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1 ***Blastococcus atacamensis* sp. nov., a novel strain adapted to life in the Yungay core**
2 **region of the Atacama Desert**

3

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23

24 Running title: *Blastococcus atacamensis* sp. nov.

25

26 Subject category: New taxa: Actinobacteria

27

28 The GenBank accession numbers for the 16S rRNA gene and genome sequences of strain
29 P6^T are KX926540 and POQU00000000, respectively. The genome accession number of
30 *Blastococcus saxobsidens* DSM 44509^T is POQT00000000.

31

32 Abbreviations: A₂pm, diaminopimelic acid; ANI, average nucleotide identity; dDDH, digital
33 DNA–DNA hybridization; GGDC, Genome-to-Genome Distance Calculator; NJ, neighbour-
34 joining; ML, maximum-likelihood; MP, maximum-parsimony; MUSCLE, Multiple
35 Sequence Comparison by Log-Expectation; T3PKS, type III polyketide synthase.

36

37 Keywords: polyphasic taxonomy, stress and biosynthetic genes, whole-genome sequences

38

39 **Abstract**

40 A polyphasic study was undertaken to establish the taxonomic status of a *Blastococcus* strain
41 isolated from an extreme hyper-arid Atacama Desert soil. The isolate, strain P6^T, was found
42 to have chemotaxonomic and morphological properties consistent with its classification in
43 the genus *Blastococcus*. It was shown to form a well-supported branch in the *Blastococcus*
44 16S rRNA gene tree together with the type strains of *Blastococcus capsensis* and
45 *Blastococcus saxobsidens* and was distinguished from the latter, its closest phylogenetic
46 neighbour, by a broad range of phenotypic properties. The draft whole genome sequence of
47 isolate P6^T showed 83.6% average nucleotide identity, 83.0% average amino acid identity
48 and a digital DNA:DNA hybridisation value of 27.8% in comparison with the genome
49 sequence of *B. saxobsidens* DSM 44509^T, values consistent with its assignment to a separate
50 species. Based on these data it is proposed that isolate P6^T (NCIMB 15090^T = NRRL B-
51 65468^T) be assigned to the genus *Blastococcus* as *Blastococcus atacamensis* sp. nov.
52 Analysis of the whole genome sequence of *B. atacamensis* P6^T, with 3,778 open reading
53 frames and a genome size of 3.9 Mb showed the presence of genes and gene clusters that
54 encode for properties that reflect its adaptation to the extreme environmental conditions that
55 prevail in Atacama Desert soils.

56

57 The actinobacterial genus *Blastococcus* was proposed by Ahrens and Moll [1] and the
58 description subsequently emended by Urzì *et al.* [2], Lee [3] and Hezbri *et al.* [4],
59 respectively. The genus *Blastococcus* together with the genera *Cumulibacter* [5],
60 *Geodermatophilus* [6], *Klenkia* [7] and *Modestobacter* [8] belong to the family
61 *Geodermatophilaceae* [9, 10] of the order *Geodermatophilales* [11]. Members of all of these
62 taxa share genomic features, as exemplified by multiple copies of the *trwC* gene (conjugative

63 relaxase) [12], have modest growth requirements [13], show unusual resistance to oxidative
64 stress [14] and tend to be associated with arid biomes, such as desert and high altitude soils
65 and with the surfaces of ancient monuments and natural stones [2, 15, 16].

66 Blastococci form a well-supported clade in the *Geodermatophilaceae* 16S rRNA gene tree
67 [4, 17] and can be distinguished from members of other genera classified in this family using
68 a combination of phenotypic features [13]. They are Gram-stain positive, coccoid-shaped
69 bacteria that may be motile or non-motile and which may propagate by budding and multiple
70 fission; they have *meso*-A₂pm in the peptidoglycan, mainly unsaturated and iso-branched
71 fatty acids; and complex phospholipid profiles which may include diphosphatidylglycerol,
72 phosphatidylcholine, phosphatidylethanolamine and phosphatidylinositol [4, 18]. At the time
73 of writing the genus encompasses 5 validly named species, namely *Blastococcus aggregatus*
74 [1, 4, 19], the type species, *Blastococcus capsensis* [4], *Blastococcus endophyticus* [4, 17],
75 *Blastococcus jejunensis* [3, 4], and *Blastococcus saxobsidens* [4, 19], and one validly named
76 strain, *Blastococcus colisei* [20]. These bacteria were isolated from the Baltic Sea, an
77 archaeological Roman pool in Tunisia, the leaves of the medicinal Chinese plant
78 *Camptotheca acuminata*, beach sand off the coast of South Korea, a limestone sample from
79 a church in Malta and an archaeological amphitheatre, respectively, and can be distinguished
80 using a range of phenotypic properties [4]. In addition, “*Candidatus Blastococcus*
81 *massiliensis*” was identified, from a stool sample of a patient with anorexia nervosa [21].

82 In a continuation of our studies on actinobacterial diversity in Atacama Desert habitats,
83 several strains were recovered from an extreme hyper-arid soil that had colonial and
84 morphological properties typical of blastococci. One of these isolates, strain P6^T, was the
85 subject of a polyphasic taxonomic study which showed that it represents a new *Blastococcus*
86 species, for which the name *Blastococcus atacamensis* sp. nov. is proposed.

87

88 *Blastococcus* strains were isolated from an extreme hyper-arid soil sample in November 2011
89 from the Yungay core region of the Atacama Desert on the eastern flank of the Cerro Aguas
90 Blancas (24°06'18.6"S/70°01'55.6W) at 1,033 metres above sea level. One gram of the soil
91 sample was suspended in 4.0 ml of ¼ strength Ringer's solution (Oxoid, product No.
92 BO0332D), this suspension was shaken on a tumble shaker prior to heating at 55°C for six
93 minutes. Aliquots (100µl) of the 10⁻¹ and 10⁻² dilutions were spread, in triplicate, over GYM

94 *Streptomyces* (DSMZ medium No. 65) and *Geodermatophilus obscurus* media [22]
95 supplemented with nalidixic acid ($10\mu\text{g}\cdot\text{ml}^{-1}$), cycloheximide and nystatin (each at $25\mu\text{g}\cdot\text{ml}^{-1}$).
96 The isolation plates were dried for 15 minutes at room temperature before incubation, as
97 recommended by Vickers and Williams [23]. After incubation at 28°C for 2 weeks, the
98 presumptive *Blastococcus* isolates were counted and expressed as the number of colony
99 forming units (cfu) per gram dry weight soil.

100 Small numbers of strains growing on the isolation plates were assigned to the genus
101 *Blastococcus* as they formed characteristic small, circular, reddish pink colonies with entire
102 margins; the highest count, $3.7\cdot 10^3$ cfu/g dry weight soil, was recorded from the *G. obscurus*
103 medium plates and the corresponding number on the GYM *Streptomyces* medium plates was
104 $2.7\cdot 10^3$ cfu/g dry weight soil. A representative *Blastococcus* strain, isolate P6^T, was taken
105 from one of the GYM *Streptomyces* plates and along with the type strains of *Blastococcus*
106 species was maintained on GYM slopes at room temperature and as suspensions of cells in
107 20%, v/v glycerol at -20°C and -80°C .

108 The isolate was examined for motility and morphological properties using procedures
109 described by Trujillo *et al.* [24]. Cultural features were recorded on modified Bennett's
110 (DSMZ medium No. 894), GPHF (DSMZ medium No. 553), GYM *Streptomyces* (DSMZ
111 medium No. 65), Luedemann's [6], potato dextrose (DSMZ medium No. 129), Reasoner's
112 2A (DSMZ medium No. 830) and from tryptone-yeast extract, yeast extract-malt extract,
113 oatmeal, inorganic salts-starch, glycerol-asparagine, peptone-yeast extract-iron and tyrosine
114 agar (International *Streptomyces* Project [ISP] media 1–7; [25]) plates following incubation
115 at 28°C for 3 weeks. The isolate was found to be a Gram-stain positive, non-motile
116 actinobacterium that formed rods and coccoid shaped cells with evidence of budding (Fig.
117 1). Strain P6^T was observed to grow well on GYM *Streptomyces*, GPHF, modified Bennett's,
118 potato dextrose, Luedemann's, Reasoner's 2A and yeast extract-malt extract agar, as
119 exemplified in Figure S1, but poorly on ISP media 1, 3 to 7, generally producing red-orange
120 or yellowish pink pigments; diffusible pigments were not observed on any of these media.

121 Biomass for most of the chemotaxonomic analyses carried out on isolate P6^T was harvested
122 from 1,000 ml yeast extract-malt extract broth ISP medium 2 that had been shaken in 500 ml
123 baffled Erlenmeyer flasks, each flask containing 200 ml of medium, at 180 revolutions per
124 minute (rpm) at 28°C for 2 weeks; the biomass was washed twice in distilled water and

125 freeze-dried. Biomass for the fatty acid analysis was prepared on PYGV agar (DSMZ
126 medium No. 621), modified by the inclusion of 2 g of peptone instead of casein, 2 g of yeast
127 extract and 10 ml of a 20% w/v glucose, after incubation at 20°C for 16 days and washed
128 twice in sterile distilled water.

129 Isolate P6^T was examined for chemotaxonomic markers known to be of value in the
130 systematics of microorganisms classified in the genus *Blastococcus* [4, 18]. Standard
131 chromatographic procedures were used to determine the isomers of A₂pm [26], isoprenoid
132 quinones [27], whole-cell sugars [28] and polar lipids [29], as modified by Kroppenstedt and
133 Goodfellow [30]. Isoprenoid quinones extracted from *Micromonospora luteifusca* GUI2^T
134 [31] were used as standards. In turn, fatty acids extracted from the isolate were methylated,
135 analysed using the protocol of the Sherlock Microbial Identification (MIDI) system, version
136 5 [32] and the resultant peaks identified using the ACTIN 6 database.

137 In general, the chemotaxonomic properties of isolate P6^T are consistent with its classification
138 in the genus *Blastococcus* [4, 18, 20]. The organism contains *meso*-A₂pm as the diagnostic
139 diamino acid (Fig. S2); MK-9(H₂) and MK-9(H₄) as predominant isoprenologues in a
140 proportion of 3:2 (Fig. S3); iso-C_{16:0} (38.9%), iso-C_{16:1} H (17.7%), iso-C_{15:0} (14.2%) and 9-
141 methyl-C_{16:0} (5.7%) as major fatty acids; a polar lipid profile that includes
142 diphosphatidylglycerol, phosphatidylcholine, phosphatidylethanolamine,
143 phosphatidylglycerol and phosphatidylinositol (Fig. S4) and galactose and glucose as major
144 sugars with a lesser proportion of ribose and traces of arabinose and xylose (Fig. S5).

145 Genomic DNA for 16S rRNA gene sequencing was extracted from 25 ml ISP 2 broth
146 incubated at 28°C, shaken at 180 rpm for 10 days; 5 ml of the fully grown culture were used
147 for genomic DNA extraction, following the protocol used by Kieser *et al.* [33] though in this
148 case incubation with protease K was conducted at 60°C until the solution became clear
149 (~1.5h); the quality of the isolated genomic DNA was checked in a 1%, w/v agarose gel run
150 at 70V for 1.5h. PCR-mediated amplification of the 16S rRNA gene was performed in a final
151 volume of 25 µl using the standard primers 27F and 1525r [34], 100 ng of genomic DNA and
152 MyFi™ Mix (Bioline, UK), following the manufacturer's instructions; the PCR conditions
153 were 5min at 95°C followed by 30 cycles of 30sec at 95°C, 30sec at 55°C and 23sec at 72°C.
154 Two µl of the resulting PCR mixture was passed through a 1%, w/v agarose gel from which
155 a single band of the expected size (about 1,500 bp) was visualised. The rest of the PCR

156 mixture was cleaned with exonuclease I and shrimp alkaline phosphatase (NEB, UK;
157 #E2622S) and sent to Geneius (Cramlington, UK) for sequencing, using a BigDye®
158 terminator v3.1 cycle sequencing kit (Thermo Fisher Scientific Inc.) on an ABI sequencer
159 model 3730xl; the sequence was assembled using Pregap4 and Gap4 from Staden Package
160 version 2.0.0b9 [35].

161

162 The fragment of the 16S rRNA gene sequence of isolate P6^T (1,340 bp) was compared with
163 corresponding 16S rRNA gene sequences of the type strains of the *Blastococcus* species, the
164 sequence of “*Candidatus B. massiliensis*” AP3 and those of *Cumulibacter manganitolerans*
165 DSM 103787^T, *G. obscurus* DSM 43160^T, ‘*Klenkia marina*’ DSM 45722^T and
166 *Modestobacter multiseptatus* DSM 44406^T all of which were retrieved from the EzBioCloud
167 server (<http://www.ezbiocloud.net/>; [36]). Alignment of the nucleotide sequences was
168 achieved with MUSCLE [37]. Phylogenetic trees were inferred with the MEGA7 suite of
169 programs version 7.0 [38] using the NJ [39], ML [40] and MP algorithms [41] with 1,000
170 bootstrap repetitions [42]; evolutionary distances were calculated with the Kimura 2-
171 parameters model [43]. Sequence similarity values were calculated based on the alignment
172 generated with MUSCLE [37], using PHYDIT software version 3.0. In addition, pairwise
173 sequence similarities and phylogenetic reconstruction were performed using the method
174 recommended by Meier-Kolthoff *et al.* [44] and the GGDC web server [45] available at
175 <http://ggdc.dsmz.de/phylogeny-service.php#> and the Genome-to-Genome Distance
176 Calculator (GGDC; [44]) to validate the results obtained using the MEGA7 software. The
177 resultant trees were rooted with *G. obscurus* DSM 43160^T, *K. marina* YIM M1315 and *M.*
178 *multiseptatus* DSM 44406^T using FigTree version 1.4.2
179 (<http://tree.bio.ed.ac.uk/software/figtree/>). All of the Figures were edited in Inkscape version
180 0.9 (<https://inkscape.org/en/download/>).

181 Strain P6^T was found to form a well-supported subclade in the *Blastococcus* 16S rRNA gene
182 tree together with the type strains of *B. capsensis* and *B. saxobsidens* (Fig. 2). It is most
183 closely related to type strain of the *B. saxobsidens* sharing a 16S rRNA gene similarity of
184 99.5%, a value that corresponds to 7 nucleotide (nt) differences at 1,339 locations. The
185 corresponding 16S rRNA gene sequence similarity value between P6^T and *B. capsensis* BMG
186 804^T was 99.1%, which equates to 10 nt differences at 1,241 sites. The 16S rRNA gene

187 sequence similarities between strain P6^T and the other *Blastococcus* type strains fell within
188 the range 97.7 and 98.1%, values corresponding to between 26 and 31 nt differences. The
189 same pairwise similarity values were recorded between the isolate and the type strains of *B.*
190 *capsensis* and *B. saxobsidens* using the GGDC server.

191 Isolate P6^T and *B. saxobsidens* DSM 44509^T were examined for a broad range of phenotypic
192 properties. Enzyme profiles were determined using API ZYM strips (BioMérieux) by
193 following the manufacturer's instructions, while GEN III microplates (Biolog Inc., Hayward,
194 CA, USA) were used to test for the ability of the strains to oxidise carbon and nitrogen
195 sources and to determine resistance to inhibitory compounds using inoculating fluid (IF-C,
196 Biolog Inc.) and a cell density of 86% transmittance in an OmniLog instrument (Biolog Inc.)
197 set at 28°C. Data from the triplicated cultures recorded in Phenotypic Mode from the GENIII
198 microplates were analysed using opm package 1.0.6 [46] for R [47], using R studio [48].
199 Many of the remaining tests were carried out using ISP 2 agar [25] as the basal medium. The
200 strains were examined for their ability to grow at a range of pH values (pH 5–10 at single
201 unit intervals; pH was adjusted by adding drops of either 1N NaOH or 1N HCl as described
202 by Montero-Calasanz *et al.* [49]) and temperatures (4, 10, 20, 28, 37, 40, 45 and 50°C) and
203 in the presence of various concentrations of sodium chloride (1.0, 1.5, 3.0, 5.0, 7.0, 15.0 and
204 20%, w/v). Apart from the temperature tests these features were recorded after incubation at
205 28°C. Results of all of these tests were recorded after incubation for 3 weeks. The ISP 2
206 medium was also used to test the capacity of the strains to degrade casein (1%), cellulose
207 (1%), elastin (0.3%), guanine (0.5%), hypoxanthine (0.4%), L-tyrosine (0.4%), uric acid
208 (0.5%) and xanthine (0.4%), their ability to degrade tributyrin was determined using
209 tributyrin agar (Sigma-Aldrich). Results of all of these tests were recorded after incubation
210 at 28°C for 14 days. Aesculin (0.1%) and arbutin (0.1%) hydrolysis was established using the
211 media and methods described by Williams *et al.* [50], the hydrolysis of urea (0.2%, w/v) after
212 Christensen [51] and nitrate reduction following Schaal *et al.* [52]. Catalase production was
213 detected by the formation of bubbles after mixing a fresh drop of 3% hydrogen peroxide to
214 fresh growth of the cultures on glass slides. Oxidase activity was determined in a 1%, w/v
215 solution of *N-N-N'-N'*-tetramethyl-1,4-phenyldiamine (Sigma-Aldrich) and the development
216 of a blue purple colour was recorded as a positive result [53]. The degradation and tolerance

217 tests were carried out in triplicate using a cell suspension equivalent to 5.0 on the McFarland
218 scale [54].

219 The triplicated tests on isolate P6^T and *B. saxobsidens* DSM 44509^T gave identical results for
220 all of the phenotypic tests, apart from the ability of isolate P6^T to degrade arbutin.

221 The phenotypic properties of isolate P6^T and *B. saxobsidens* DSM 44509^T strains were
222 compared with those of the other type strains of *Blastococcus* species, which had mainly been
223 examined using the same procedures, as exemplified by the API, GENIII microplate,
224 tolerance and catalase tests [4, 20]. It can be seen from Table 1 that the isolate can be
225 distinguished from all of the *Blastococcus* type strains, including *B. capsensis* DSM 46835^T
226 and *B. saxobsidens* DSM 44509^T, its nearest phylogenetic neighbours, using a combination
227 of phenotypic features. It can be distinguished from each of these organisms by its ability to
228 grow at 10 and 45°C, to reduce nitrate to nitrite and use dextrin, α -keto-butyric acid and D-
229 malic acid, and by its inability to use glucuronamide and D-saccharic acid as sole carbon
230 sources. It can also be separated from the *B. saxobsidens* type strain by its capacity to oxidise
231 pectin and D-salicin. It is also evident from Table 1 that several chemotaxonomic features
232 support the separation of isolate P6^T from the type strains of *Blastococcus* species. Isolate
233 P6^T and *B. saxobsidens* DSM 44509^T were also found to share many phenotypic features:
234 they produce acid and alkaline phosphatases, α -chymotrypsin, cysteine arylamidase, esterase
235 (C4), esterase lipase (C8), α -glucosidase, naphthol-AS-B1-phosphohydrolase and valine
236 arylamidase; degraded aesculin, tributyrin and starch; but do not produce lipase (C14), α -
237 mannosidase, or α -fucosidase or hydrolyse allantoin and urea, or degrade casein, cellulose,
238 elastin, guanine, hypoxanthine, L-tyrosine, uric acid, or xanthine. In addition, they can utilise
239 α - and β -hydroxy-butyric acid, D-cellobiose, D-fucose, β -gentiobiose, D-gluconic acid, α -D-
240 lactose, D-salicin, sucrose and D-turanose but not L-alanine, D-arabitol, D-aspartic acid,
241 glycyl-proline, gelatin, mucic acid or *p*-hydroxy-phenylacetic acid.

242 Biomass for sequencing the whole-genome of isolate P6^T was prepared in a 1.5 ml of brain-
243 heart infusion broth at 28°C in a shaking incubator (180 rpm) for 2 days. Genomic DNA of
244 strains was extracted using the QIAamp DNA extraction kit (Qiagen, USA) according to the
245 manufacturer's instructions. The purity and concentration of the extracted genomic DNA
246 were measured using the Nanodrop spectrophotometer (NanoDrop Technologies, UK).
247 Genome sequencing was performed on an Illumina MiSeq instrument (Illumina); the reads

248 were assembled by using SPAdes 3.9.0 [55] and contigs smaller than 1,000 bp in size were
249 discarded. The draft assemblies have been submitted to the GenBank (accession numbers:
250 POQU000000000 and POQT000000000) and is publicly available.

251 The genomes were annotated using the RAST annotation pipeline [56] and a sequenced based
252 comparison performed in the SEED Viewer [56, 57]. A digital DNA:DNA hybridisation
253 (dDDH) value was calculated between the genomes of strain P6^T and *B. saxobsidens* DSM
254 44509^T using the GGDC server [45]. BLAST-based ANI and AAI between the strains were
255 calculated using the online resource from the K. Konstantinidis group (<http://enve-omics.ce.gatech.edu/>; [58]).

257 The draft genomes of strain P6^T and *B. saxobsidens* DSM 44509^T, contained 3,778 and 4,348
258 open reading frames, respectively, and were 3.9 Mb and 4.5 Mb in size with average *in silico*
259 DNA G+C contents of 73.1 and 74.3 mol%. The dDDH value between the genome of the
260 two strains was 27.8% (C. I. 25.4-30.3%), which is well below the 70% threshold used to
261 confirm the species status of novel strains [59]. The corresponding ANI and AAI indices
262 were 84.6± 4.5 and 83.0± 13.0, values below the threshold used for prokaryotic species
263 delineation [58, 60, 61].

264 The draft genomes of isolates P6^T and *B. saxobsidens* DSM 44509^T were examined using the
265 antiSMASH server [62] to detect putative biosynthetic gene clusters. The genome of isolate
266 P6^T was found to encode for a T3PKS and corresponding residues that make up the catalytic
267 triad found in RppA, a T3PKS involved in the biosynthesis of pentaketide 1,3,6,8-
268 tetrahydroxynaphthalene in *Streptomyces griseus* [63, 64]. The T3PKS of isolate P6^T showed
269 94% sequence identity with a corresponding sequence detected in the genome of the *B.*
270 *saxobsidens* strain and 83% identity with a putative T3PKS encoded in the genome of *G.*
271 *obscurus* DSM 43160^T (Gobs_4821; UniProt [65], accession number: D2S5V1). The gene
272 that encodes for the T3PKS of isolate P6^T was surrounded by other biosynthetic genes, such
273 as one encoding for a methyltransferase and others encoding regulatory and transport
274 proteins, thereby suggesting the presence of a biosynthetic gene cluster though the
275 functionality and product generated by this putative biocluster has still to be established. The
276 genomes of isolate P6^T and the *B. saxobsidens* strain were also found to harbour genes
277 encoding for polyprenyl synthetase and phytoene synthase, enzymes involved in the
278 biosynthesis of terpenoid compounds [66, 67]. The genome of the *B. saxobsidens* strain

279 contains two genes that encode for proteins that contain the IucA/IucC domain (Pfam [68]
280 accession: pfam04183) which is involved in the biosynthesis of siderophore compounds [69].
281 The genome of the type strain of *Modestobacter caceserii* which, like P6^T, was isolated from
282 an extreme hyper-arid soil sample collected from the Yungay core region of the Atacama
283 Desert, contained a siderophore gene cluster predicted to encode for deferoxamine; the
284 genome of this organism also contained gene clusters encoding for type II and III polyketides
285 and terpenes [70]. These preliminary datasets suggest that the genomes of
286 *Geodermatophilaceae* strains have the capacity to produce specialised metabolites such as
287 polyketides and siderophores. However, antiSMASH does not necessarily detect all of the
288 gene clusters in genomes, as exemplified by the failure to identify the hygromycin A gene
289 cluster in *Streptomyces leewenhoekii* C34^T [71], moreover an improved genome assembly is
290 required for a more precise interpretation of predicted biosynthesis gene clusters in the
291 genome of isolate P6^T.

292

293 A comparison of the genomes of isolate P6^T and *B. saxobsidens* DSM 44509^T showed that
294 the genome of the former contains 474 genes that are absent from the genome of the latter,
295 including those involved in stress responses (2 copies of *terA* and 3 copies of *terD* genes; P6-
296 peg 850-P6-peg 855) [72]. In contrast, the genome of the *B. saxobsidens* strain harbours
297 2,848 genes that are absent from isolate P6^T (data not shown); these genes include multiple
298 copies of the *tetA*, *tetB*, and *tetC* genes that are involved in tricarboxylate/citrate transport.
299 However, most of the unique genes (54-66%) found in the genomes of these strains encode
300 for hypothetical proteins.

301 The genomes of isolate P6^T and the *B. saxobsidens* DSM 44509^T contained 119 and 147
302 genes, respectively that are associated with stress responses (Table S1). Each of the strains
303 contained two genes involved in carbon starvation, one encoding for carbon starvation
304 protein A and the other for a carbon storage regulatory protein indicating that they are adapted
305 to life in low carbon environments [73-75]. Similarly, four genes belonging to the CspA
306 family associated with responses to cold-shock [76], 13 genes of the *dnaK* gene cluster that
307 respond to heat shock [77] and seven genes associated with the biosynthesis, uptake and
308 utilisation of trehalose, which are considered to help in responses to heat and desiccation
309 stress [78], are conserved in each of the strains. The genomes of the strains also contain genes

310 belonging to the *uvrABC* DNA repair system that assists in UV resistance [79], as well as
311 Rec proteins (RecA, RecX and those involved in the RecBCD and RecFOR pathways) that
312 are responsible for stabilising genomes [80]. The P6^T genome contains a *coxGMLS* gene
313 cluster and a *coxD* gene, whereas the *B. saxobsidens* strain has five copies of the *coxM* gene
314 and two copies of the *coxS* gene, though the gene encoding for the *coxG* protein is absent;
315 *cox* genes code for the utilisation of carbon monoxide, thereby indicating that these
316 organisms may have a chemolithoautotrophic lifestyle [81].

317 The proteins involved in responses to oxidative stress are, with minor exceptions, conserved
318 in the genomes of isolate P6^T and *B. saxobsidens* DSM 44509^T (Table S1). The P6^T genome
319 contains eight genes involved in responses to osmotic stress, notably choline dehydrogenase,
320 and ABC transporter proteins for betaine, glycine and L-proline uptake and a high affinity
321 choline uptake protein (*betT*) [82, 83]. In turn, only the *B. saxobsidens* strain has a *sox* gene
322 cluster encoding the subunit of sarcosine oxidase, along with additional copies of *betT* and
323 transport proteins. Sarcosine oxidase is associated with responses to osmotic stress [84, 85].

324

325 It is clear from the chemotaxonomic, genomic, morphological and phylogenetic data that
326 isolate P6^T is a *bona fide* member of the genus *Blastococcus*. It can be distinguished from the
327 type strains of *Blastococcus* species using a broad range of phenotypic features and from *B.*
328 *saxobsidens* BC448^T, its close phylogenetic relative, by low ANI and AAI indices and by a
329 low *in silico* DNA:DNA pairing value. It can be concluded that the isolate should be
330 recognised as a new *Blastococcus* species, for which we propose the name *Blastococcus*
331 *atacamensis* sp. nov.

332

333 This is the first description of a novel *Blastococcus* species from the Atacama Desert though
334 there are grounds for believing that others will follow [86], especially since culture-
335 independent studies show that blastococci are part of the core microbiome of hyper- and
336 extreme hyper-arid soils of the desert landscape [87]. It is also interesting that the genomes
337 of *B. atacamensis* P6^T and *M. caceserii* KNN 45-2b [70], another isolate from the Yungay
338 core region, contain genes or gene clusters associated with an ability to cope with low levels
339 of carbon [73-75], osmotic stress [82], high UV radiation [79] and heat tolerance and
340 desiccation (biosynthesis and uptake of trehalose; [78]). The genomes of these strains also

341 contain multiple *cox* genes suggesting that *Geodermatophilaceae* strains from the Atacama
342 may be able to use carbon monoxide as a carbon and energy source, an observation in line
343 with the suggestion that facultatively chemoautotrophic bacteria may sustain microbial
344 communities in the nutrient impoverished high altitude Atacama Desert soils [88, 89].
345 Biological adaptations such as these may account for the presence of blastococci in habitats
346 characterised by scarcity of available water, low nutrient availability and extremes of
347 temperature and UV radiation levels [2, 14].

348

349 **Description of *Blastococcus atacamensis* sp. nov.**

350 *Blastococcus atacamensis* (a.ta.cam.en'sis. N.L. masc. adj. *atacamensis*; belonging to the
351 Atacama Desert, the source of the isolate).

352

353 Gram-stain positive, oxidase-negative actinobacterium which forms non-motile, rod-and
354 coccoid-elements with evidence of budding. Round orange colonies with entire margins are
355 formed on yeast extract-malt extract agar. Grows from 10–45°C, optimally ~35°C, from pH
356 6–12, optimally ~pH 7.0 and in the presence of 3%, w/v sodium chloride. Degrades starch
357 and tributyrin but not guanine. Arbutin is hydrolysed. Utilises L-glutamic acid (amino acid),
358 D-glucose, glycerol, α -methyl-D-glucoside, *N*-acetyl-D-glucosamine (sugars), acetic acid,
359 acetoacetic acid, γ -amino-*n*-butyric acid, α - and β -hydroxybutyric acid, α -ketobutyric acid,
360 D-gluconic acid, α -ketoglutaric acid, D-malic acid and propionic acid (organic acids), but not
361 D-mannose, D-melibiose, *N*-acetyl-neuraminic acid or L-rhamnose (sugars) or butyric acid,
362 mucic acid, D-saccharic acid, α -hydroxyphenylacetic acid or bromosuccinic acid (organic
363 acids); is resistant to aztreonam (antibiotic), lithium chloride and potassium tellurite (heavy
364 metals) and Tween 40 (surfactant) but is sensitive to fusidic acid, lincomycin, minocycline,
365 rifamycin SV, troleandomycin and vancomycin (antibiotics), guanidine HCl (chaotropic
366 agent), tetrazolium blue and tetrazolium violet (redox indicators), sodium bromide, sodium
367 formate and sodium lactate (salts) and niaproof (surfactant) (GENIII microplates). Additional
368 phenotypic features are cited either in the text or in Table 1. The predominant fatty acids are
369 iso-C_{16:0}, iso-C_{16:1} H, iso-C_{15:0} and 9-methyl-C_{16:0}; the major sugars are galactose, glucose
370 and ribose; the polar lipid profile contains diphosphatidylglycerol, phosphatidylcholine,
371 phosphatidylethanolamine, phosphatidylglycerol and phosphatidylinositol, an unidentified

372 lipid and an unidentified phosphoglycolipid. Additional chemotaxonomic properties are
373 typical of the genus. The *in silico* DNA G+C content of the type strain is 73.1 mol%. The
374 type strain, P6^T (=NCIMB 15090^T = NRRL B-65468^T) was isolated from an extreme hyper-
375 arid soil sample from the Yungay core region of the Atacama Desert.

376

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389 **Conflicts of interest**

390 The authors declare that they do not have any conflict of interest.

391 **Ethical statement**

392 The authors have not carried out any studies involving human participants or animals.

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635 *Geophysical Research: Biogeosciences* 2012;117(G2):G02028.

636

639 **Table 1:** Phenotypic properties that differentiate isolate P6^T from the *Blastococcus* type strains.

Characteristics	1	2	3	4	5	6	7
Cell shape	Cocci	Cocci, rods, vibrios ^d	Cocci	Cocci, rods, vibrios	Cocci ^e	Cocci, rods ^c	Cocci ^d
Bud formation	+	+ ^d	–	+	– ^e	+ ^c	– ^d
Germ tube	+	+ ^d	–	+	– ^e	– ^c	– ^d
Motility	–	+ ^d	–	–	– ^e	+ ^c	+ ^d
Pigmentation	Light-pink to mild red-orange	Pink	Bright orange	Coral	White to pink to black ^e	Apricot	Pink to orange
Temperature growth range (°C)	10–45	10–40 ^d	20–30	10–40	10–45 ^e	10–37 ^c	20–37 ^d
Biochemical tests:							
Catalase	+	+	–	–	+ ^e	+ ^c	+ [*]
Nitrate to nitrite reduction	+	– ^e	– ^b	–	– ^e	+ ^e	– ^e
API ZYM tests:							
Acid phosphatase	+	– ^e	– ^b	–	+ ^e	– ^e	+ [‡]
Alkaline phosphatase	+	– ^e	+ ^b	+	+ ^e	– ^e	+ ^e
Esterase lipase (C 8)	+	+ ^e	+ ^b	–	+ ^e	– ^e	+ ^e
α-Glucosidase	+	– ^e	+ ^b	+	+ ^e	– ^e	+ [‡]
Naphthol-AS-BI-phosphohydrolase	+	– ^e	– ^b	–	+ ^e	– ^e	+ ^e
Valine arylamidase	+	– ^e	– ^b	+	+ ^e	+ ^e	+ [‡]
GENIII Biolog microplates:							
Oxidation of Amino acids:							
L-Alanine	–	+	–	+	v	+	–

Glycyl-L-proline(dipeptide)	–	+	v	–	–	–	–
Monosaccharides:							
<i>N</i> -Acetyl-neuraminic acid	–	+	–	–	–	–	v
Glucuronamide	–	v	+	–	+	v	+
Disaccharide:							
β -Gentiobiose	+	+	–	+	+	+	+
Sugar alcohol:							
D-Salicin	+	–	–	v	v	–	+
Polymers:							
Dextrin	+	+	–	+	+	–	–
Gelatin	–	+	–	–	v	+	–
Pectin	+	–	+	v	–	–	–
Tween 40	+	+	–	+	+	+	v
Organic acids:							
D-Gluconic acid	+	–	+	+	+	v	+
β -hydroxy-Butyric acid	+	–	+	+	v	+	+
α -keto-Butyric acid	+	v	–	+	–	+	–
D-Malic acid	+	+	+	v	+	+	–
Methyl pyruvate	+	–	–	v	–	–	v
Mucic acid	–	–	–	v	–	+	–
D-Saccharic acid	–	–	–	+	+	v	+
Phospholipids	DPG, L, PC PE, PG, PGL, PI	DPG, PI, GPL, PC	DPG, PE, PC, PI, 2PL	DPG, PC, PI, GPL, PE, OH-PE, 6PL	DPG, PI, PE, PC	DPG, PC, PME, PE, PI, GPL	DPG, PE, PC, PI, GLP, 3PL
Diagnostic sugars	Glu, Gal, Rib; traces of Ara, Xyl	Rib, Ara, Man, Glu	Glu, Rha, Rib	Glu, Gal, Rib	Glu, Gal; traces of	Rha, Rib, Xyl, Glu;	Glu, Gal; traces of Rib, Man

Menaquinones (MK)	MK-9(H ₄), MK-9(H ₂)	MK-8(H ₄), MK-9(H ₄)	MK-9(H ₄), MK-9, MK- 9(H ₂)	MK-9(H ₄), MK-9, MK- 8(H ₄)	Rha, Rib, Man, Ara MK-9(H ₄), MK-9, MK- 8(H ₄)	traces of Man MK-9(H ₄), MK-9	MK-9(H ₄), MK-8(H ₄), MK-9, MK- 9(H ₆), MK- 9(H ₂)
DNA G + C content (mole %)	73.1	73.9 ^d	73.7	73.2	71.6 ^e	72.3 ^c	73.5

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642 Strains: **1**, Isolate P6^T; **2**, *B. aggregatus* DSM 4725^T; **3**, *B. capsensis* DSM 46835^T; **4**, *B. colisei* DSM 46837^T; **5**, *B. endophyticus* DSM
643 45413^T; **6**, *B. jejuensis* DSM 19597^T; **7**, *B. saxobsidens* DSM 44509^T. Data for **1** and **7** are from this study; those for **2** to **6** are from
644 Hezbri et al. [20] unless indicated. + positive; – negative; v variable. Data taken from ^a Ahrens and Moll [1]; ^b Hezbri et al. [4]; ^c Lee [3];
645 ^d Urzì et al. [19] and ^e Zhu et al. [17]; * recorded as negative by Hezbri et al. [4]; † recorded as negative by Zhu et al. [17]. Abbreviations:
646 Ara, arabinose; Glu, glucose; Gal, galactose; Man, mannose; Rha, rhamnose; Rib, ribose; Xyl, xylose; DPG, diphosphatidylglycerol;
647 GPI, glycoposphatidylinositol; PE, phosphatidylethanolamine; PME, phosphatidyl-*N*-methylethanolamine; PC, phosphatidylcholine;
648 PI, phosphatidylinositol; unidentified: GPL, glycopospholipid, L, lipid, PL, phospholipid; MK, menaquinone.

649 **Legends for Figures**

650

651 **Figure 1.** Phase contrast image of isolate P6^T following growth on ISP 2 at 28°C for 7 days
652 showing the presence of coccoid and rod-shaped elements and evidence of budding. Scale
653 bar: 5µm.

654 **Figure 2.** Neighbour-joining tree based on partial 16S rRNA gene sequences (1,239
655 nucleotides) showing the relationships between isolate P6^T and the type strains of
656 *Blastococcus* species and the candidatus strain. Asterisks indicate branches of the tree that
657 were also found using the maximum-likelihood (ML) and maximum-parsimony (MP) tree-
658 making algorithms. Numbers at the nodes indicate levels of bootstrap support (%) above 50%
659 based on a neighbour-joining analysis of 1,000 resampled datasets. Genbank accession
660 numbers are indicated in parentheses. The scale bar indicates the number of substitutions per
661 nucleotide position.

662

663 **Supplementary Figures**

664 Fig. S1. Light microscopy images of colonies of isolate P6^T following growth on ISP 2 at
665 28°C for 3 weeks.

666 Fig. S2. Thin-layer chromatographs showing (a) the presence of diaminopimelic acid (A₂pm)
667 isomers and (b) the presence of *meso*-A₂pm in whole-cell hydrolysates of isolate P6^T.

668 Fig. S3. Menaquinone profile of isolate P6^T. Isoprenoid quinones extracted from
669 *Micromonospora luteifusca* GUI2^T [1] were used as standards.

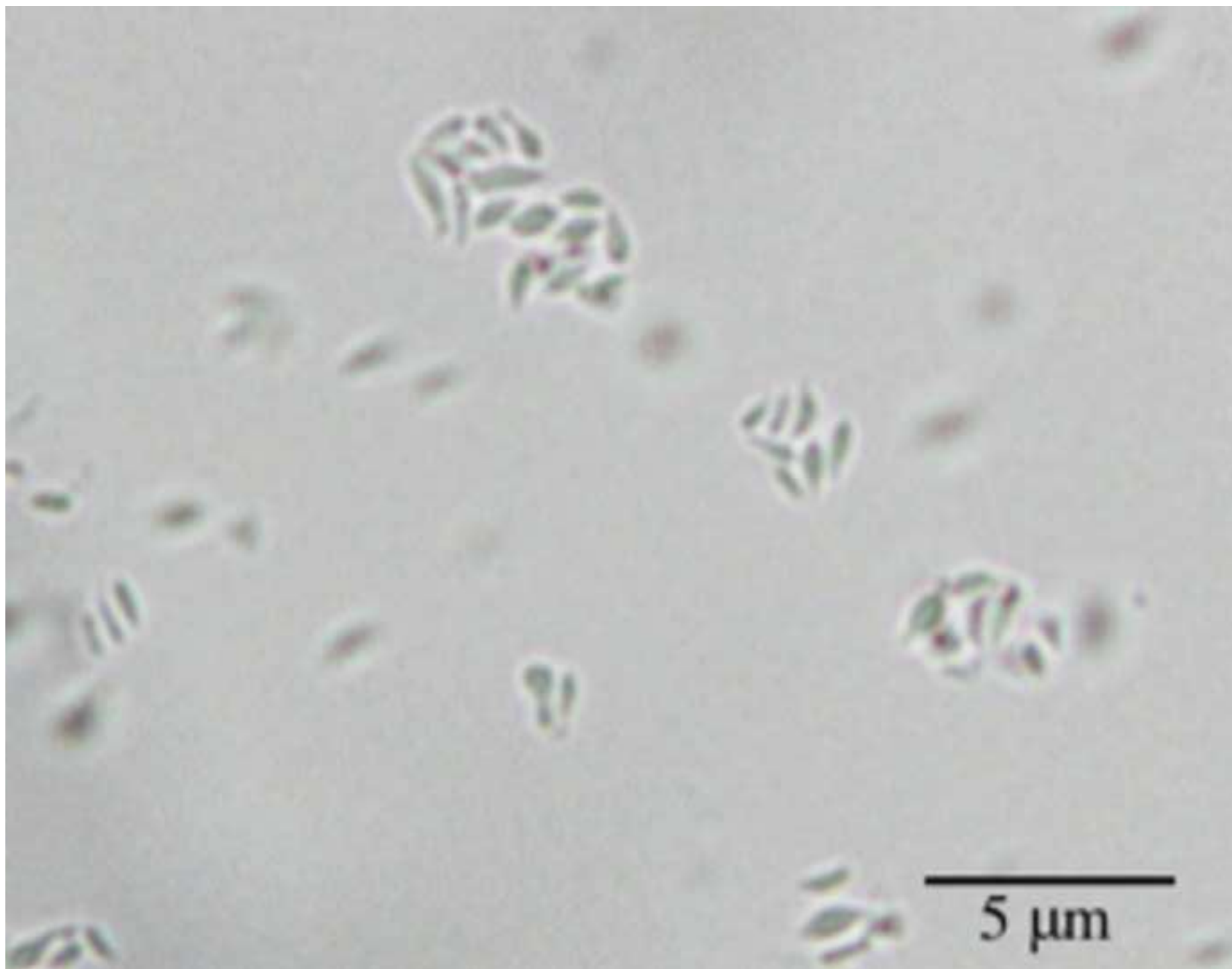
670 Fig. S4. Bi-dimensional thin-layer chromatography of polar lipids of isolate P6^T.

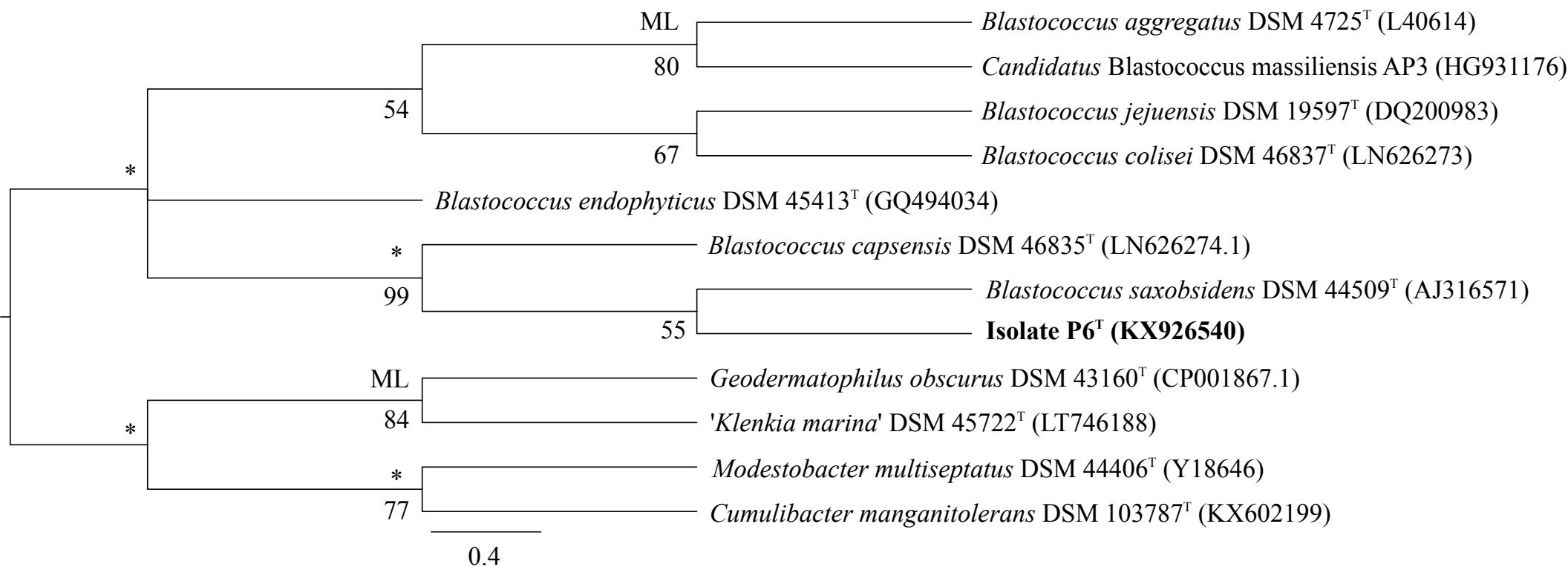
671 Fig. S5. Thin-layer chromatographs showing the presence of (a) standard sugars and (b)
672 sugars in whole-cell hydrolysates of isolate P6^T.

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International Journal of Systematic and Evolutionary Microbiology**Supplementary data*****Blastococcus atacamensis* sp. nov., a novel strain adapted to life in the Yungay core region of the Atacama Desert**

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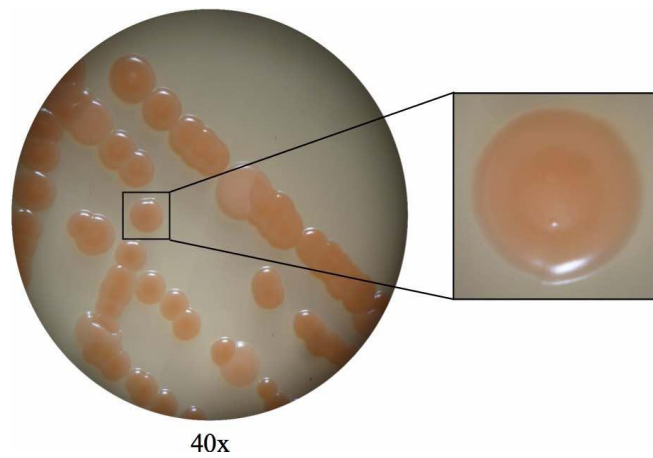
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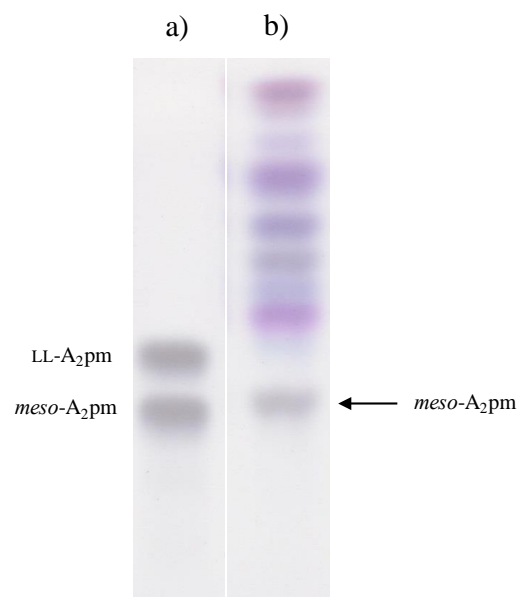
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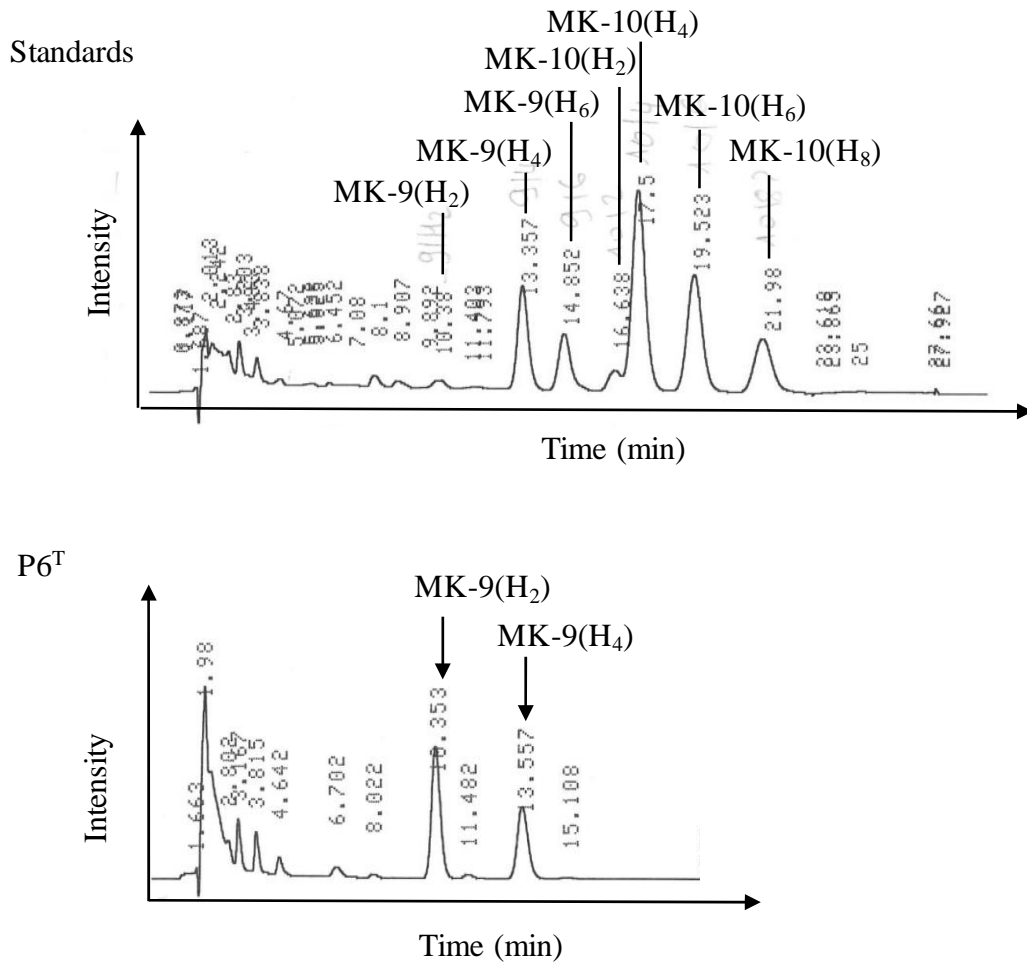
*To whom correspondence should be addressed: phone +44-(0)191 2087706; email: m.goodfellow@ncl.ac.uk.



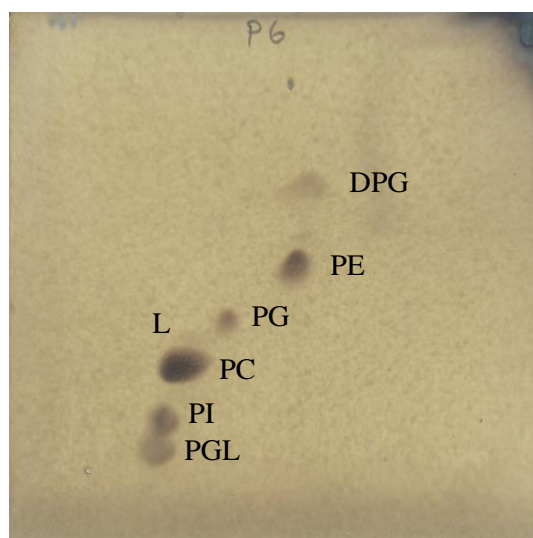
Supplementary Figure S1: Light microscopy images of colonies of isolate P6^T following growth on ISP 2 at 28°C for 3 weeks.



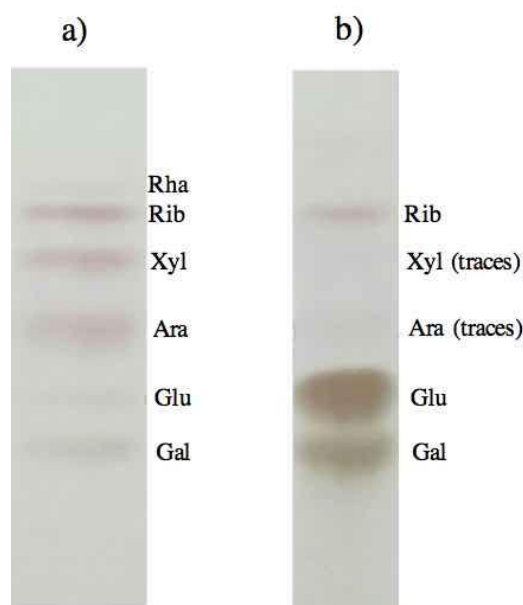
Supplementary Fig. S2: Thin-layer chromatographs showing (a) the presence of diaminopimelic acid (A₂pm) isomers and (b) the presence of *meso*-A₂pm in whole-cell hydrolysates of isolate P6^T. a) Standards A₂pm; b) isolate P6^T.



Supplementary Fig. S3: Menaquinone profile of isolate P6^T. Isoprenoid quinones extracted from *Micromonospora luteifusca* GUI2^T [1] were used as standards.



Supplementary Fig. S4: Bi-dimensional thin-layer chromatography of polar lipids of isolate P6^T using molybdato-phosphoric acid reagent (5 %). DPG: diphosphatidylglycerol; L: unidentified lipid; PC: phosphatidylcholine; PE: phosphatidylethanolamine; PG: phosphatidylglycerol; PGL: Unknown phosphoglycolipid; PI: phosphatidylinositol. Solvent 1: chloroform:methanol:distilled water (65:25:4); solvent 2: chloroform:glacial acetic acid:methanol:distilled water (80:12:15:4).



Supplementary Fig. S5. Thin-layer chromatographs showing the presence of (a) standard sugars and (b) sugars in whole-cell hydrolysates of isolate P6^T. a) Standard sugars; b) sugars present in isolate P6^T. Rha: rhamnose; Rib: ribose; Xyl: xylose; Ara: arabinose; Glu: glucose; Gal: galactose.

Supplementary Table 1. A list of stress related genes present in strains P6^T and DSM 44509^T

Presence	Category	Role	Organism A	Organism B
P6 ^T and DSM 44509 ^T	Cold shock	Cold shock protein CspA	P6_peg1370, P6_peg2786	DSM44509_peg149, DSM44509_peg3572
P6 ^T and DSM 44509 ^T	Cold shock	Cold shock protein CspC	P6_peg575	DSM44509_peg176
P6 ^T and DSM 44509 ^T	Cold shock	Cold shock protein CspG	P6_peg2467	DSM44509_peg2911
P6 ^T	Detoxification	Various polyols ABC transporter, periplasmic substrate-binding protein	P6_peg1164, P6_peg1169	
P6 ^T and DSM 44509 ^T	Heat shock	Chaperone protein DnaJ	P6_peg274, P6_peg1954	DSM44509_peg382, DSM44509_peg2184
P6 ^T and DSM 44509 ^T	Heat shock	Chaperone protein DnaK	P6_peg226	DSM44509_peg384
P6 ^T and DSM 44509 ^T	Heat shock	Heat shock protein GrpE	P6_peg227	DSM44509_peg383
P6 ^T and DSM 44509 ^T	Heat shock	Heat-inducible transcription repressor HrcA	P6_peg1955	DSM44509_peg2183
P6 ^T and DSM 44509 ^T	Heat shock	HspR, transcriptional repressor of DnaK operon	P6_peg275	DSM44509_peg381
P6 ^T and DSM 44509 ^T	Heat shock	Hypothetical radical SAM family enzyme in heat shock gene cluster, similarity with CPO of BS HemN-type	P6_peg1959	DSM44509_peg2178
P6 ^T and DSM 44509 ^T	Heat shock	Nucleoside 5-triphosphatase RdgB (dHAPTP, dITP, XTP-specific) (EC 3.6.1.15)	P6_peg6	DSM44509_peg2317
P6 ^T and DSM 44509 ^T	Heat shock	Ribonuclease PH (EC 2.7.7.56)	P6_peg5	DSM44509_peg2316
P6 ^T and DSM 44509 ^T	Heat shock	Ribosomal RNA small subunit methyltransferase E (EC 2.1.1.-)	P6_peg1953	DSM44509_peg2185
P6 ^T and DSM 44509 ^T	Heat shock	Translation elongation factor LepA	P6_peg1981	DSM44509_peg2165
P6 ^T and DSM 44509 ^T	Heat shock	rRNA small subunit methyltransferase I	P6_peg2916	DSM44509_peg235
P6 ^T and DSM 44509 ^T	Heat shock	tmRNA-binding protein SmpB	P6_peg3717	DSM44509_peg3854

P6 ^T and DSM 44509 ^T	Osmotic stress	Choline dehydrogenase (EC 1.1.99.1)	P6_peg827, P6_peg3514	DSM44509_peg321, DSM44509_peg1542
P6 ^T and DSM 44509 ^T	Osmotic stress	Glycine betaine ABC transport system permease protein	P6_peg1488	DSM44509_peg112
P6 ^T and DSM 44509 ^T	Osmotic stress	High-affinity choline uptake protein BetT	P6_peg2419	DSM44509_peg286, DSM44509_peg1388, DSM44509_peg1393, DSM44509_peg1579
P6 ^T and DSM 44509 ^T	Osmotic stress	L-proline glycine betaine ABC transport system permease protein ProV (TC 3.A.1.12.1)	P6_peg944, P6_peg1490	DSM44509_peg110, DSM44509_peg1084
P6 ^T and DSM 44509 ^T	Osmotic stress	L-proline glycine betaine ABC transport system permease protein ProW (TC 3.A.1.12.1)	P6_peg1489	DSM44509_peg111, DSM44509_peg1082, DSM44509_peg1083
P6 ^T and DSM 44509 ^T	Osmotic stress	L-proline glycine betaine binding ABC transporter protein ProX (TC 3.A.1.12.1)	P6_peg1487	DSM44509_peg113, DSM44509_peg279
DSM 44509 ^T	Osmotic stress	Sarcosine oxidase alpha subunit (EC 1.5.3.1)		DSM44509_peg1385
DSM 44509 ^T	Osmotic stress	Sarcosine oxidase beta subunit (EC 1.5.3.1)		DSM44509_peg1383
DSM 44509 ^T	Osmotic stress	Sarcosine oxidase delta subunit (EC 1.5.3.1)		DSM44509_peg1384
DSM 44509 ^T	Osmotic stress	Sarcosine oxidase gamma subunit (EC 1.5.3.1)		DSM44509_peg1386
DSM 44509 ^T	Osmotic stress	Aquaporin Z		DSM44509_peg31
P6 ^T and DSM 44509 ^T	Oxidative stress	Putative Holliday junction resolvase YggF	P6_peg2241	DSM44509_peg1712
P6 ^T and DSM 44509 ^T	Oxidative stress	Ribosomal RNA small subunit methyltransferase E (EC 2.1.1.-)	P6_peg1953	DSM44509_peg2185
P6 ^T and DSM 44509 ^T	Oxidative stress	Flavohemoprotein (Hemoglobin-like protein) (Flavohemoglobin) (Nitric oxide dioxygenase) (EC 1.14.12.17)	P6_peg3406	DSM44509_peg3484, DSM44509_peg3485
P6 ^T and DSM 44509 ^T	Oxidative stress	Hydroxyacylglutathione hydrolase (EC 3.1.2.6)	P6_peg65, P6_peg2230	DSM44509_peg1701, DSM44509_peg2761
P6 ^T and DSM 44509 ^T	Oxidative stress	Lactoylglutathione lyase (EC 4.4.1.5)	P6_peg2435	DSM44509_peg1561
P6 ^T and DSM 44509 ^T	Oxidative stress	Acetyl-CoA:Cys-GlcN-Ins acetyltransferase, mycothiol synthase MshD	P6_peg3623	DSM44509_peg3541
P6 ^T and DSM 44509 ^T	Oxidative stress	Formaldehyde dehydrogenase MscR, NAD/mycothiol-dependent (EC 1.2.1.66)	P6_peg2339	DSM44509_peg334, DSM44509_peg1381
P6 ^T and DSM 44509 ^T	Oxidative stress	Glycosyltransferase MshA involved in mycothiol biosynthesis (EC 2.4.1.-)	P6_peg3580	DSM44509_peg3513
P6 ^T and DSM 44509 ^T	Oxidative stress	L-cysteine:1D-myo-inositol 2-amino-2-deoxy-alpha-D-glucopyranoside ligase MshC	P6_peg3347	DSM44509_peg1070
P6 ^T and DSM 44509 ^T	Oxidative stress	Mycothiol S-conjugate amidase Mca	P6_peg321	DSM44509_peg2370

P6 ^T and DSM 44509 ^T	Oxidative stress	N-acetyl-1-D-myo-inosityl-2-amino-2-deoxy-alpha-D-glucopyranoside deacetylase MshB	P6_peg1013	DSM44509_peg1794
P6 ^T and DSM 44509 ^T	Oxidative stress	Putative hydrolase in cluster with formaldehyde/S-nitrosomycothioli reductase MscR	P6_peg2338	DSM44509_peg333
P6 ^T and DSM 44509 ^T	Oxidative stress	S-nitrosomycothioli reductase MscR	P6_peg2339	DSM44509_peg334, DSM44509_peg1381
P6 ^T and DSM 44509 ^T	Oxidative stress	Uncharacterized protein Rv0487/MT0505 clustered with mycothioli biosynthesis gene	P6_peg3579	DSM44509_peg3512
P6 ^T and DSM 44509 ^T	Oxidative stress	Alkyl hydroperoxide reductase subunit C-like protein	P6_peg3238	DSM44509_peg984
P6 ^T and DSM 44509 ^T	Oxidative stress	Catalase (EC 1.11.1.6)	P6_peg613	DSM44509_peg1819, DSM44509_peg3715
P6 ^T and DSM 44509 ^T	Oxidative stress	Organic hydroperoxide resistance protein	P6_peg279	DSM44509_peg379
P6 ^T and DSM 44509 ^T	Oxidative stress	Organic hydroperoxide resistance transcriptional regulator	P6_peg280	DSM44509_peg378
P6 ^T and DSM 44509 ^T	Oxidative stress	Phytochrome, two-component sensor histidine kinase (EC 2.7.3.-)	P6_peg28, P6_peg2209	DSM44509_peg2040, DSM44509_peg4012
P6 ^T and DSM 44509 ^T	Oxidative stress	Redox-sensitive transcriptional regulator (AT-rich DNA-binding protein)	P6_peg3266, P6_peg3267	DSM44509_peg3930, DSM44509_peg3931
P6 ^T and DSM 44509 ^T	Oxidative stress	Transcriptional regulator, FUR family	P6_peg612, P6_peg3592	DSM44509_peg3526, DSM44509_peg3714
P6 ^T and DSM 44509 ^T	Oxidative stress	Zinc uptake regulation protein ZUR	P6_peg1798	DSM44509_peg407, DSM44509_peg1351, DSM44509_peg4073
P6 ^T and DSM 44509 ^T	Oxidative stress	bacteriophytochrome heme oxygenase BphO	P6_peg29	DSM44509_peg4013
P6 ^T and DSM 44509 ^T	Oxidative stress	Catalase (EC 1.11.1.6)	P6_peg613	DSM44509_peg1819, DSM44509_peg3715
P6 ^T and DSM 44509 ^T	Oxidative stress	NAD-dependent glyceraldehyde-3-phosphate dehydrogenase (EC 1.2.1.12)	P6_peg874	DSM44509_peg3078
P6 ^T and DSM 44509 ^T	Oxidative stress	NAD-dependent protein deacetylase of SIR2 family	P6_peg1289, P6_peg2601	DSM44509_peg219, DSM44509_peg3132
P6 ^T and DSM 44509 ^T	Oxidative stress	Nicotinamidase (EC 3.5.1.19)	P6_peg366	DSM44509_peg1160, DSM44509_peg2303
P6 ^T and DSM 44509 ^T	Oxidative stress	Nicotinate phosphoribosyltransferase (EC 2.4.2.11)	P6_peg365	DSM44509_peg2304
P6 ^T	Oxidative stress	CoA-disulfide reductase (EC 1.8.1.14)	P6_peg1645	
DSM 44509 ^T	Oxidative stress	Peroxidase (EC 1.11.1.7)		DSM44509_peg3715
DSM 44509 ^T	Oxidative stress	transcriptional regulator, Crp/Fnr family		DSM44509_peg2379

P6 ^T and DSM 44509 ^T	no subcategory	Flavoheomoprotein (Hemoglobin-like protein) (Flavoheomoglobin) (Nitric oxide dioxygenase) (EC 1.14.12.17)	P6_peg3406	DSM44509_peg3484, DSM44509_peg3485
P6 ^T and DSM 44509 ^T	no subcategory	Hemoglobin-like protein HbO	P6_peg1865	DSM44509_peg685
P6 ^T and DSM 44509 ^T	no subcategory	diguanylate cyclase/phosphodiesterase (GGDEF & EAL domains) with PAS/PAC sensor(s)	P6_peg301, P6_peg436, P6_peg521, P6_peg665, P6_peg699, P6_peg726, P6_peg1016, P6_peg1019, P6_peg1054, P6_peg1080, P6_peg1176, P6_peg1199, P6_peg1204, P6_peg1205, P6_peg1216, P6_peg1449, P6_peg1524, P6_peg1682, P6_peg1833, P6_peg1919, P6_peg1934, P6_peg1935, P6_peg2120, P6_peg2153, P6_peg3156, P6_peg3327, P6_peg3329, P6_peg3342, P6_peg3368, P6_peg3376, P6_peg3647, P6_peg3742	DSM44509_peg440, DSM44509_peg460, DSM44509_peg522, DSM44509_peg523, DSM44509_peg538, DSM44509_peg739, DSM44509_peg915, DSM44509_peg916, DSM44509_peg924, DSM44509_peg947, DSM44509_peg1062, DSM44509_peg1369, DSM44509_peg1372, DSM44509_peg1510, DSM44509_peg1787, DSM44509_peg1791, DSM44509_peg1814, DSM44509_peg1826, DSM44509_peg1922, DSM44509_peg2018, DSM44509_peg2150, DSM44509_peg2427, DSM44509_peg2461, DSM44509_peg2476, DSM44509_peg2477, DSM44509_peg2814, DSM44509_peg2873, DSM44509_peg3354, DSM44509_peg3770, DSM44509_peg3968, DSM44509_peg4177, DSM44509_peg4271
P6 ^T and DSM 44509 ^T	no subcategory	Carbon starvation protein A	P6_peg1951	DSM44509_peg975
P6 ^T and DSM 44509 ^T	no subcategory	Carbon storage regulator	P6_peg3731	DSM44509_peg4059
P6 ^T and DSM 44509 ^T	no subcategory	ABC-type Fe ³⁺ -siderophore transport system, permease 2 component	P6_peg780, P6_peg1125	DSM44509_peg709, DSM44509_peg3042
P6 ^T and DSM 44509 ^T	no subcategory	Flavoheomoprotein (Hemoglobin-like protein) (Flavoheomoglobin) (Nitric oxide dioxygenase) (EC 1.14.12.17)	P6_peg3406	DSM44509_peg3484, DSM44509_peg3485
P6 ^T and DSM 44509 ^T	no subcategory	GTP-binding protein HflX	P6_peg208	DSM44509_peg1601
P6 ^T and DSM 44509 ^T	no subcategory	Anti-sigma B factor antagonist RsbV	P6_peg1497	DSM44509_peg715, DSM44509_peg716, DSM44509_peg1537
P6 ^T and DSM 44509 ^T	no subcategory	RNA polymerase sigma factor SigB	P6_peg600, P6_peg2515	DSM44509_peg2957, DSM44509_peg3720
P6 ^T and DSM 44509 ^T	no subcategory	Serine phosphatase RsbU, regulator of sigma subunit	P6_peg602, P6_peg1036, P6_peg1041, P6_peg1190, P6_peg1192, P6_peg1237, P6_peg1322, P6_peg2184, P6_peg2763, P6_peg2953, P6_peg2961, P6_peg3019	DSM44509_peg298, DSM44509_peg490, DSM44509_peg714, DSM44509_peg1766, DSM44509_peg2084, DSM44509_peg2260, DSM44509_peg2460, DSM44509_peg2490, DSM44509_peg2491, DSM44509_peg2564, DSM44509_peg2572, DSM44509_peg2959, DSM44509_peg3627, DSM44509_peg3647, DSM44509_peg4104, DSM44509_peg4267, DSM44509_peg4269
P6 ^T and DSM 44509 ^T	no subcategory	Serine-protein kinase RsbW (EC 2.7.11.1)	P6_peg601	DSM44509_peg2958
DSM 44509 ^T	no subcategory	Putative SigmaB associated two-component system sensor protein		DSM44509_peg2261