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New Global Species Biodiversity: Soil soars, Ocean flounders

Robert J. Blakemore PhD

VermEcology, 101 Suidomichi, Nogeyama, Kanagawa-ken 231-0064, Japan. C/- Kanagawa Prefectural Museum of Natural History, Odawara, Kanagawa-ken 247-0007. Email: <u>rob.blakemore@gmail.com</u> . Updated 25th Sept., 2022.

ABSTRACT

Based on topographic field data, an argument is advanced that Soil houses ~2.1 x 10^{24} taxa and supports >99.9% of global species biodiversity, mostly Bacteria or other microbes. Contradictory claims that Soil is home to only a quarter of biota while Ocean harbours 80–99% of Life on Earth are both dismissed. Earlier guesstimates of 8.8 million taxa (2.2 million or 25% marine), of 1–6 billion, or over a trillion species worldwide are likely underestimations. Recent studies show >10¹² microbial OTU (just 10¹⁰ or <1% in Ocean) soon raised to 10¹⁴ and speculated as high as 10^{23} species. Scaling of simple topsoil samples herein ups total to 2.1 x 10^{24} or 20x. Biomass at 2 x 10^{-13} g/cell of 2.1 x 10^{30} soil cells = 4 x 10^{17} g or 400 Gt (with 200 Gt Carbon, 48 Gt Nitrogen).

An Addendum estimates microbes in gut of two key soil taxa: Megadrile earthworms $(10^{21}-10^{30}$ cells) and termites $(10^{22}-10^{23}$ cells) that strictly add to Soil's totals. Although ranges are large, it is unclear how much 65 x 10^{15} earthworms contribute to Earth's soils count of ~2.1 x 10^{30} cells.

INTRODUCTION

An honest inventory of our global biotic stock is vitally important in order to estimate biodiversity and track extinctions. False claims that soil has "*more than 25% of our planet's biodiversity*" (<u>Ref</u>.) and simultaneously ocean has 80% (<u>Ref</u>.) or "97 percent of life in the world, maybe in the universe" (<u>Ref</u>.) or "99% of the habitable space on this planet" (<u>Ref</u>.) are clearly misguided.

Species	Earth			Otean		
	Catalogued	Fredicted	±SE	Catalogued	Predicted	:50
Eukaryotes	1000 C 1000 C		21010205	A CONTRACT	a second second	
Animula	953,494	2,770.000	958,000	375,082	2,1542,6840	145,000
Chipmiste	13,035	27,598	30.509	4.859	7,498	\$1,640
Foregi	43,271	811,000	2907,000	1,097	8,835	11,100
Plantae	315.644	299,000	8.200	8,690	16.009	9,130
Protossia	6,118	35,438	6,690	8,118	30,435	6,090
Tenir	6,288,000	8,740,000	1,300,000	181,754	2,210,000	182,000
Prokaryokes						
Archave	562	455	166	.W.1	0 C	00
Bacteria	10,898	8.682	8,470	812	1,820	436
Foruit	10.890	10.100	3.630	451	1.330	436
Grand Total	1,244,200	8,756,009	1,300.000	104.409	3,210,060	182,000



In a highly biased survey range of 3–100 million taxa (10^6-10^8) , Mora *et al.* (2011: tab. 2) chose a mere ~8.8 million (~2.2 million or 25% marine). Whereas Larsen *et al.* (2017) proposed a new Pie of Life projected for >1–6 billion (10^9) species on Earth dominated by Bacteria (~70–90% of total) which they mainly considered for insect hosts. Mundanely, they also found on average six cryptic species per morphologically described arthropod taxon (their <u>tab. S1</u>) quite counterbalancing approximately 20% of published eukaryote names that are synonyms (<u>Ref.</u>).

Bahram *et al.* (2018) conclude soils are Earth's most diverse biomes but fail to give figures. For soil's 3 x 10^{29} cells Flemming & Wuertz (2019) and Bar-On & Milo (2019) give no species data.

Subsequently, Louca *et al.* (2019) claimed only "*about 2.2–4.3 million full-length OTUs* [unique taxa] *worldwide*" (10^6) refuting predictions that billions or trillions of prokaryotic OTUs exist. Wiens (2021) explained how Louca *et al.* (2019) had made entirely avoidable underestimation errors whilst also revising Larsen *et al.*'s (2017) projected 1–6 billion estimate downwards to 0.183 to 4.2 billion (10^8 – 10^9) species with 58–88% Bacteria, again most of these in insect hosts.

High-throughput genomic sequencing and bioinformatics studies allow scaling values based on Locey & Lennon (2016: fig. 3 below) showing Earth with ~10¹² microbial OTU taxa (just 10¹⁰ or ~1% in global Ocean). These totals were later raised to 10^{12} – 10^{14} microbial taxa by Lennon & Locey (2020) and then by Fishman & Lennon (2022) who had "*a soft upper constraint of 10^{22}–10^{23} due to neutral drift*". Most of these taxa at any time are likely dormant (Ref.) and/or unculturable as fewer than 1% of soil species are culturable (Schloss & Handelsman, 2006), or possibly as low as only 0.001% to 0.1% (Ref.). So all bacterial species counts are likely minima.



Fig. 3. The microbial richness-abundance scaling relationship (dashed red

Figure 2. From Locey & Lennon (2016: fig. 3) with Earth's >10¹² taxa (just 10¹⁰ or <1% Oceanic).

However, to date, none of the estimates of terrestrial biodiversity have considered terrain that may easily double soil surface area (Blakemore, <u>2018</u>b). All estimates based upon planimetrically flat land areas are manifestly deficient since land is hilly and soil is bumpy. Thus this study aims to estimate soil microbial totals taking terrain into consideration for the first time.

As Blakemore (2018b) stated: "A single gramme (~1 cm³) of fertile topsoil may have three billion microbes (Bacteria, Actinomycetes, Archaea, Fungi, Protozoa, etc.), up to 60 km of fungal hyphae, with 10,000 to 50,000 microbial species having 1,598 km of DNA some dating to the beginning of life four billion years ago." Average soil carbon is ~1–2% and global soil organic carbon (SOC) alone amounts to >10,000 Gt, thus if one gram of soil has >10,000 spp, its total biodiversity must be truly astronomical and lower values or figures above are likely most modest.

METHODS

Two main steps to determine the status of soil biota are to review sample surveys to obtain a consensus per gram then to determine the soil matrix extent to give an overall estimate of totals.

How many species per gram or tonne of soil?

Curtis *et al.* (2002) stated: "the entire bacterial diversity of the sea may be unlikely to exceed 2 x 10^6 , while a ton of soil could contain 4×10^6 different taxa" (or 4 million taxa) which seems a substantial underestimation error as "species of bacteria per gram of soil vary between 2,000 and 8.3 million" (Gans *et al.*, 2005; Roesch *et al.* 2007) (= 10^4 – 10^6 spp/g or 10^{10} – 10^{12} spp/t that, if all unique taxa, is equivalent to twenty billion up to a trillion spp per ton of topsoil). [Discrepancies in Gans *et al.* are samples of 10 g soil so strictly 0.83 x 10^6 spp/g, yet their fig. 4 shows total species number computed as up to 10^7 thus a million or so spp per g seems correct].

Raynard & Nunan (2014) had: "densities commonly found in bulk soil (10^8 cells g⁻¹ soil)" or "a single gram of soil can harbour up to 10^{10} bacterial cells and an estimated species diversity of between 4 x 10^3 [1] to 5 x 10^4 species [2]" (= 10^{14} – 10^{16} cells/t and 4 x 10^9 –5 x 10^{10} spp/t).

Bickel & Or (2020) found: "bacterial phylotypes ranges between 10^2 and 10^6 per gram of soil^{1.2.4}, with high values similar to the diversity in all of earths environments³" (= 10^8 – 10^{12} spp/t).

Madison James *et al.* (2022) summarize: "Soil microorganisms are the largest biodiversity pool on earth, with more than 10^{30} microbial cells [total surely!?], $10^4 - 10^6$ species, and nearly 1,000 Gbp of microbial genome per gram of soil (Vogel et al., 2009; Mendes and Tsai, 2018)".

Soils naturally include root-zone Rhizosphere: "the most diverse microbiomes on Earth, containing up to 10¹¹ microbial cells and ~30,000 bacterial species per gram of root [1]. The rhizosphere microbiome exists through an interwoven tapestry of bacteria, viruses, archaea,

protists, fungi, nematodes, and small arthropods interacting directly with plant roots and each other" (White et al., 2021). McNear (2013) has " 10^{10} - 10^{12} cells per gram rhizosphere soil".

Thus soil has up to 10^8 – 10^{12} cells/g or 10^{14} – 10^{18} cells/t, there being 10^6 grams in a tonne.

Soil biodiversity ranges $10^2 - 10^6$ spp/g or $10^8 - 10^{12}$ spp/t, there being 10^6 grams in a tonne.

How many tonnes of topsoil on Earth?

Whitman *et al.* (<u>1998</u>: tab. 2 below) cite 2.6 x 10^{29} prokaryotic cells in 1.2 x 10^{14} m² soil (or 10^{15} cells/m²). They footnote 1 m of topsoil ranges 10^7 to 10^9 cells per gram (median 10^8 /g) with 1.3 t per cubic metre to give a global total of (1.2 x 1.3 =) ~**1.6 x 10^{14} t of topsoil to 1 m depth**.

prokaryotes in soil

Ecosystem type*	Area, × 10^{12} m ²	No. of cells, [†] $\times 10^{27}$
Tropical rain forest	17.0	1.0
Tropical seasonal forest	7.5	0.5
Temperate evergreen forest	5.0	0.3
Temperate deciduous forest	7.0	0.4
Boreal forest	12.0	0.6
Woodland and shrubland	8.0	28.1
Savanna	15.0	52.7
Temperate grassland	9.0	31.6
Desert scrub	18.0	63.2
Cultivated land	14.0	49.1
Tundra and alpine	8.0	20.8
Swamps and marsh	2.0	7.3
Total	123.0	255.6

*From ref. 73.

[†]For forest soils, the number of prokaryotes in the top 1 m was 4×10^7 cells per gram of soil, and in 1–8 m, it was 10^6 cells per gram of soil (16). For other soils, the number of prokaryotes in the top 1 m was 2×10^9 cells per gram of soil, and in 1–8 m, it was 10^8 cells per gram of soil (18). The boreal forest and tundra and alpine soils were only 1 m deep. A cubic meter of soil was taken as 1.3×10^6 g.

Figure 3. Whitman *et al.* (<u>1998</u>: tab. 2) include desert, exclude glaciers and ignore terrain (<u>Ref.</u>). Conversely, Blakemore (<u>2018</u>b: fig. 4; tab. 5) has "*habitable land*" of 104×10^6 km² presumably with rich humic topsoil, say ~1 m deep (and ~1 t per m³), there being 10^6 m² in a km², thus 1.04 $\times 10^{14}$ t or 104,000 Gt. Doubled for terrain is about 208,000 Gt or ~2.1 $\times 10^{14}$ t global topsoil.

RESULTS

If $10^{14}-10^{18}$ cells/t soil x 2.1 x 10^{14} t the range is 2.1 x $10^{28}-10^{32}$ cells (median ~2.1 x 10^{30}).

A range of $10^8 - 10^{12}$ spp/t x 2.1 x 10^{14} t gives 2.1 x $10^{22} - 10^{26}$ spp total (median ~2.1 x 10^{24}).

Biomass carbon Whitman *et al.* (<u>1998</u>) took as half mean soil prokaryotic dry wt./cell (C : N = 1 : 0.24), so if 2×10^{-13} g/cell of all 2.1 x 10^{30} cells = $\sim 4 \times 10^{17}$ g or 400 Gt (200 Gt carbon : 48 Gt N).

DISCUSSION

Properly allowing for terrain, Earth's surface as exposed to Sun, air and rain of Land vs. Ocean from Blakemore (2018b) is 64 : 36%; thus Soil has by far the largest area supporting most biota.

Whitman *et al.* (<u>1998</u>) initially estimated microbial prokaryotes cell numbers in Soil vs. Ocean as 2.6 x 10^{29} vs. 1.2 x 10^{29} , respectively (68% vs. 32%); biomass soil carbon was 26 vs. 2.2 Gt C (92% vs. 8%). Soil was to 1 or 8 m depth, Ocean included open water and 10 cm of sediment. My present value of ~2.1 x 10^{30} soil cells to just 1 m increases their Soil tally by about ten times.

Remote subsurface biota are not a major concern. Whitman *et al.*'s (<u>1998</u>: tab. 5) 3.6 x 10^{30} & 2.5 x 10^{30} cells in Oceanic & Terrestrial subsurfaces were revised by Kallmayer *et al.* (<u>2012</u>) & Magnabosco *et al.* (<u>2018</u>) to just 3–5 x 10^{29} & 2–6 x 10^{29} , with subsurface biomass of 4 & 23–31 Gt C, respectively. A total range of 0.5–1.1 x 10^{30} cells (cf. <u>Ref.</u>, <u>Ref.</u>). New Soil cell total herein of ~2.1 x 10^{30} is two to four times as large and Soil microbe biomass of ~200 Gt C over six times.

For global biodiversity, a reasonable status summary by Fishman & Lennon (2022) had: "*the present number of bacterial and archaeal taxa* $S_{present}$ *is between* 10⁶ *and* 10²³." My present ~2.1 x 10²⁴ soil species increases their upper value by a factor of about twenty times but a lower estimate of ~2.1 x 10²⁰ soil species—as calculated below—is within their range. Adding weight to the argument, the methods to achieve these similar conclusions were calculated independently.

Whereas terrain doubles area at metre scale, at cm–mm sampling scale it is likely quadrupled. Blakemore (2018b: tabs. 5–6) had 1x, 2x and 4x soil as 162,000 Gt (agreeing with Whitman *et al.*, <u>1998</u>: tab. 2), 324,000 and 648,000 Gt, respectively; or 1.6, 3.2 and 6.5 x 10^{14} t soil. My median scaling estimate of around ~2.1 x 10^{24} soil microbial taxa is therefore likely a low value that may be readily doubled (see - <u>https://vermecology.wordpress.com/2022/08/04/different-f3/</u>). Accounting for neglected terrain & topography at cm–mm scale, if soil microbial tallies double again would total ~4.2 x 10^{30} cells, ~4.2 x 10^{24} spp and biomass total of ~800 Gt (= ~400 Gt C).

Accepting ~2.1 x 10^{30} soil microbial cells with ~2.1 x 10^{24} taxa implies one unique taxon per million microbial cells. Albeit if as many as 99% of soil microbes were eventually proven ubiquitous "species", ~2.1 x 10^{22} different genetic phylotypes would remain. Soils are often deeper than 1 m, but if such high species richness occurred in only the top 10 cm of 10% of Earth's topsoils, it would yet total in the order of ~2.1 x 10^{20} species. Higher values seem reasonable as random samples separated by up to 9,000 km had only <1.5% bacterial taxa in common to all, the majority of 88% OTUs unique to just one soil (Fulthorp *et al*, 2008). A similar figure of 80% wholly endemic taxa per soil sample was found by Schloss & Handelsman (2006). Bickel & Or (2021) found only 0.4% of soil bacterial species common, 99.6% classified as rare. Moreover, Operational Taxonomic Unit (OTU) often applies to Genera comprising many species.

Such findings attest to and support a massive Soil microbial biodiversity, orders of magnitude above that a cold, dark, densely saline, nutrient- and oxygen-depleted and ever-mixing Ocean's.

For the Ocean, Ferrer *et al.* (2019) categorically state: "*It is estimated that the ocean.*. *hosts the largest population of microbes on Earth. More than 2 million eukaryotic and prokaryotic species are thought to thrive both in the ocean and on its surface.*" Hoshino *et al.* (2020) claim: "Global marine sedimentary taxonomic richness predicted by species–area relationship models is 7.85 $\times 10^3$ to 6.10×10^5 for Archaea and 3.28×10^4 to 2.46×10^6 for Bacteria as amplicon sequence variants, which is comparable to the richness in seawater and that in topsoil [!]" also "the global diversity of marine prokaryotes (Archaea and Bacteria) in the near-surface ocean (0 to 1,000 m below sea level [mbsl]) was estimated (3.75×10^4 operational taxonomic units)".

Remarkably, a couple of grams of topsoil has an equivalent to all Ocean's total 10⁴–10⁶ taxa!

Furthermore, Whitman *et al.* (<u>1998</u>) had 5 x 10¹⁹ cfu (colony forming units) in the atmosphere, and, since only 0.001-1% of Bacteria are culturable, this likely equates to >>5 x 10²¹ cells. That is at least 500 times the estimate of Curtis *et al.* (<u>2002</u>) who said: "*The atmosphere is thought to have an NT* [total "individuals" (sic for microbes with binary fission!) of aerobiotic cells] *value of* 10^{19} , *which is sufficient to accommodate* 4×10^6 *taxa*" or 4 million species (most taxa shared with soils?). This is also twice the Ocean's best tally whilst also exceeding it as the Earth's largest biome. Aquatic taxa totals are proportionately infinitesimal. Prior to latest microbial totals mainly in soil, land was already shown to support 99.7% of biomass and 98.0% of biodiversity (Blakemore, <u>2020</u>: fig. 2; - <u>https://vermecology.wordpress.com/2020/05/27/realms-of-the-soil/</u>).



Figure 4. Blakemore (2020: fig. 2) Global soil biomass & biodiversity (~90% now upped >99.9%).

How much soil biota is currently known and why is it so important?

Soil provides 99.7% human food (Pimental & Burgess, 2013), all our fibres & building materials, many of our medicines such as Penicillin, Streptomycin or Ivermectin (from one site in Japan!), filters & stores most of our freshwater supplies, and supports >99.9% of biodiversity albeit we know <0.0001% of soil fauna, fungi or flora (an estimate below is 310,000 soil species currently described, most microbes or invertebrates such as earthworms that enhance microbes, *etc.*). Moreover, an argument is to include plant seeds/roots as valid soil flora (~4–500,000 spp - Ref.).

Soil Invertebrate Group:	Individuals m ²	Biomass g.	Known	No.
Batteria and Archant	(approx.)o	20-50040	B 2000	5.00 Willin
Englis	/southands	20.5004-	30.000	0.5660
Destarant	10007 10007	6,300	1.5050	8950
Potifiers / Ddalloid and antifersin	1032	10.000	300	0.000
Nematoda"	1081	1,300	35,000	-1.546.77
Laboradia (Onechanhera)	14.2	1-70-	-1 100m	000000
Tardigrada)0		125	~1,2000	
Lobopoula (Onychophara,)=	ha.	24	<2000	50%87
Lobopodia (Tardigrada)=	9	*	~1,9900	· 7= ·
Arachnida, Opdionest:	0	0	6,3000	?¤
Arachnida, Pseudoscorpionida@	0		3,3000	700
Actari (mites)0	1042	0.2.40	45,2000	4960
Hexapoda (totals)=	10 ⁴ m	0.2-44	-9,000=	17%sn
Hexayoda/Collembolai=	Lip to 100.000=	-	6,500=	1
Henapoda (Diphraia	0.	京	800×	0
Hexapoda (Protrana coneheads)=	- P	-	733=	SH.
Soil Insects and their larvae:	50-5000	4.50	55,000+/10	20%/72
Myriapoda (centi-, millipedes)0	100-1100:	1.5-22.50	18,0000	20%40
Miriapada (Sympinia)	5	I	7600	
Pauropoda (Mariapodorelative)=	早ららいないの	-	300=	D.
lsopoda(slaters, woodlice, etc.)c	Up to 18000	<40	5,0000	192
Isoptera (Termites):	Colonies:	70	2,6000	60%20
Blattodsa (Cockroaches)5	70.	10	4,5000	70
Ants (Hymenoptera - Formicidae)=	Colomest .	- 20	13,0000	.50%=
Mollunes (Seil Gastropods)	20	10	24,0000	409630
Terrestrial Turbellaria (Planarians)	⁹ Π	70	\$30+0	. 90
Terrestrial Polychaeta:	fπ.	30	71	7c
Oligochaeta (Mega-, Microdriles)=	50-2,000=	28-500=	10,000=	20%7=
Microdyller/Exclatracidaeia	1.000-300.000=	1.53=	~.709=	7=
Microdriler (excluding enclatmend)	71	30	7-2,899270	9±
Megadriles (mue cartloroms)=	59-2.020=	20-303*=	7,009=	<20%
Total species (approx.)0	0	- 22	310.000:	20

earthurem values are 'from Lee (1985: tab. ??) in NZ pastarse (mear 2.020 m² with 305 gm² from McColl & Lautour, 1978): Enclyraed maxima ore from Springett (<u>1967</u> fig. 24):Gragg (<u>1963</u>, tab. ?) from Moor Hruue, UK (mis)quoted by Spain & Lavelle (<u>2001</u>).¶



Blakemore (<u>2012</u>: tab. 1, <u>2016</u>: tab. 3) compiled list of >310,000 spp is greater than a \$1 billion, 10 yr Census of Marine Life of 230,000 spp (<u>Ref</u>.). After this CoML survey completed, Mora *et al.* (<u>2011</u>: tab. 2) totalled just 194,409 catalogued Ocean spp; both predicted only ~2 million total taxa. The current study substantially increases Soil taxa total, thus disproportionate funding for Marine research is illogical when Soil is so rich and, via runoff, provisions or pollutes the Ocean.

As biodiversity estimates grow so too they decline as soil erosion and species extinctions take their toll (Blakemore, <u>2018a</u>; <u>Ref.</u>, <u>Ref.</u>). Despite vital importance and a massive biodiversity inventory, not a single **SOIL ECOLOGY INSTITUTE** yet exists anywhere on Earth (except perhaps on my humble office desk – see: <u>https://vermecology.wordpress.com/2015/12/10/isei-international-soil-ecology-institute-and-soil-ecology-exchange-yokohama-seexy-open-day-5th-december-2015-for-unfao-international-soil-day/). For rational continuation of our reasonable survival, this major Soil research deficiency requires redress with urgent redirection of resources.</u>

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ADDENDUM

Global Microbiome Counts in Intestines of Earthworms and Termites (with a Footnote on Abundance of Ants and Soil Viruses)

Robert J. Blakemore PhD

VermEcology, 101 Suidomichi, Nogeyama, Kanagawa-ken 231-0064, Japan.

Email: rob.blakemore@gmail.com. Date: 22nd Sept., 2022.

ABSTRACT

Earthworms and termites are two of the wholly soil-dwelling invertebrate faunal groups. Both are known to support vibrant and enhanced microbial gut communities of symbionts and parasites. Estimates of gut microbial abundance for all 65×10^{15} earthworms with 4.5×10^{15} grams dry biomass – including their soil ingesta – is an intestinal count of $10^{6}-10^{15}$ cells/g thus $10^{21}-10^{30}$ cells total. Termite gut microbiome range of around $10^{22}-10^{23}$ cells is minor. Although large, the contribution earthworms make to total soil counts is unclear: Total soil microbial biomass for median ~2.1 x 10^{30} cells (in range $10^{28}-10^{32}$) is ~4 x 10^{17} g or 400 Gt (with 200 Gt C & 48 Gt N).

A footnote revises ants up to 40 x 10^{15} with 0.024 Gt C, yet less than earthworm abundance at 65 x 10^{15} (or up to 10^{19}) with 2.25 Gt C. The soil virus estimate is ~ 10^{30} virions with ~0.03 Gt C.

INTRODUCTION

Earthworms (Annelida: Oligochaeta: Megadrilacea) are the major component of healthy soils. Earthworms and microorganisms are co-evolved and interdependent thus their interactions regulate Ecology of terrestrial soils. Megadrile earthworms comprise 20 families, approximately 600 genera (several un-necessary sub-genera) and ~7,000 described species or sub-species with an expected total of over 35,000 species (Blakemore, 2012, 2016b). They represent up to 90% of invertebrate biomass present in soil and microbes are known to increase during digestion up to x 1,000 and after gut passage in their castings (Lee, 1985: 27, 206). Their total abundance [from Darwin (1881) 1.3×10^{15} ; Lee (1985: tab. 7) 27 x 10^{15} ; Blakemore (2000, 2016b, 2018a) 65×10^{15}] range is therefore $1.3-65 \times 10^{15}$ worms or up to 65 quadrillion with mean 15 Gt wet biomass thus 30% and about 4.5 Gt dry mass or 4.5×10^{15} g, with ~2.25 Gt C. (See – https://vermecology.wordpress.com/2017/02/12/nature-article-to-commemorate-charles-darwins-birthday-on-12th-feb/ and https://vermecology.wordpress.com/2021/02/12/cd-bd/).

Bar-On et al. (2018: tab. S1) Annelids total of 10¹⁸ (Megadriles + Microdriles) is feasible. *

Soil terrain consideration (Blakemore, <u>2018</u>b) doubles evenly dispersed earthworm counts, but taxa such as ants or termites in discrete colonies or nests per unit area are not so enhanced.

Earthworms digestive systems have muscular gizzards, calciferous glands, typhlosoles and/or caeca thus, during their passage, ingested micro-organisms may increase 10 to 1,000 times

(<u>Ref.</u> below quoting Parle, 1963). Whereas Parle (<u>1963</u>a) says the microbes only increase ten times in mid- to hind-gut he showed x 10^3 in antilogs of his table 1. Later, Parle (<u>1963</u>b: tab. 1) reported soil to casts for Actinomycetes & Bacteria went from 4×10^7 to 6×10^9 & from 8×10^8 to 8×10^{10} , respectively, or an increase about 100 times. Stockli (1928) had shown Actinomycetes & Bacteria increased 600 times & 1,000 times in intestines with hindgut counts raised to 15×10^9 & 4.4×10^{11} cells, respectively (his table highlighted in blue below).



ing passage through the gut. Stockli (1928) reported great increases in total numbers of bacteria and actinomycetes (Table 1).

Similarly, Ponomareva (1953) found that numbers of actinomycetes, pigmented bacteria and other bacteria of the *Bacillus cereus* group increased as food moves backwards along the earthworm intestine. One of the most detailed studies of earthworm nutrition to date was that of Parle (1963), who also reported that actinomycetes and bacteria multiply as they pass through the gut of *L. terrestris*, *A. caliginosa* and *A. longa*, with about 1000 times more of these organisms in the hind gut than the foregut. Supporting evidence that multiplication in numbers of microorganisms occurs during passage of materials through the gut comes from the many reports of greatly increased numbers of microorganisms in the casts of earthworms, compared with those in the soil

TABLE 1

Numbers ($\times 10^6$) of microorganisms in different parts of the intestine of L. terrestris (Stöckli, 1928)

Taxa	Foregut	Midgut	Hindgut
Actinomycetes	26	358	15 000
Bacteria	475	32 900	440 700

Figure 1. With report of 600–1,000 times increase in microbes in earthworms gut (data in blue).

Of note is many such studies use culturable cfu plate counts that produce a small proportion of total soil microbes, perhaps just 1% (<u>Ref.</u>) or possibly as low as 0.001% to 0.1% (<u>Ref.</u>) of all bacterial species! Thus final results or current estimates of 10^8-10^{12} microbe cells/g in bulk topsoil ingesta as taken as initial base range, may both be far too low.

Soil digestion rate per earthworm per day vary widely by species, size, substrate, season and habitat: from 0.1 g/g for large European forest worms up to 36 g/g for juveniles in tropical savannah (Lee, 1985: 17-22). A reasonable ordinal average is likely ~1 x body weight per worm per day with passage time also about one day (as per <u>Ref</u>.). Lee (1985) reported earthworms can process up to 25% of organic Ah soil horizon in 1 year, or all in four years (cf. Darwin, 1881).

Parle (<u>1963</u>a: tab. 1 below) for three reasonably widely distributed arable earthworm species had about the same number of Yeast or Fungi cells in surrounding soil compared to earthworms' gut, but Actinomycetes & Bacteria were substantially increased on average from 2×10^7 to 5×10^8 /g and from 1×10^8 to 1×10^{11} /g, respectively. Thus antilog counts, rather than logs, were increased by one to three orders of magnitude or ten to 1,000 times. Later studies have similar range (<u>Ref.</u>, <u>Ref.</u>), supporting claims that "*ingested soil may contain 10*⁹ or more microbial cells per gram dry weight" (<u>Ref.</u>), the earthworm gut microbiome can have ~ 10^{10} cells per gram (<u>Ref.</u>).

or passage through their digestive system increases microbes up to 1,000 times (<u>Ref.</u>; <u>Ref.</u>; <u>Ref.</u>; <u>Ref.</u>; Lee, 1985: 27; cf. <u>Ref</u>. "*a Review*" that surprisingly ignores any references prior to 1986!).

	Yeasts	Fungi × 10 ⁶	Actino- mycetes × 10 ⁶	Bacteria × 10 ⁶
	Lu	mbricus terrest	ria	
Gut content	4·89 ±0·122	-0.25 ± 0.122	1-97 ±0-154	$4-91 \pm 0.166$
Soil	4-70 ± 0-178	-0.50 ± 0.164	0-95 ± 0-226	2.07 ± 0.244
	Allol	obophora caligi	nosa	
Gut content	5.06 ±0-070	-0.08 ± 0.081	3.12 ± 0.198	4.42 ± 0.342
Soil	4·79 ± 0·108	-0.89 ± 0.119	1-54 ± 0-290	2.06 ± 0.500
		A. longa		
Gut content	5.46 ±0.074	0.52 ± 0.068	3.25 ± 0.102	5·80 ±0·070
Soil	5-00 ± 0-109	$0-15 \pm 0-099$	1.00 ±0.149	2.08 ± 0.103

Table 1. Counts of yeasts, fungi, actinomycetes and bacteria in worm intestinal contents compared with soil. Mean log. number of organisms per g. dry wt. of gut contents or soil

Figure 2. After Parle (<u>1963</u>a: tab. 1) with ~10–1,000 times microbe increase.

Soil values of Fungi, Actinomycetes & Bacteria all at x 10^{5-6} per gram compare to Suzuki (2007: tab. 4.1) of Algae x 10^{4-5} , Fungi x 10^{5-6} , Actinomycetes x 10^{7-8} , Bacteria x 10^{8-9} per gram soil:

		Made from the Soil 121
TABLE 4.1:		
Relative Number of	Soil Flora and Fau	na in Surface Soil
ORGANISMS	NUMBER/METRE ¹	NUMBER/GRAM
Microflora		
Bacteria	10 ¹³ -10 ¹⁴	108-104
Actinomycetes	10 ¹¹ -10 ¹¹	10 ⁷ -10 ⁸
Fungi	10 ¹⁰ 10 ¹⁷	10 ⁵ -t0 ⁶
Algae	10 ⁸ -10 ¹⁰	104-105
Microfauna		
Protozoa	10 ⁹ -10 ¹⁰	104-103
Nematoda	108-107	10~10 ²
Other fauna	10%-103	
Earthworms	30300	

Figure 3. After Suzuki (2007: tab. 4.1 from Ref.); note Earthworms relationship to microbes.

Earthworm-rich Organic vs. Conventional farm soils in Philippines had higher Fungi & Bacteria populations (+55% & +44%) with counts up to 5×10^4 & 1×10^7 cfu/g, while vermicompost Fungi & Bacteria were higher yet at 1.5×10^5 & 1.2×10^7 & cfu/g (Blakemore, <u>2016</u>b: tabs. 5 & 7).

For termites, Whitman *et al.* (<u>1998</u>: tab. 4) have prokaryote cells per termite hindgut of 2.7 x 10^6 and, for a world population of 2.4 x 10^{17} of these insects, they conclude as 6.5 x 10^{23} cells total (cf. <u>Ref.</u>; <u>Ref</u>.). An estimate of global termite biomass is 4.4 x 10^8 t wet (<u>Ref</u>.) or ~2.2 x 10^8 t dry.

METHODS

Earthworm and termite total biomass estimates are multiplied by their known microbial residents.

RESULTS & DISCUSSION

Earthworm global dry biomass of 4.5×10^{15} g with $10^6 - 10^{11}$ cells/g of Acinomycetes, Bacteria & Archaea (unknown at time of Stockli's or Parle's studies) totals $4.5 \times 10^{21} - 10^{26}$ cells. Or, Kieft & Simmonds (2015: fig. 1) of 10^8 microbes per worm for $1.3-65 \times 10^{15}$ worms = $0.1-6.5 \times 10^{24}$ cells. If actual increase during passage is $10-10^3$ with bulk starting soil range of $10^8 - 10^{12}$ cells/g (from Blakemore, 2022) this presumably raises to $10^9 - 10^{15}$ cells/g worm by 4.5×10^{15} g worms = $4.5 \times 10^{24} - 10^{30}$ microbial cells in total. The upper value is twice a mean global cell count so perhaps is not tenable. Alternatively, it may signal the median total soil microbes of 2.1×10^{30} soil cells is too modest and support an argument that earthworms contribute greatly to overall soil microbiota. Unique species are acknowledged but poorly known so no estimate is possible.

Interestingly, if an earthworm biomass of 15 Gt wet (4.5 Gt dry) on average processes its body weight of soil per day this would total 5,500 Gt wet (1,540 Gt dry) topsoil per year. Such passage also stimulates a 10–1,000 microbial increase in their cast plus the aeration and water infiltration in their burrows benefits microbes too. Re-estimation of soil microbes at 400 Gt with 200 Gt C must then correspond with earthworm activity to a large extent and relate to humus.

For termites, 2.2 x 10^{14} g for 2.4 x 10^{17} population = 10^{-3} g/termite (dry). Hindgut count of 2.7 x 10^{6} cells/termite is 2.7 x 10^{9} cells/g; by 2.2 x 10^{14} g = 6 x 10^{23} cells. Or, Kieft & Simmonds (2015: fig. 1) of 10^{6} cells/termite for 10^{-2} g/termite (wet); by 4.4 x 10^{14} g = 4.4 x 10^{22} cells. Slightly smaller than Whitman *et al.*'s 6.5 x 10^{23} cells, also a minor contribution to soil biomass.

CONCLUSION

Although terrain consideration increases all soil biometrics, new estimates of resident microbes of earthworm intestine or mostly tropical termite gut make large and lesser contributions to total soil microbial median abundances of 2.1 x 10^{30} cells at 2 x 10^{-13} g/cell = 400 Gt (200 Gt C, 48 Gt N) and 10^{24} taxa (Blakemore, 2022). Currently the contribution of earthworms to unique, host-specific parasitic or symbiotic microbial species are unknown in scope of total soil biodiversity.

Despite estimates of probable numbers of unknown soil taxa increase, there is a corresponding rapid decline in species with the intensification of agriculture and excessive meat eating (Blakemore, 2018a). Solutions are to farm organically, study Permaculture, and to eat less meat.

As Darwin (<u>1881</u>) said: "*Finally, no one who considers the facts given in this chapter—on the burying of small objects and on the sinking of great stones left on the surface—on the vast number of worms which live within a moderate extent of ground—on the weight of the castings ejected from the mouth of the same burrow—on the weight of all the castings ejected within a known time on a measured space—will hereafter, as I believe, doubt that worms play an important part in nature." Certainly their relationships with soil microbes are intimate & intricate.*

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FOOTNOTE

A recent 2022 study by several ant academics found: "20 × 10¹⁵ (20 quadrillion) ants on Earth, with a total biomass of 12 megatons of dry carbon... equivalent to ~20% of human biomass" Their average in tropics was 54.9 ants/m² and in temperate regions 2.56 ants/m². This totals about 57.5 ants/m². Actual data were: "global epigaeic ant abundance = 3.02×10^{15} (±0.74×10¹⁵) global arboreal ant abundance = 1.34×10^{15} (±0.36×10¹⁵)" they multiplied these by ~5x to allow for non-foraging nest ants. As I explained above, when true land surface area is doubled for terrain (Blakemore, 2018b: tab. 5) it doubles dispersed organisms such as earthworms or foraging ants, but not nest ants that are concentrated in oft measured nests/unit-area.

Seemingly they used the usual planimetrical and plainly wrong "flat-Earth" model – www.pnas.org/doi/suppl/10.1073/pnas.2201550119/suppl_file/pnas.2201550119.sapp.pdf.

Doubled for terrain & topography (Blakemore, 2018b: tab. 5) their total is about 40 x 10^{15} ants which is yet less than earthworms quadrillions with 65 x 10^{15} global abundance. And earthworm biomass of 2.25 Gt C dry weight carbon is about 100 times larger than their paltry 0.012 Gt C doubled to 0.024 Gt C. Again this begs query why earthworms and soil ecology are so ignored. Earthworms mean number 273 worms/m² serves to confirm my calculations above as in range.

Regarding viruses, aside from questions on whether they are "living" or not, their abundance in soils may be miscalculated by orders of magnitude. Viruses in Bar-On *et al.* (2018: tab. S1 copied below) have biomass of 0.2 Gt C from global abundance of 10^{31} . Yet a summary paper (Ref.) found: "While many soils contain large numbers of viral particles (10^7 to 10^9 virus particles per gram of soil [37, 40–42]), knowledge of soil viral ecology has come mainly from the fraction that desorbs easily from soils (<10% in reference 43) and the much smaller subset that has been isolated (44)." This would give a range of > 10^8-10^{10} /g. Like bacteria, their soil numbers are massive. A reasonable published estimate of 10^9 viruses/g soil (Ref.), that if also <10% gives 10^{10} /g soil. Thus Earth's ~ 2.1×10^{20} g soil may have ~ 2.1×10^{30} virions and, if 10^{31} is 0.2 Gt C, this equates to ~0.04 Gt C. Compare to Bar-On *et al.*'s best estimate (Supplement: 55-56): "assuming a mean soil depth of 10 meters (276), we get to an estimate of ~ $10^{29} -10^{30}$ virions in soil globally" that they average as ~ 6×10^{29} soil viruses (mainly phages). Another simple calculation is to double their estimate for terrain 2x to ~ 1.2×10^{30} with about 0.02 Gt C. This is just over a quadrillion-quadrillion but, as already mentioned, it may be out by orders of scale and whether it truly adds living matter to an ever-evolving soil biota tally is up for debate.

Table S1. Summary of estimated total biomass and abundance of various abundant taxonomic groups.

Values are based on a literature survey as detailed in the *SI Appendix*. Reported values have been rounded to the closest order of magnitude in log scale thus reflecting the associated level of uncertainty.

Taxon		Mass [Gt C = 1015 g C]		Abundance	
Plants	Trees			450	1013
Bacteria	Terrestrial deep subsurface		60		1034
	Marine deep subsurface		7	i - 1	1024
	Soil		7		1028
	Marine		1.3	1 8	1029
	Total			70	10 ³⁰
Fungi				12	1023
Archaea	Terrestrial of	leep subsurface	4		1021
	Marine deep	p subsurface	3		1029
	Soil		0.5	l í	1028
	Marine		0.3	1 3	1028
	Total			7	1024
Protists				4	1023
	Chordates	Fish	0.7		10/01
Animals		Livestock	0.1		10.00
		Humans	0.06		10%
		Wild mammals	0.007		
		Wild birds	0.002		1011
	Arthropods	Terrestrial	0.2		1018
		Marine	1		1039
	Annelids		0.2		1011
	Molluses		0.2		10 ¹⁰
	Cnidarians		0.1		1026
	Nematodes		0.02		1021
	Total			2	1021
Viruses				0.2	1011

*Figure 5. From Bar-On *et al.* (2018: tab. S1 - http://rpdata.caltech.edu/publications/Bar-On_2018_SI.pdf); updated in Blakemore (2018b: fig. 12, tab. 10). For Annelids, i.e., megadrile earthworms (+ microdriles but they exclude marine polychaetes!) on pages 31-32 they estimate "300 individual earthworms per m^2 [soil]", 5 mg C per worm & 1.5 g C m⁻² (= 3 g dry, 10 g wet m⁻²) supporting my abundance numbers of 273 m⁻² & fresh weight 63 g m⁻². Their 0.2 Gt total earthworm biomass is now 2.25 Gt C (Blakemore, 2022). Abundance they have as 10¹⁸ yet on page 61 say: "For annelids... we use our biomass estimates and divide them by the mean individual body mass"; i.e., 0.2 Gt C / 5 mg C per worm = 4 x 10¹⁶ or 40 quadrillion, not 1,000 quadrillion as they calculated! However, enchytraeid abundances range 10³-10⁵ m⁻² (Ref.) so if megadriles at 10² m⁻² total 10¹⁵-10¹⁶, this computes as 10¹⁶-10¹⁹ for total worms. Please note too how their Soil has about 24 Gt C (92%) including Fungi and Protists while Ocean is only about 2 Gt C (8%), before terrain adjustment that doubles Soil to at least 50 Gt C and also adds about half the Plant biomass (e.g. roots, logs, litter, etc.) to probably total >250 Gt C (>99%). In their Supplement they also state "≈98% of the total microbial biomass is found in the top 1 meter of soil" further supporting my microbial total estimates based upon 1 m depth of global soils.