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Developmentally regulated volatiles geosmin and 2-methylisoborneol attract a soil arthropod to *Streptomyces* bacteria promoting spore dispersal

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Supplementary Information for:

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Supplementary Discussion

Distribution of *geoA* in *Streptomyces* genomes

To examine the distribution of the gene encoding geosmin synthase (*geoA*) in *Streptomyces* species, we interrogated all the *Streptomyces* genome sequences marked “completed” in the NCBI databases (ftp://ftp.ncbi.nlm.nih.gov/genomes/genbank/bacteria/assembly_summary.txt, accessed in April 2018). The 109 genomes for which gene predictions are available were searched with BLASTP and the *geoA* sequence from *S. coelicolor* (SCO6073). All 109 genomes were found to contain a clear homologue of *geoA*. In one of the genomes (*Streptomyces* sp. CB01881), the gene was misannotated as giving rise to shorter protein, but a closer inspection of the nucleotide sequence showed that a full-length version of the *geoA* open reading frame was present. By using TBLASTN, a total of 122 *Streptomyces* genomes (include some lacking gene predictions) were searched. Of these, 120 contained *geoA*. Of the other two, the *Streptomyces* sp. S063 genome sequence is marked as having many frame-shifted proteins (https://www.ncbi.nlm.nih.gov/assembly/GCA_002832675.1/), and *Streptomyces xinghaiensis* S187⁶¹ was isolated from marine sediment and may have lost *geoA* in adapting to this environment. Overall, our survey shows that, with possible rare exceptions, essentially all streptomycetes encode a geosmin synthase.

Absence of genes encoding olfactory receptors (OR) and the OR co-receptor (ORCO) from springtail genomes

The two published Collembola genome sequences, those of *Orchesella cincta* and *Folsomia candida*^{62,63}, were interrogated for homologues of OR and Orco. The genomes and predicted proteomes of these two springtails were searched using Orco and OR sequences from *Drosophila melanogaster* (Orco, isoform A, NP_001097687.1; Orco, isoform B, NP_524235.2; OR 13a, Q9VXL0.2; OR 43a NP_523647.2; OR 74a, CBA13679.1); no homologues were identified.

Supplementary Table

Supplementary Table 1. | Strains, Plasmids and Oligonucleotide primers used in this study

Strains	Relevant genotype/comments	Source/reference
<i>S. venezuelae</i>		
NRRL B-65442	Wild-type	64
SV6	$\Delta whiG::apr$	65
SV7	$\Delta whiB::apr$	66
SV8	$\Delta whiH::apr$	This work
SV10	$\Delta whiI::apr$	67
SV11	$\Delta whiA::apr$	68
SV13	$\Delta bldM::apr$	67
SV8/pIJ6973	$\Delta whiH::apr$ attB Φ BT1::pIJ6973	This work
<i>S. coelicolor</i>		
M145	Prototrophic SCP1 ⁺ SCP2 ⁺	69
J2192	$\Delta geoA \Delta(mibA-mibB)::apr$	This work
J2400	$whiG::hyg$	70
J3003	$\Delta geoA$	71
<i>E. coli</i>		
ET12567/pUZ8002	<i>dam-13::Tn9 dcm-6 hsdM</i> pUZ8002	72
BW25113	$\Delta(araD-araB)567 \Delta(rhaD-rhaB)568 \Delta lacZ4787 (::rrnB-3) hsdR514 rph-1$	73,74
Plasmids		
pIJ773	Plasmid template for amplification of the <i>apr oriT</i> cassette for 'Redirect' PCR-targeting	71
pIJ790	Modified λ RED recombination plasmid [<i>oriR101</i>] [<i>repA101(ts)</i>] <i>araBp-gam-bet-exo</i>	71
pMS82	Plasmid cloning vector for the conjugal transfer of DNA from <i>E. coli</i> to <i>Streptomyces</i> spp. Integrates site-specifically at the Φ BT1 attachment site (Hyg ^R).	75
pIJ6793	pMS82 carrying sequence encoding WhiH-3xFLAG driven from <i>whiH</i> promoter.	This work
pIJ10770	Derivative of pMS82.	76
pIJ10646	<i>geoA</i> and <i>eshA-mibA-mibB</i> cloned in pIJ10770.	This work
Primers		
<i>mib</i> _DEL_F	CTCCGCCGCGAGCAAGGAGCCACCGATGCCCGACTCCGGGATTCCGGGG ATCCGTCGACC	
<i>mib</i> _DEL_R	GACGGGCGCATCGTGCTGGGACCACTCGTTAGCCGGGTGTAGGCTG GAGCTGCTTC	
<i>whiH</i> _DEL_F	CTCAAGGGTCAACGAAGCAACGCCGACAAAGGATGCGTGATTCCGGGGA TCCGTCGACC	
<i>whiH</i> _DEL_R	GCCCCGACTCCGCACTCCCGGCCACGGCCCGCGGATCATGTAGGCTG GAGCTGCTTC	
<i>geoA</i> _F	GGGCGGATATCGACGTGGTCACCGGGCTCG	
<i>geoA</i> _R	GGGCGGATATCTCGGTCAGCGCGTCAGTGCG	
<i>mib</i> _F	GGCGAAGCTTGGTCCAGTGATCCTCCACGGC	
<i>mib</i> _R	GCTGCCTAGGGTGCTGGGACCACTCGTTCAGC	

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