

Supplementary Material

Table S1. Genomic features of different *B. megaterium* strains.

Strain	Genome size (Mbp)	G+C content (%)	Number of coding sequences (CDS)	Number of RNA genes	Reference
LVN01	5.22	37.8	4846	21	This work
QM B1551	5.52	38.2	5710	176	(Eppinger <i>et al.</i> , 2011)
DSM 319	5.09	38.1	5323	148	(Eppinger <i>et al.</i> , 2011)
WSH-002	5.07	38.2	5330	149	(Liu <i>et al.</i> , 2011))
SF 185	5.05	38.1	5346	138	(Di Luccia <i>et al.</i> , 2016)
Q3	5.23	38.2	5370	171	(Liu <i>et al.</i> , 2014)
RIT 381	5.86	37.6	6083	193	(Polter <i>et al.</i> , 2015)
BMS	5.62	37.7	5858	153	(Daligault <i>et al.</i> , 2014)
PE5-112	5.43	38.2	5604	168	(van Zyl <i>et al.</i> , 2016)
BGH1.1	6.26	37.3	6528	130	(Wang <i>et al.</i> , 2016))
NBCR 15308	5.75	37.8	5970	166	(Arya <i>et al.</i> , 2014))
MSP20.1	4.37	36.5	4207	66	(Pal <i>et al.</i> , 2014)

Table S2. PHB content in *B. megaterium* LVN01 and *B. megaterium* BmGD.

Time (h)	PHB Content (mg _{PHB} / g _{cell})	
	<i>B. megaterium</i> BmGD	<i>B. megaterium</i> LVN01
6	2.8	5.0
24	24.6	39.2
30	80.5	54.6
48	64.3	58.1
52	30.9	23.4

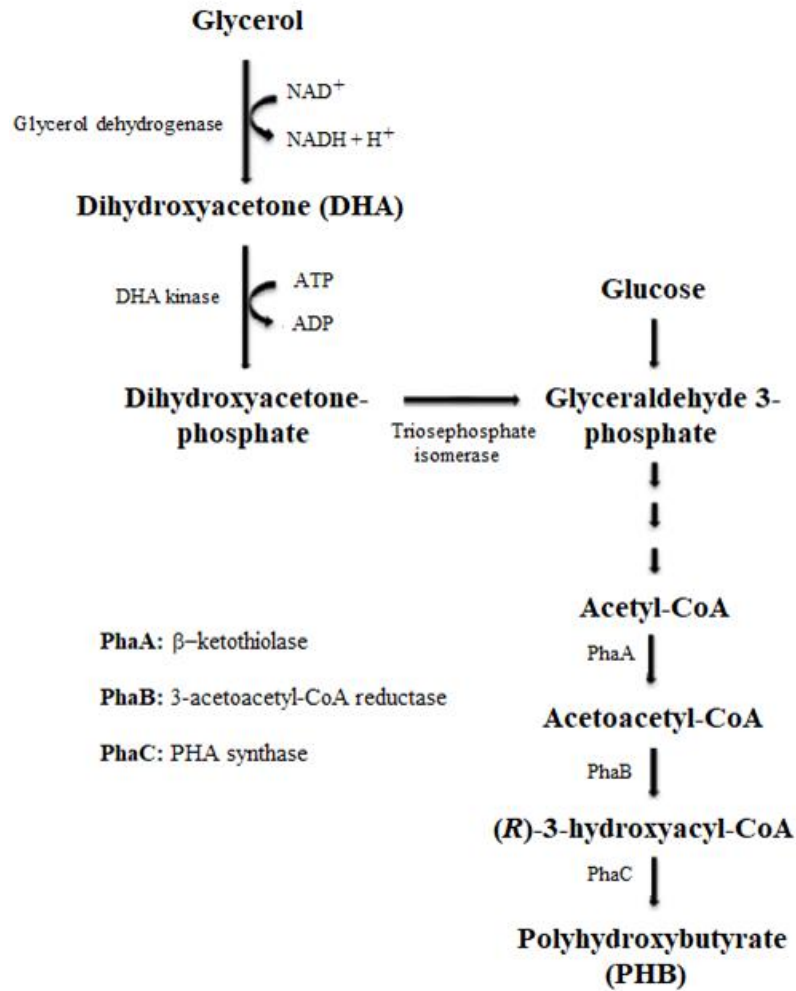


Figure S1. Synthesis of PHA by *B. megaterium*. Adapted from Mozejko-Ciesielska and Kiewisz, 2016.

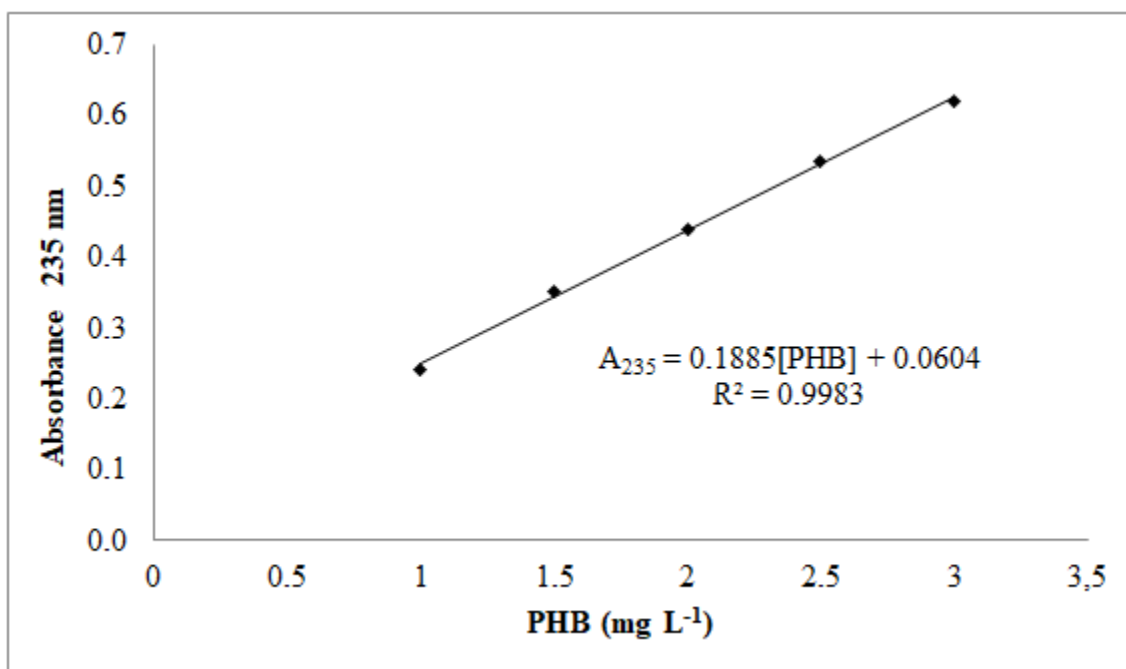


Figure S2. Calibration curve for PHB quantification.

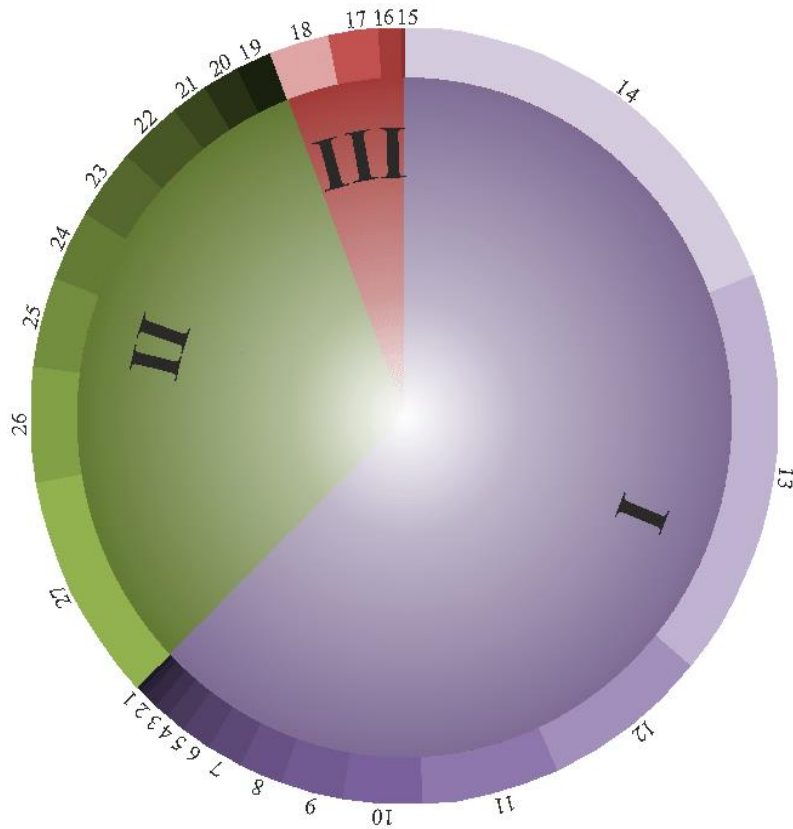


Figure S3. Distribution of subsystem categories in *B. megaterium* LVN01 according to RAST (Aziz *et al.*, 2008). Subsystems were clustered in three main groups named **(I)** metabolism, **(II)** cellular processes and signaling, and **(III)** information and others. Subsystems: **(1)** Iron acquisition and metabolism, **(2)** secondary metabolism, **(3)** potassium, **(4)** DNA metabolism, **(5)** nitrogen, **(6)** sulfur, **(7)** aromatic compounds, **(8)** phosphorous, **(9)** nucleosides and nucleotides, **(10)** respiration, **(11)** fatty acids, lipids and isoprenoids, **(12)** protein, **(13)** amino acids and derivatives, **(14)** carbohydrates, **(15)** phages, prophages, transposable elements, plasmids, **(16)** miscellaneous, **(17)** DNA, **(18)** RNA, **(19)** dormancy and sporulation, **(20)** cell division and cell cycle, **(21)** regulation and cell signaling, **(22)** motility and chemotaxis, **(23)** virulence, disease and defense, **(24)** membrane transport, **(25)** cell wall and capsule, **(26)** stress response, and **(27)** cofactor, vitamins, prosthetic groups and pigments.

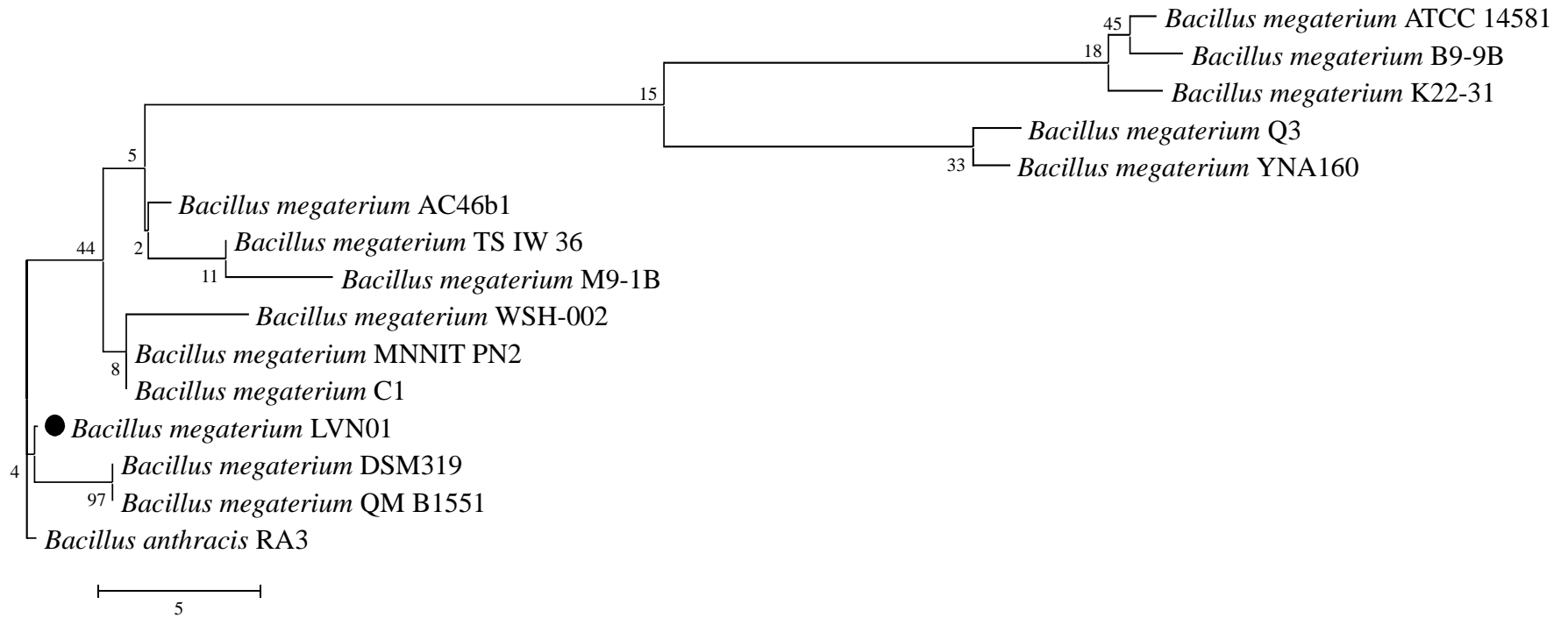


Figure S4. Dendrogram of *B. megaterium* strains. Analysis of genes by MEGA6 (Tamura *et al.*, 2013) was employed for conducting sequence alignment and inferring phylogenetic trees, employing Maximum Likelihood as statistical method, Jones-Taylor-Thornton as substitution model and Nearest-Neighbor-Interchange as Maximum Likelihood heuristic method. The test of phylogeny considered the bootstrap method (500 bootstrap replications).

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BmGDLVN01      1 MRKAFISPSKYIQGENEILNLGYFVKTFGT SALLIAHPEDIKRVQDKLDATEAEKYGITFFEGGFNGECSRPEISRLOEIA 80
BmGDWSH-002    1 MRKAFISPSKYIQGEDEILNLGYFVKTFGT SALLIAHPEDIKRVQDKLDATEAEKYGITFFEGGFNGECSRPEISRLOEIA 80
BmGDQM B1551   1 MRKAFISPSKYIQGENEILNLGYFVKTFGT SALLIAHPEDIKRVQDKLDATEAEKYGITFFEGGFNGECSRPEISRLOEIA 80

BmGDLVN01     81 KENNCDCITIGLGGGKAIDTAKCVAEAGEGLIIVPTIAATDAPTS HSAVIYTPGAFDDYAYFKQSPSVVLIDTTVIANAPT 160
BmGDWSH-002   81 KENNCDCITIGLGGGKAIDTAKCVAEAGEGLIIVPTIAATDAPTS HSAVIYTPGAFDDYAYFKQSPSVVLIDTTVIANAPT 160
BmGDQM B1551  81 KENNCDCITIGLGGGKAIDTAKCVAEAGEGLIIVPTIAATDAPTS HSAVIYTPGAFDDYAYFKQSPSVVLIDTTVIANAPT 160

BmGDLVN01    161 RFLVSGMGDALSTYFEARATARSFSNVNAGLPCGVREDLCAPAKGTNAALVLA KHCYNTLLEDGVKAKAASDHNVVPAL 240
BmGDWSH-002  161 RFLVSGMGDALSTYFEARATARSFSNVNAGLPCGVREDLCAPAKGTNAALVLA EHCYNTLLEDGVKAKAASDHNVVPAL 240
BmGDQM B1551  161 RFLVSGMGDALSTYFEARATARSFSNVNAGLPCGVREDLCAPAKGTNAALVLA KHCYNTLLEDGVKAKAASDHNVVPAL 240

BmGDLVN01    241 ENIIEANILLSGLGFESGGLAGAHAIHDGLTLLLESAHYFHGEKVAFGTLAQLVLENAPTEEIEEVLD FCLAVGLPVCLA 320
BmGDWSH-002  241 ENIIEANILLSGLGFESGGLAGAHAIHDGLTLLLESAHYFHGEKVAFGTLAQLVLENAPTEEIEEVLD FCLAVGLPVCLA 320
BmGDQM B1551  241 ENIIEANILLSGLGFESGGLAGAHAIHDGLTLLLESAHYFHGEKVAFGTLAQLVLENAPTEEIEEVLD FCLAVGLPVCLA 320

BmGDLVN01    321 DIGVEQITQEELMEVANKACIPEESIYSMPFFITPESVAAAI IAADQIGNDYKKRLI 377
BmGDWSH-002  321 DIGVEQITQEELMEVANKACIPEESIYSMPFFVNPESVAAAILA ADQIGNDYKKRLI 377
BmGDQM B1551  321 DIGVEQITQEELMEVANKACIPEESIYSMPFFITPESVAAAI IAADQIGNDYKKRLI 377

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Figure S5. A multiple sequence alignment of glycerol dehydrogenases from *B. megaterium* strains. Non-conserved residues among the sequences are highlighted in red. BmGDLVN01: glycerol dehydrogenase from *B. megaterium* LVN01, BmGDWSH-002: glycerol dehydrogenase from *B. megaterium* WSH-002, BmGDQM B1551: glycerol dehydrogenase from *B. megaterium* QM B1551.

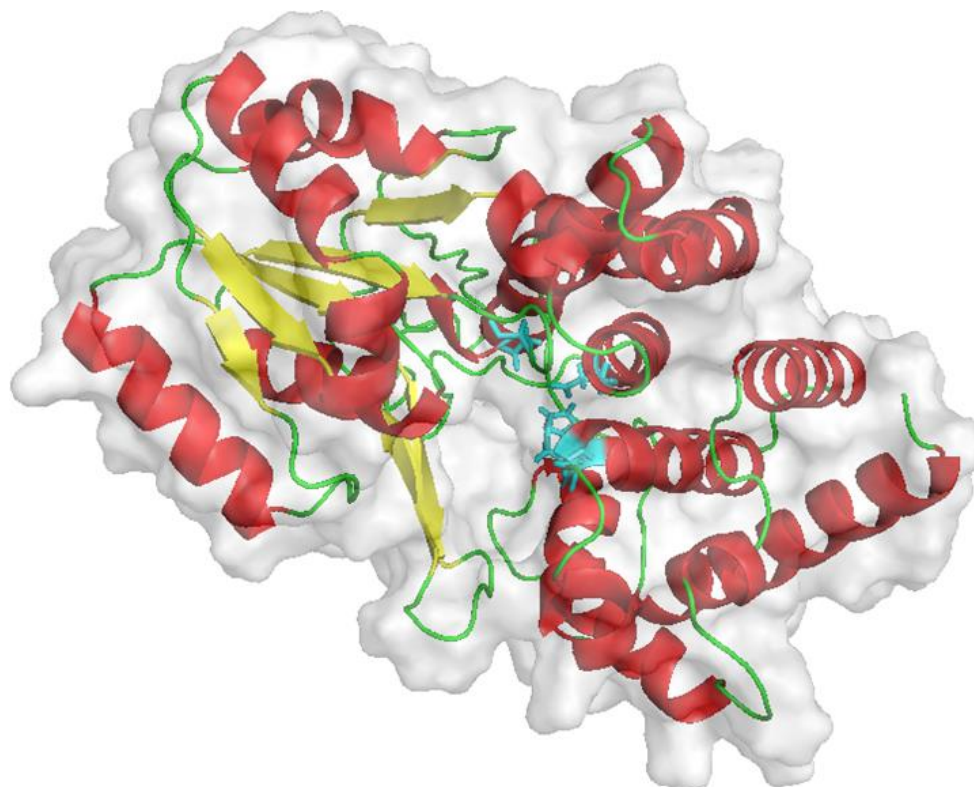


Figure S6. Model for a monomer of *BmGD* showing its catalytic pocket. The glycerol binding site is highlighted in cyan (clockwise direction, from top left to bottom right D119, D169, H264 and H281).

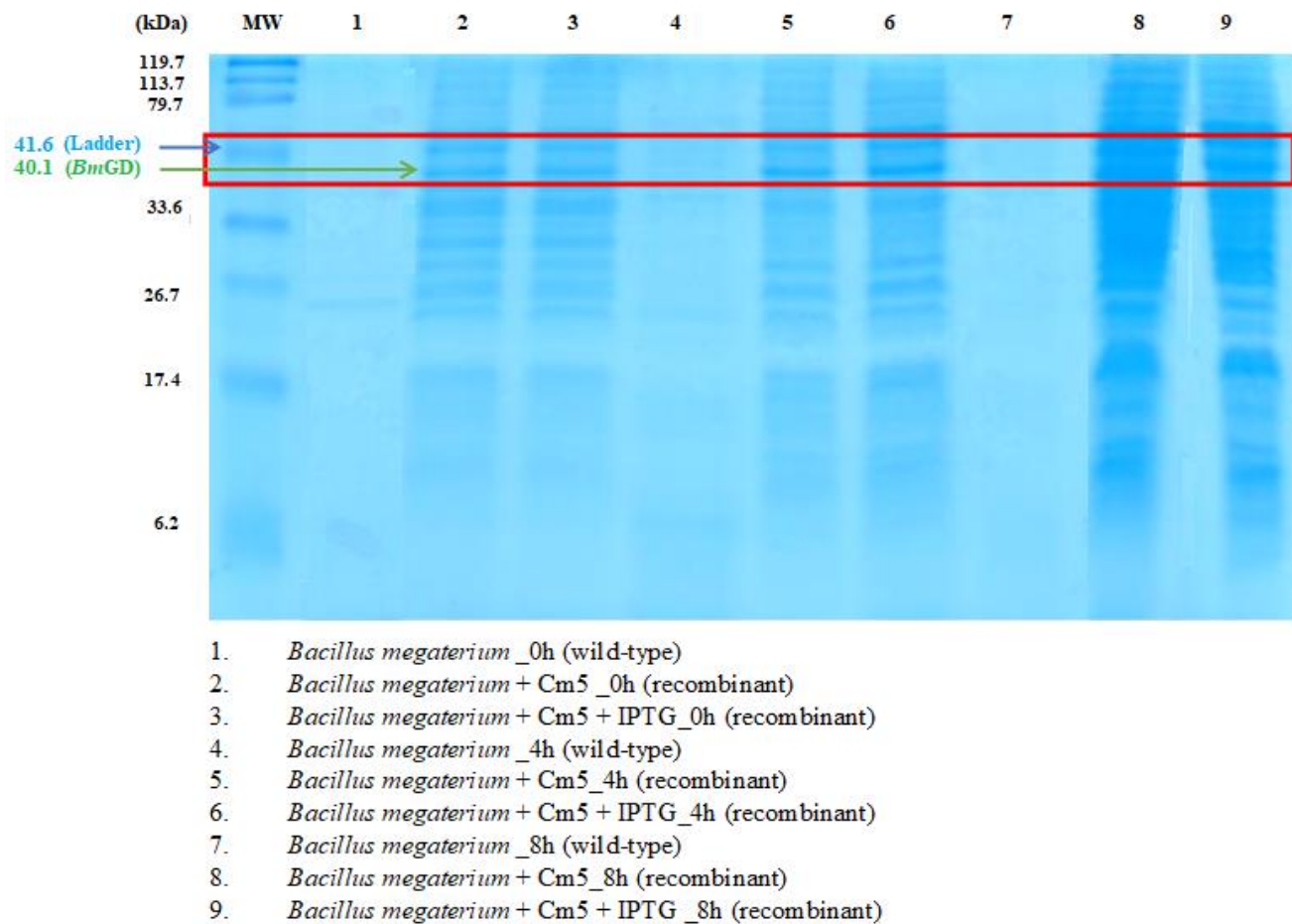


Figure S7. SDS-PAGE analysis of whole-cell extracts of *B. megaterium* LVN01 (wild-type) and *B. megaterium* carrying pHT01-*bmgd* (recombinant) without and with IPTG induction. The green arrow to the left indicate the position of *BmGD* enzyme. Cm5: 5.0 $\mu\text{g mL}^{-1}$ Chloramphenicol

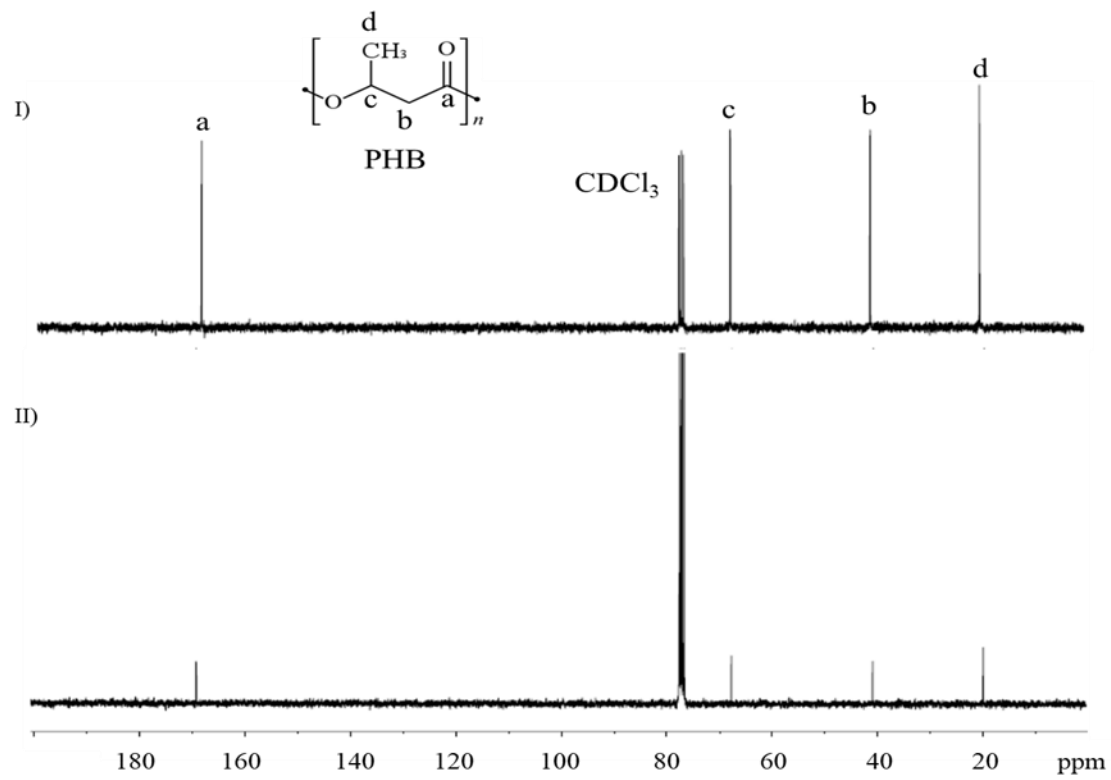


Figure S8. Spectrum 300 MHz ^{13}C -NMR of (I) commercial PHB compared to (II) the PHB spectrum obtained from *B. megaterium* BmGD.

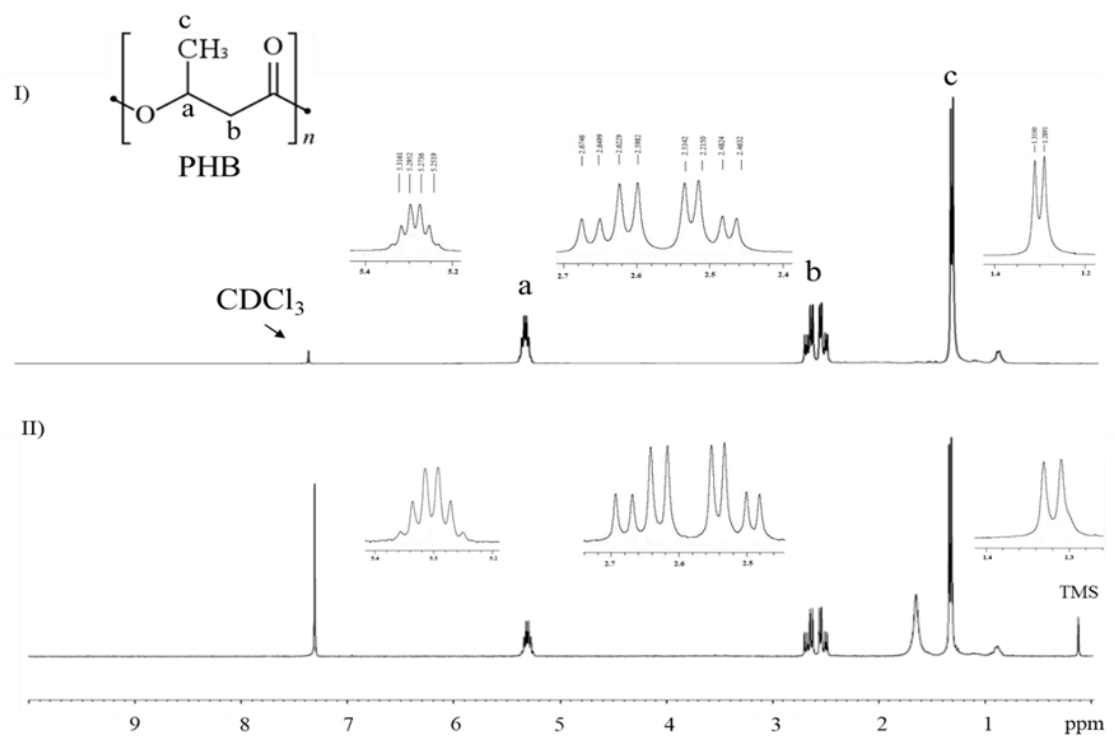


Figure S9. Spectrum 300 MHz ¹H-NMR of **(I)** commercial PHB compared to **(II)** the PHB spectrum obtained from *B. megaterium* BmGD.

References

- Arya, G., Petronella, N., Crosthwait, J., Carrillo, C. D., & Shwed, P. S. 2014. Draft genome sequence of *Bacillus megaterium* type strain ATCC 14581. *Genome Announcements* 2(6): e01124-14.
- Aziz, R. K., Bartels, D., Best, A. A., DeJongh, M., Disz, T., Edwards, R. A., Formsma, K., Gerdes, S., Glass, E. M., Kubal, M., Meyer, F., Olsen, G. J., Olson, R., Osterman, A. L., Overbeek, R. A., McNeil, L. K., Paarmann, D., Paczian, T., Parrello, B., Pusch, G. D., Reich, C., Stevens, R., Vassieva, O., Vonstein, V., Wilke, A. & Zagnitko, O. 2008. The RAST Server: Rapid Annotations using Subsystems Technology. *BMC Genomics* 9(1): 75.
- Daligault, H. E., Davenport, K. W., Minogue, T. D., Bishop-Lilly, K. A., Broomall, S. M., Bruce, D. C., Chain, P. S., Coyne, S. R., Frey, K. G., Gibbons, H. S., Jaissle, J., Koroleva, G. I., Ladner, J. T., Lo, C.-C., Munk, C., Palacios, G. F., Redden, C. L., Rosenzweig, C. N., Scholz, M. B., & Johnson, S. L. 2014. Twenty whole-genome *Bacillus* sp. assemblies. *Genome Announcements* 2(5): e00958-14.
- Di Luccia, B., D'Apuzzo, E., Varriale, F., Baccigalupi, L., Ricca, E., & Pollice, A. 2016. *Bacillus megaterium* SF185 induces stress pathways and affects the cell cycle distribution of human intestinal epithelial cells. *Beneficial Microbes* 7(4): 609–620.
- Eppinger, M., Bunk, B., Johns, M. A., Edirisinghe, J. N., Kutumbaka, K. K., Koenig, S. S. K., Creasy, H. H., Rosovitz, M. J., Riley, D. R., Daugherty, S., Martin, M., Elbourne, L. D. H., Paulsen, I., Biedendieck, R., Braun, C., Grayburn, S., Dhingra, S., Lukyanchuk, V., Ball, B., Ul-Qamar, R., Seibel, J., Bremer, E., Jahn, D., Ravel, J. & Vary, P. S. 2011. Genome sequences of the biotechnologically important *Bacillus megaterium* strains QM B1551 and DSM319. *Journal of Bacteriology* 193(16): 4199–4213.
- Liu, L., Li, Y., Zhang, J., Zou, W., Zhou, Z., Liu, J., Li, X., Wang, L., & Chen, J. 2011. Complete genome sequence of the industrial strain *Bacillus megaterium* WSH-002. *Journal of Bacteriology* 193(22): 6389–6390.
- Liu, M., Luo, K., Wang, Y., Zeng, A., Zhou, X., Luo, F., & Bai, L. 2014. Isolation, identification and characteristics of an endophytic quinclorac degrading bacterium *Bacillus megaterium* Q3. *PLoS ONE*, 9(9): e108012.
- Pal, K. K., Dey, R., Sherathia, D., Vanpariya, S., Patel, I., Dalsania, T., Savsani, K., Sukhadiya, B., Mandaliya, M., Thomas, M., Ghorai, S., Rupapara, R., Rawal, P., Shah, & A., Bhayani, S. 2014. Draft genome sequence of a moderately halophilic *Bacillus megaterium* strain, MSP20.1, isolated from a saltern of the little Rann of Kutch, India. *Genome Announcements* 2(1): e01134-13.
- Polter, S. J., Caraballo, A. A., Lee, Y. P., Eng, W. W. H., Gan, H. M., Wheatley, M. S., Savka, M. A., Thomas, B. N., & Hudson, A. O. 2015. Isolation, identification, whole-genome sequencing, and annotation of four *Bacillus* species, *B. anthracis* RIT375, *B. circulans* RIT379, *B. altitudinis* RIT380, and *B. megaterium* RIT381, from internal stem tissue of the insulin plant *Costus igneus*. *Genome Announcements* 3(4): e00847-15.
- Tamura, K., Stecher, G., Peterson, D., Filipski, A., & Kumar, S. 2013. MEGA6: Molecular Evolutionary Genetics Analysis version 6.0. *Molecular Biology and Evolution* 30(12): 2725-2729.
- van Zyl, L. J., Matobole, R., Augustin Nsole Biteghe, F., Klein, T., Kirby, B., & Trindade, M. 2016. Draft genome sequences of three *Bacillus* species from South African marine sponges. *Genome Announcements* 4(2): e00143-16.
- Wang, W., Zheng, S.-S., Sun, H., Cao, J., Yang, F., Wang, X.-L., & Li, L.-X. 2016. Draft genome sequence of *Bacillus megaterium* BHG1.1, a strain isolated from bar-headed goose (*Anser*

indicus) feces on the Qinghai-Tibet Plateau. *Genome Announcements* 4(3): e00317-16.