

Piezophilic phenotype is growth condition dependent and correlated with the regulation of two sets of ATPase in deep-sea piezophilic bacterium *Photobacterium profundum* SS9

An-qi Li^{1,2,3}, Wei-jia Zhang^{3,4,5*}, Xue-gong Li^{3,4,5}, Xu-chong Bao^{2,3}, Xiao-qing Qi^{3,4,5}, Long-Fei Wu^{4,6} and Douglas Bartlett⁷

- ¹ Institute of Oceanology, Chinese Academy of Sciences, Qingdao 266071, China
- ² University of Chinese Academy of Sciences, Beijing 101408, China
- ³ Laboratory of Deep-Sea Microbial Cell Biology, Institute of Deep-sea Science and Engineering, Chinese Academy of Sciences, Sanya 572000, China
- ⁴ International Associated Laboratory of Evolution and Development of Magnetotactic Multicellular Organisms, CNRS-Marseille/CAS-Sanya, Sanya 572000, China
- ⁵ Institution of Deep-sea Life Sciences, IDSSE-BGI, Hainan Deep-sea Technology Laboratory, Sanya 572000, China
- ⁶ LCB, Aix Marseille University and CNRS, 13402 Marseille, France
- ⁷ Marine Biology Research Division, Scripps Institution of Oceanography, University of California San Diego, La Jolla, CA 92093, USA

* Correspondence: Dr. Wei-Jia Zhang; E-mail: wzhang@idsse.ac.cn; Tel: +86 898 8821 1771.
Laboratory of Deep-Sea microbial cell biology, Institute of Deep-sea Science and Engineering, Chinese Academy of Sciences, Sanya, Hainan 572000, China.

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Table S1. Primers used in this study

Name	Sequence (5'-3')	Description
I-up-F	CGAGCTCTATTTAGCCCTCGCTGGTG	Deletion of <i>atpI</i>
Fusion-I-R	CAGCCATAACTCATCGTATTCGTTCCCTCG	Deletion of <i>atpI</i>
Fusion-I-F	CGAGGAACGAATACGATGAGTTATGGCTG	Deletion of <i>atpI</i>
I-down-R	TGCTCTAGAGTACCAAGGGAAGCAAG	Deletion of <i>atpI</i>
SE1-up-F	CCCCCGGGAGTTATCTTATTTGCCATTGTTTAC	Deletion of <i>atpE1</i>
SE1-up-R	TGCTCTAGATATCTCCAGTTTATAGTTGTCGGC	Deletion of <i>atpE1</i>
SE1-d-F	TGCTCTAGATCAATAGCTGTTTTAACCTTGTGG	Deletion of <i>atpE1</i>
SE1-d-R	TCCCCGCGGCACTGCAATTCAGCTTAACTTTACG	Deletion of <i>atpE1</i>
SE2-up-F	CCCCCGGGAATAAGAGTCACTAGAACGGTCAAG	Deletion of <i>atpE2</i>
SE2-up-R	TGCTCTAGAAATTTTTTATAAATTAAATTAACG	Deletion of <i>atpE2</i>
SE2-d-F	TGCTCTAGATAAATGACTACTGCGGTGATACAG	Deletion of <i>atpE2</i>
SE2-d-R	TCCCCGCGGACTCAACAGTGCGAGAAAG	Deletion of <i>atpE2</i>
Q-I-F	TTAATCGGTGGTGGCATC	RT-PCR analysis
Q-I-R	GGTCAGTTTGAGGGGAAC	RT-PCR analysis
Q-E1-F	GTTTCGCTCTTCTTGGTGG	RT-PCR analysis
Q-E1-R	AGCTAGCTGGGCAACAAATG	RT-PCR analysis
Q-E2-F	ACGTTGCTGGCGCAC	RT-PCR analysis
Q-E2-R	TACCAACACCGATCATTGG	RT-PCR analysis
G3PDH-F	CGGCCTTCTAACAGGTATGG	RT-PCR analysis
G3PDH-R	AGTTAGCGCAACACCAGC	RT-PCR analysis

Table S2. The genetic characterization of ATPase components in SS9 strain

	ATPase-I		ATPase-II		Gene product	Similarity in amino acid sequences (%)
	Gene locus	Length (amino acids)	Gene locus	Length (amino acids)		
<i>atpI</i>	PBPRA3611	129	-	-	Subunit I	-
<i>atpB</i>	PBPRA3610	262	PBPRB0130	255	Subunit a	55.73
<i>atpE</i>	PBPRA3609	86	PBPRB0131	82	Subunit c	44.74
<i>atpF</i>	PBPRA3608	156	PBPRB0132	134	Subunit b	57.48
<i>atpH</i>	PBPRA3607	177	PBPRB0133	182	Subunit δ	44.51
<i>atpA</i>	PBPRA3606	513	PBPRB0134	513	Subunit α	82.85
<i>atpG</i>	PBPRA3605	288	PBPRB0135	291	Subunit γ	68.49
<i>atpD</i>	PBPRA3604	468	PBPRB0136	461	Subunit β	83.12
<i>atpC</i>	PBPRA3603	140	PBPRB0137	147	Subunit ϵ	58.39

Table S3. The growth characteristics of wild-type and mutant strains in MB2216 and MG media

	MB2216 medium				MG medium			
	μ_{\max}		Biomass (OD _{600nm})		μ_{\max}		Biomass (OD _{600nm})	
	0.1 MPa	28 MPa	0.1 MPa	28 MPa	0.1 MPa	28 MPa	0.1 MPa	28 MPa
WT	0.25 ± 0.00	0.25 ± 0.00	0.19 ± 0.01	0.30 ± 0.02	0.04 ± 0.00	0.10 ± 0.01	0.42 ± 0.01	0.54 ± 0.01
<i>$\Delta atpI$</i>	0.26 ± 0.03	0.31 ± 0.02	0.18 ± 0.00	0.35 ± 0.00	0.03 ± 0.00	0.12 ± 0.01	0.61 ± 0.01	0.75 ± 0.02
<i>$\Delta atpE1$</i>	0.22 ± 0.01	0.28 ± 0.00	0.15 ± 0.01	0.34 ± 0.01	0.02 ± 0.00	0.08 ± 0.00	0.59 ± 0.00	0.66 ± 0.01
<i>$\Delta atpE2$</i>	0.29 ± 0.01	0.33 ± 0.00	0.20 ± 0.01	0.30 ± 0.01	0.03 ± 0.01	0.19 ± 0.01	0.43 ± 0.01	0.60 ± 0.01

Table S4. Effect of pressure on the expression of ATPase genes

		WT	$\Delta atpI$	$\Delta atpE1$	$\Delta atpE2$
	<i>atpI</i>	1.47 ± 0.08 ^{#, **}	-	1.35 ± 0.25	0.74 ± 0.13 ^{***}
MB2216	<i>atpE1</i>	0.90 ± 0.07	1.50 ± 0.13 ^{**}	-	0.97 ± 0.02
	<i>atpE2</i>	1.22 ± 0.11	0.81 ± 0.10 [*]	1.35 ± 0.20 [*]	-
	<i>atpI</i>	0.26 ± 0.01 ^{***}	-	1.95 ± 0.16 ^{**}	0.45 ± 0.04 ^{***}
MG	<i>atpE1</i>	0.40 ± 0.01 ^{***}	0.92 ± 0.03	-	0.43 ± 0.05 ^{***}
	<i>atpE2</i>	1.32 ± 0.14	1.68 ± 0.04 ^{***}	0.86 ± 0.05	-

[#] The values show the relative expression at 28 MPa normalized to that at 0.1 MPa.

All data are shown as mean ± standard deviation. The statistical analysis of comparisons between two groups was performed using an unpaired-sample *t* test, a two-tailed *p* < 0.05 was considered significant, *, *p* < 0.05; **, *p* < 0.01; ***, *p* < 0.001.

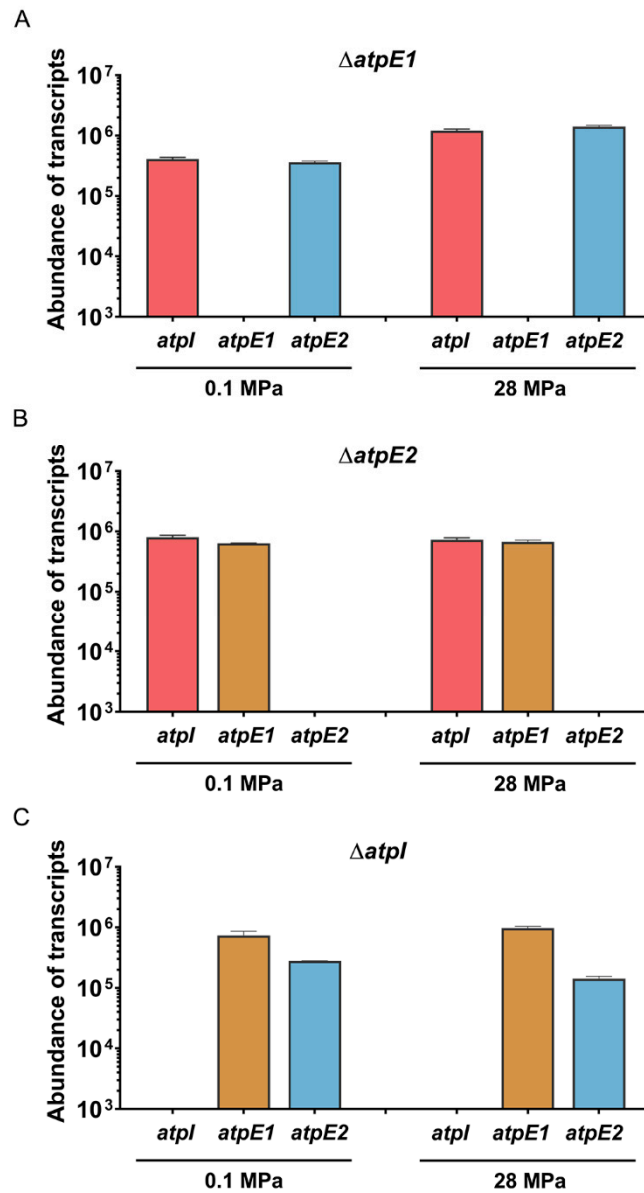


Figure S1. Abundance of ATPase gene transcripts in mutant strains grown in MB2216 medium. Panel A, B and C show the quantification of ATPase genes in $\Delta atpE1$, $\Delta atpE2$ and $\Delta atpI$ mutants, respectively. The genes analyzed and pressure condition are marked below the bars.

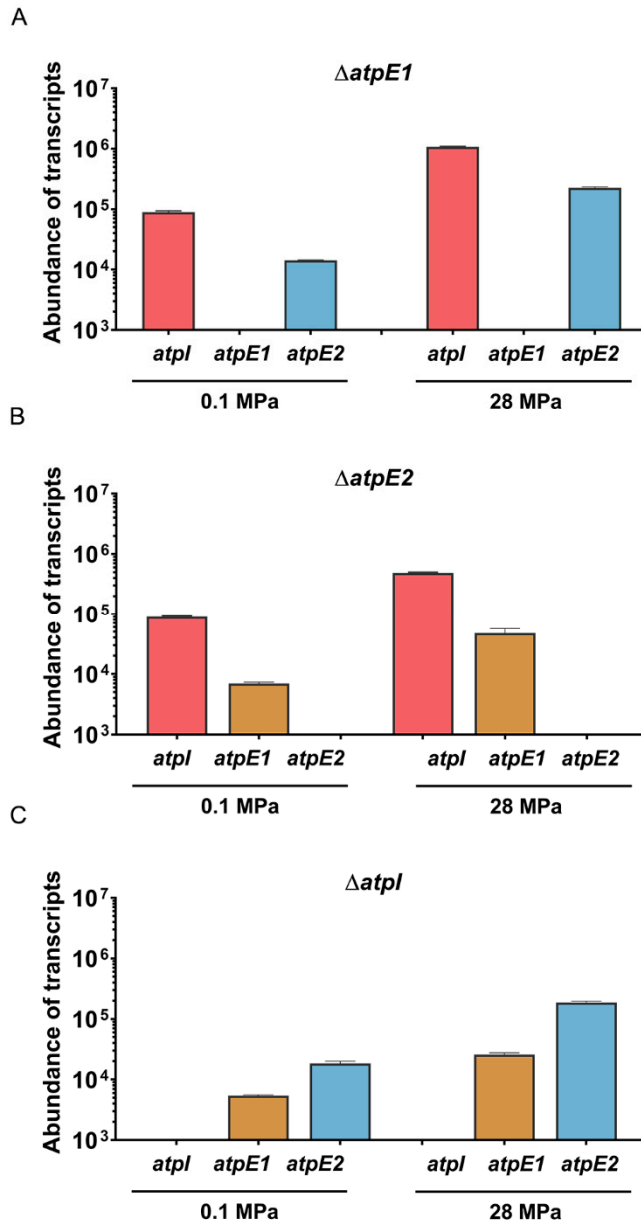


Figure S2. Abundance of ATPase gene transcripts in mutant strains grown in MG medium. Panel A, B and C show the quantification of ATPase genes in $\Delta atpE1$, $\Delta atpE2$ and $\Delta atpI$ mutants, respectively. The genes analyzed and pressure condition are marked below the bars.