFLNVSTQEAANLTNELLNSRLYQFDMTAVTELPFLFEEYG	298			
GeoA1	199	DLVEHAFAVEVPKAAATRPMPVLKDTFADGVHLRNDLFSYQREVEEENGNSNCVLVVERFLNVSTQEAANLTNELLNSRLYQFDMTAVTELPFLFEEYG	298			
GeoA2	199	DLVEHAFAVEVPKAAATRPMPVLKDTFADGVHLRNDLFSYQREVEEENGNSNCVLVVERFLNVSTQEAANLTNELLNSRLYQFDMTAVTELPFLFEEYG	298			
NPUNMOD	199	DLVEHAFAVEVPKAAATRPMPVLKDTFADGVHLRNDLFSYQREVEEENGNSNCVLVVERFLNVSTQEAANLTNELLNSRLYQFDMTAVTELPFLFEEYG	298			
Sav2163	194	GLVEYAT--AEVPAAGSRPLRVLMETFSGAVHLRNDLFSYQREVEEENGNSNCVLVVERFLNVSTQEAANLTNELLNSRLYQFDMTAVTELPFLFEEYG	292			
Sco6073	195	GLVEYAT--AEVPAAGSRPLRVLMETFSGAVHLRNDLFSYQREVEEENGNSNCVLVVERFLNVSTQEAANLTNELLNSRLYQFDMTAVTELPFLFEEYG	293			
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GSY1	299	VDPVERVNVLLYIKGLQDWQSGGHEWHMSSRYMNMKEPDNSGTSVTLGGPTGLGTSAAARLESLS--TTLGLRRFKSFTHVPYQTVGPVKLPKFIYMPFSTT	397			
GSY2	299	VDPVERVNVLLYIKGLQDWQSGGHEWHMSSRYMNMKEPDNSGTSVTLGGPTGLGTSAAARLESLS--TTLGLRRFKSFTHVPYQTVGPVKLPKFIYMPFSTT	397			
GeoA1	299	IDPVEQANVLLYIKGLQDWQSGGHEWHMSSRYMNMKEPDNSGTSVTLGGPTGLGTSAAARLESLS--TTLGLRRFKSFTHVPYQTVGPVKLPKFIYMPFSTT	397			
GeoA2	299	VDPVERVNVLLYIKGLQDWQSGGHEWHMSSRYMNMKEPDNSGTSVTLGGPTGLGTSAAARLESLS--TTLGLRRFKSFTHVPYQTVGPVKLPKFIYMPFSTT	397			
NPUNMOD	299	LDPVARVNVLLYIKGLQDWQSGGHEWHMSSRYMNMKEPDNSGTSVTLGGPTGLGTSAAARLESLS--TTLGLRRFKSFTHVPYQTVGPVKLPKFIYMPFSTT	396			
Sav2163	293	LTPPEVAAAYARGLDQWQSGGHEWHMSSRYMNMKEPDNSGTSVTLGGPTGLGTSAAARLESLS--TTLGLRRFKSFTHVPYQTVGPVKLPKFIYMPFSTT	388			
Sco6073	294	LTPPEVAAAYARGLDQWQSGGHEWHMSSRYMNMKEPDNSGTSVTLGGPTGLGTSAAARLESLS--TTLGLRRFKSFTHVPYQTVGPVKLPKFIYMPFSTT	387			
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Geo1	398	LNPMLDAARNSHKEWARRMGLSLEIPDAFIWVDHFKFDVADVALCGAWIHPNGSEHELNLTAACWLWVGYAADDYFPAIYGNRDLACAGVFNARLSAF	497			
Geo2	398	LNPMLDAARNSHKEWARRMGLSLEIPDAFIWVDHFKFDVADVALCGAWIHPNGSEHELNLTAACWLWVGYAADDYFPAIYGNRDLACAGVFNARLSAF	497			
GeoA1	399	VNPHLEAVRLHKAIAIANGMLSPGEDSLNLGIWDERKFDLMMNLAFASVTNPDLITIQLEIADWCVWVFFDDYFPAIYGNRDLACAGVFNARLSAF	497			
GeoA2	398	LNPMLDAARNSHKEWARRMGLSLEIPDAFIWVDHFKFDVADVALCGAWIHPNGSEHELNLTAACWLWVGYAADDYFPAIYGNRDLACAGVFNARLSAF	497			
NPUNMOD	397	LNPMLDAARNSHKEWARRMGLSLEIPDAFIWVDHFKFDVADVALCGAWIHPNGSEHELNLTAACWLWVGYAADDYFPAIYGNRDLACAGVFNARLSAF	496			
Sav2163	388	LSPHLEGARARLTTPWMSIGMLQEG-----VWDEKLTATYDLPCLCAGLDPAATFDELDSLRLWAGTYGDDYFPAIYGNRDLACAGVFNARLSAF	480			
Sco6073	389	LSPHLEGARARLTTPWMSIGMLQEG-----VWDEKLTATYDLPCLCAGLDPAATFDELDSLRLWAGTYGDDYFPAIYGNRDLACAGVFNARLSAF	481			
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Geo1	498	MPLDNTSTPP--VATNPVEKGLADIWSRTAGPMSSTARTEFRRAIQDMTDSVWVWELANQTNRIPIPDIDYIEMRRKTFGSDLTMSLSRLSQGGEIPMIIYYS	596			
Geo2	498	MPLDNTSTPP--VATNPVEKGLADIWSRTAGPMSSTARTEFRRAIQDMTDSVWVWELANQTNRIPIPDIDYIEMRRKTFGSDLTMSLSRLSQGGEIPMIIYYS	596			
GeoA1	499	MPVDLTSTPP--VATNPVEKGLADIWSRTAGPMSSTARTEFRRAIQDMTDSVWVWELANQTNRIPIPDIDYIEMRRKTFGSDLTMSLSRLSQGGEIPMIIYYS	597			
GeoA2	498	MPLDNTSTPP--VATNPVEKGLADIWSRTAGPMSSTARTEFRRAIQDMTDSVWVWELANQTNRIPIPDIDYIEMRRKTFGSDLTMSLSRLSQGGEIPMIIYYS	597			
NPUNMOD	497	MPLDNTSTPP--VATNPVEKGLADIWSRTAGPMSSTARTEFRRAIQDMTDSVWVWELANQTNRIPIPDIDYIEMRRKTFGSDLTMSLSRLSQGGEIPMIIYYS	596			
Sav2163	481	MPVDGEEVP--APVNGMERGLIDLWAIITTAEMTPDERTRFASVDVMTESVWVWELANQTNRIPIPDIDYIEMRRKTFGSDLTMSLSRLSQGGEIPMIIYYS	579			
Sco6073	482	MPLDNTSTPP--VATNPVEKGLADIWSRTAGPMSSTARTEFRRAIQDMTDSVWVWELANQTNRIPIPDIDYIEMRRKTFGSDLTMSLSRLSQGGEIPMIIYYS	580			
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Geo1	597	RPMSLENAAADFACFTNDIFSQKEIEFEIGEIHNCVLVQNFNLCDLLKAVEIVNNLMTARAQQFQHIIVETELPALFDDFNLKNTREKLLKYIEKLEQ	696			
Geo2	597	RPMSLENAAADFACFTNDIFSQKEIEFEIGEIHNCVLVQNFNLCDLLKAVEIVNNLMTARAQQFQHIIVETELPALFDDFNLKNTREKLLKYIEKLEQ	696			
GeoA1	598	RQIQSLNNITIDWIGLSNDIFSQKEIEFEIGEIHNCVLVQNFNLCDLLKAVEIVNNLMTARAQQFQHIIVETELPALFDDFNLKNTREKLLKYIEKLEQ	696			
GeoA2	598	RPMSLENAAADFACFTNDIFSQKEIEFEIGEIHNCVLVQNFNLCDLLKAVEIVNNLMTARAQQFQHIIVETELPALFDDFNLKNTREKLLKYIEKLEQ	697			
NPUNMOD	597	RPMSLENAAADFACFTNDIFSQKEIEFEIGEIHNCVLVQNFNLCDLLKAVEIVNNLMTARAQQFQHIIVETELPALFDDFNLKNTREKLLKYIEKLEQ	696			
Sav2163	580	GPVRSLENAADVYGLINDIFSQKEIEFEIGEIHNCVLVQNFNLCDLLKAVEIVNNLMTARAQQFQHIIVETELPALFDDFNLKNTREKLLKYIEKLEQ	679			
Sco6073	581	GPVRSLENAADVYGLINDIFSQKEIEFEIGEIHNCVLVQNFNLCDLLKAVEIVNNLMTARAQQFQHIIVETELPALFDDFNLKNTREKLLKYIEKLEQ	680			
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Geo1	697	WMCGVLKWHITVDRYKEFELRNSASPIVRLNGPTGFGTSAAHIRSLVGATNSVI----	751			
Geo2	697	WMCGVLKWHITVDRYKEFELRNSASPIVRLNGPTGFGTSAAHIRSLVGATNSVI----	751			
GeoA1	697	WLAGNVVWINKTRRYNDFFVPN-LPKAEPVVSSTPFLGNSAFKIGSLVGTSNPFVNR-	753			
GeoA2	698	WMCGVLKWHITVDRYKEFELRNSASPIVRLADTSKGFDTSLTGISFISKGNHVKNG-	755			
NPUNMOD	697	WMCGVLKWHITVDRYKEFELRNSASPIVRLADTSKGFDTSLTGISFISKGNHVKNG-	753			
Sav2163	680	WMSGLKWHQDCHRYG-----AADLARRAHGFVDRAPAPPTAWAAPVAR----	725			
Sco6073	681	WMAGILNVHNRNVPYK-----AEYLACRTHGFLDRIAPPPVPRSSPALTH-----	726			

Figure S2. Amino acid sequence alignment of geosmin synthases or putative geosmin synthases. Geo1 and Geo2, synthases from *Anabaena ucrainica* CHAB 1432 and 2155; GeoA1 and GeoA2, putative synthases from *Phormidium* sp. (EF619621); NPUNMOD, synthase from *Nostoc punctiforme* PCC 73102 (CP001037); Sav2163, synthase from *Streptomyces avermitilis* MA-4680 (BA000303); Sco6073, synthase from *Streptomyces coelicolor* A3(2) (AL939126). Black-boxed residues show the possible Mg²⁺-binding sites; sites marked by * and # at the bottom indicate putative substrate binding pockets and active site lid residues, respectively.

Table S2. The NCBI accession numbers of the sequences used in phylogenetic trees.

Species	geosmin/synthetase gene	16S rRNA
<i>A. ucrainica</i> CHAB 1432	HQ404996	GU197649
<i>A. ucrainica</i> CHAB 2155	HQ404997	GU197642
<i>Phormidium</i> sp.	EF619621	HF678514
<i>Nostoc punctiforme</i> PCC 73102	FJ010203	CP001037
<i>Oscillatoria</i> sp. PCC 6506	ZP07114089	AY768397
<i>Calothrix</i> sp. PCC 7507	CP003943	CP003943
<i>Cylindrospermum stagnale</i> PCC 7417	CP003642	CP003642
<i>Myxococcus xanthus</i> DK 1622	CP000113	-
<i>Gloeobacter violaceus</i> PCC 7421	-	BA000045

		Mg ²⁺ -binding motif
Geo N-ter	1	MQPFKLPAYMFWFARLNPMLFAARVHSAWYEMGILGSKKEESQCEPIWDERKFDADHYALLCSYTHFDTPSTELNVTQVYVWVFDDHFLIYKRS
Geo C-ter	382	VCPVKLPKFFYMPFSTTLNPNLDAARNHKSKEVAROMGMLSLPEIPDAFWNDHKFDVADVALCGAWIHPNGSEHELNLTACVLVWCTYADDYFPAIYGN
NPUNMOD N-ter	1	MQPFELPEFYMPFWFARLNPMLFAARSHSAWYQMGILGSKKEAESSVWDERTFDAHDYALLCSYTHFDAPGTEDLVTQVYVWVFDDHFLIYKRT
NPUNMOD C-ter	381	VCPVTLKFFYMPFITSLNPNLDAARKHKSKEVAROMGMLSLPGIPDAFWDDHKFDVADVALCGALHPNGSGLELNLTACVLVWCTYADDYFPAIYGN
NSP7120	1	MEKITFFNLYCPFFPERKNQYFVLDQYALQVLRFKLIDSES-----LYQRFSKAKFYLLTAGAYPHCQLEELKIANDVISVLFVWDDQCDISDLGK
AVA29413	1	MEKFTFFNLYCPFFPERKNQYFVLDQYALQVLRFKLIDSES-----LYQRFSKAKFYLLTAGAYPHCQLEELKIANDVISVLFVWDDQCDISDLGK
NP73102	1	INQLLCGLYCPFFSQTNKYVDVLEEYSLVLRFLNLANES-----AYKRCKSKFFFLAASAYPDSKFEEIKITHDVLSTVFIWDDQCDLSLKK
Geo N-ter	101	QDLICAREYLDRLPAFMPIYPODNLPPFTNPVERCLADLWSRTAFTKSVWEQRFESTKNLLDESMLVELANINQNRIANPTEYIEMRRKVGCAFWASDL
Geo C-ter	482	RDLACAKVFNARLSAFMPLDNDSTP-FVATNPVEKCLADIWSRTAGPMSSTARTEERRAIQDMTDSWVVELANQTQNRIPDPIDYIEMRRKTFCSDLTMSL
NPUNMOD N-ter	101	QDMACAREYLGRLPMFMPIYPTETPPVPNPVECLADLWSRTAFTKSVWDWLRFEESTKNLLDESMLVELANINQNRVANPTEYIEMRRKVGCAFWASDL
NPUNMOD C-ter	481	RNMACAKVFNARLSAFMPLDDSTPSEVPNPVECLADIWSRTAGPMSANARTQERRAIQDMTDSWVVELANQINRIPDPIDYIEMRRKTFCSDLTMSL
NSP7120	93	K----PELLKIWCNRLEELNGAELTADDDLGLFALRDINRRIINRGSIITFFHHQVVRNFEDYFHCIEEAAHNRVTVSIPDVEAYIKIRSANAAAALCLNL
AVA29413	93	K----PELLKIWCNRLEELNGAELTADDDLGLFALRDINRRIINRGSIITFFHHQVVRNFEDYFHCIEEAAHNRVTVSIPDVEAYIKIRSANAAAALCLNL
NP73102	93	Q----PEVLNNFHQRYLEELNGAELTADDTLFSHALDLRKRRLQASIKWFNYITSYLEDYFVCQVQETATRAKGIPLDLDYIMIRSSSVGVAVLAL
Geo N-ter	201	VEHAAPVEVFAKTAATFPMRVLKDTFADCVHLRNDLFSYQREVEDEGENSNCVLVVERFLNVSTQEAANLTNELLTSLRYQDNTAVTELESLREEYGV
Geo C-ter	581	SRLSQGCEIFMQIYYSRPMRSLNAAADFACFTNDIPSYQKEIEFEGCEIHCNVLVWQNFNLCDLLKAVEIVNNMTARAQQFOHIVETELPALDDFNLD
NPUNMOD N-ter	201	VEHAVFTEIPADIASTFPMRVLKDTFADCVHLRNDLFSYQREVEDEGENSNCVLVVERFLNVSTQEAANLTNELLTSLRYQDNTAVTELESLREEYGLD
NPUNMOD C-ter	581	SRLAQCSEIFQELIYRTRTMRSLDNSAADFACLTNDIPSYQKEIEFEGCEIHCNVLVWQNFNLCDLFPQAVEVVNNMTSRALQQLIVATELPVLRDDFDLD
NSP7120	189	IEFCDRVMIFYSRLRNHDTLKKLTQMTINILAWSNDIPSAPREIAN-GEVHNLVFWIHHHQIPLERKAMLAAMHQQEVENLVKLESQIT-----YFS
AVA29413	189	IEFCDRVMIFYSRLRNHDTLKKLTQMTINILAWSNDIPSAPREIAN-GEVHNLVFWIHHHQIPLERKAMLAAMHQQEVENLVKLESQIT-----YFS
NP73102	189	SEFCNQFIIPDVLNRNHLVKKLELITTDIIAWSNDIPSAREIAS-GEVHNLVFWIHHHQIPLERKATEQVVKIHNEEVHSLIKVESSLS-----FFS
Geo N-ter	301	PVERVNVLLYIKELQDWQSCGHEVHMRSRYMNKQEPDNGSTSV-----
Geo C-ter	681	KNTREKLLKYIEKLEQWMCVLRKWHKVDRYK-EFELRNSASPIVRLLN
NPUNMOD N-ter	301	PVARVNVLLYIKELQDWQSCGHEVHMRSRYMNKKG-DNSPTSTV----
NPUNMOD C-ter	681	ASTREKLLCHVKKLEQWMCVLRKHITVDRYK-EFELRNSLAG-RLLS
NSP7120	281	AEIDAETIKYISLHAWIRENLDWYAHSCRYQITEKLELMAS-----
AVA29413	281	AETDAETIKYISLHAWIRENLDWYAHSCRYQITEKLELLAS-----
NP73102	281	EELDVEITKYISLHAWIRENLDWYCYEYRHNLERLELTFEK-----

Figure S3. Domain alignment of sesquiterpene synthases. N- and C-terminal domains of Geo, NPUNMO (from *Nostoc punctiforme* PCC 73102; CP001037; which convert FPP to geosmin) and three single-domain sesquiterpene synthases (which convert FPP to other sesquiterpene) in cyanobacteria: NSP7120 (*Nostoc* sp. PCC 7120; BA000019), AVA29413 (*Anabaena variabilis* ATCC 29413; CP000117) and NP73102 (*Nostoc punctiforme* PCC 73102; CP001037).