



Supplement of

Reviews and syntheses: Heterotrophic fixation of inorganic carbon – significant but invisible flux in environmental carbon cycling

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29 **Table S1:** Global standing stock of organic carbon in living biomass and contribution from anaplerotic CO₂
 30 fixation (only anaplerosis is considered here; other mechanisms of heterotrophic CO₂ fixation were neglected).
 31 In heterotrophs, a conservative estimate of 1-5% of the cell carbon is assumed to originate from inorganic
 32 carbon fixation (see references in text).

Continental habitats	Carbon biomass [Pg C]	Carbon in biomass derived from anaplerotic CO ₂ fixation [Pg C]	References for carbon biomass
Terrestrial animals	0.6	0.006 – 0.03	(Bar-On et al. 2018)
Soil fungi	12	0.12 – 0.6	(Bar-On et al. 2018)
Terrestrial protists	1.6	0.016 – 0.8	(Bar-On et al. 2018)
Soil prokaryotes (upper 100 cm of soil)	23.2	0.23 – 1.16	(Xu, Thornton and Post 2013)
Continental subsurface prokaryotes	2.4 – 12.6*	0.024 – 0.63	(Magnabosco et al. 2018)
Heterotrophic prokaryotes in freshwater and saline inland surface waters	0.013**	0.00013 – 0.00065	(Whitman et al. 1998)
Marine and oceanic habitats			
Marine Animals	2	0.02 – 0.1	(Bar-On et al. 2018)
Marine protists	2	0.02 – 0.1	(Bar-On et al. 2018)
Marine fungi	0.3	0.003 – 0.015	(Bar-On et al. 2018)
Marine planktonic heterotrophic prokaryotes	1.4 – 3.5***	0.014 – 0.175	(Whitman et al. 1998)
Subseafloor sedimentary prokaryotes	1.5 – 22	0.015 – 1.1	(Kallmeyer et al. 2012, Schippers et al. 2005)
Prokaryotes of the oceanic crust	0.5 – 5	0.005 – 0.25	(Bar-On et al. 2018)
Total heterotrophic carbon biomass	47 – 85	0.47 – 4.96	

33
 34 * Cell abundances ($2 – 6 \times 10^{29}$ cells) from Magnabosco et al. (2018) were converted into cell carbon using the
 35 carbon conversion factors 12 fg C cell⁻¹ and 21 fg C cell⁻¹ (Wilhartz et al. 2009, Griebler et al. 2002) for the
 36 minimum and maximum values of the range, respectively. In favor of a conservative estimate, quite low carbon
 37 conversion factors were used (at the lower end of the carbon content values for freshwater prokaryotic cells
 38 reported in literature).

39
 40 ** Cell abundance (2.3×10^{26} cells) from Whitman et al. (1998) were converted into cell carbon using a carbon
 41 conversion factor of 57 fg C cell⁻¹, which is the arithmetic mean of the minimum and maximum of a range of
 42 values (6 to 107 fg C cell⁻¹) reported for freshwater lakes and rivers of different trophic states in literature
 43 (Pedrós-Alió and Brock 1982, Bjørnsen 1986, Simon 1987, Lever et al. 2015).

44
 45 *** Cell abundances were converted into cell carbon using the carbon conversion factors 12 fg C cell⁻¹ and
 46 30 fg C cell⁻¹ (Fukuda et al. 1998) for the minimum and maximum values, respectively.

49 **Table S2:** Annual global heterotrophic carbon biomass production and contribution from heterotrophic CO₂
 50 fixation (via anaplerosis).

	Annual heterotrophic C-biomass production [Pg C yr⁻¹]	Anaplerotically fixed carbon [Pg C yr⁻¹] [§]	
<u>Marine and oceanic habitats</u>			
Marine and freshwater	2.4 – 76 [*]	0.024 – 3.8	(Cole, Findlay and Pace 1988, del Giorgio and Duarte 2002)
Oceanic subseafloor	0.1 – 9.8 ^{**}	0.001 – 0.49	(Schippers et al. 2005)
<u>Continental habitats</u>			
Aquifers and unsaturated subsurface	0.12 – 26.3 [†]	0.0012 – 1.315	(Magnabosco et al. 2018, Griebler et al. 2014)
Soils	31.3 – 133.2 ^{††}	0.313 – 6.66	(Prentice et al. 2001, Manzoni et al. 2012, Hashimoto et al. 2015, Potter and Klooster 1998)
Total heterotrophic C-biomass production	34 – 245	0.34 – 12.3	

51
 52 * Bacterial carbon production (BCP) rates from 54 marine and freshwater studies (Cole et al. 1988) were
 53 converted from [mg C m⁻²d⁻¹] into [Pg C yr⁻¹] and extrapolated to global scale using a world water surface area
 54 of 361,419,000 km² (<http://www.worldatlas.com/aatlas/infopage/oceans.htm>).

55
 56 ** The total number of living cells [1.3 x 10²⁹] was divided by the turnover time of subseafloor bacteria [0.25-22
 57 yrs], multiplied by the mean carbon content per cell [19 fg C], and converted from [fg C] to [Pg C]. All data as
 58 given in Schippers et al. (2005).

59
 60 [†] The range of bacterial carbon production rates [fg C L⁻¹ yr⁻¹] from 14 groundwater wells (sampled in spring and
 61 autumn) located in an oligotrophic porous aquifer in the Bavarian Alps (close to Mittenwald in Southern
 62 Germany) was divided by the corresponding bacterial abundance [cells L⁻¹] to obtain BCP rates per cell (data
 63 from Griebler et al. 2014). The minimum and the maximum values of these cell-specific BCP rates were then
 64 multiplied by the minimum and the maximum estimated total number of prokaryotes in the continental
 65 subsurface [2-6 x 10²⁹ cells] from Magnabosco et al. (2018), respectively, and carbon mass units were
 66 converted from [fg] to [Pg]. Note: since comprehensive, global data on microbial carbon production in aquifers
 67 are currently still missing, the level of uncertainty of this estimate is high. Therefore, in order to avoid
 68 overestimation, and in favor of obtaining a most conservative estimate, we selected out of the available data
 69 only those production rates, which were determined in pristine, highly oligotrophic environments. If all other
 70 data from the dataset in Griebler et al. (2014), in total 88 wells throughout Germany, sampled twice, as well
 71 as the data from four other available studies with sites in the USA, Austria and Denmark (Thorn and Ventullo
 72 1988, Kazumi and Capone 1994, Albrechtsen and Winding 1992, Wilhartz et al. 2009) were to be included, a
 73 much higher estimate of the global annual heterotrophic carbon biomass production in aquifers would be
 74 obtained, ranging from 0.06 to 4,829 Pg C yr⁻¹, and corresponding to 0.001 – 386 Pg C yr⁻¹ of anaplerotically
 75 fixed carbon each year.

76
 77 ^{††} Global terrestrial heterotrophic respiration in soils [55 Pg C yr⁻¹] from Prentice et al. (2001) was extrapolated
 78 to carbon biomass production assuming that respiration accounts for 30-62% of the total carbon consumed

79 (corresponding to a carbon use efficiency (CUE) of 38-70%) in the course of organic matter decomposition in
80 different types of soils (Manzoni et al. 2012).

81
82 [§] It was assumed that 1-5% of the annually produced carbon biomass of heterotrophs originate from anaplerotic
83 CO₂ fixation (see ref. in the text of the main MS). A fraction of 1% was applied to the minimum, and 5% to the
84 maximum value of the C-biomass production ranges in this table, respectively.

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