

Supporting Information: “Exposure to dairy manure leads to greater antibiotic resistance and increased mass-specific respiration in soil microbial communities”

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Table S1. Site locations and soil chemical, physical, and biological properties for the 11-paired soils. Site indicates the U.S. state of collection. SIR : substrate induced respiration; DOC : dissolved organic C; POM : particulate organic matter.

Site	Input	Latitude	Longitude	pH	SIR CO ₂ -C ($\mu\text{g g dry wt soil}^{-1} \text{ h}^{-1}$)	Microbial C (mg g dry wt soil ⁻¹)	Microbial N (mg C g dry wt soil ⁻¹)	DOC (mg C g dry wt soil ⁻¹)	C-min CO ₂ -C (mg g dry wt soil ⁻¹)	Mineral C (mg g dry wt soil ⁻¹)	Mineral N (mg g dry wt soil ⁻¹)	POM C (mg g dry wt soil ⁻¹)	POM N (mg g dry wt soil ⁻¹)	Total C (mg g dry wt soil ⁻¹)	Total N (mg g dry wt soil ⁻¹)	Sand (%)	Clay (%)	Silt (%)	Fungal-to-bacterial dominance	tefO (log copies g soil ⁻¹)	tefW (log copies g soil ⁻¹)	ampC (log copies g soil ⁻¹)	ermB (log copies g soil ⁻¹)
NY	+Manure	42.4407	-76.2454	7.2	9.1	0.63	0.05	0.4	2.48	82.0	9.4	53.2	5.08	135.2	14.4	53.53	7.75	38.73	0.0028	5.06	5.12	5.18	1.10
FL	+Manure	29.7765	-82.4166	7.4	2.56	0.06	0.006	0.33	0.44	5.5	0.5	3.6	0.38	9.1	0.8	97.50	2.50	0.00	0.1054	6.75	6.57	5.56	2.01
GA2	+Manure	33.9130	-83.2452	5.9	13.03	0.6	0.046	0.3	1.78	58.9	6.6	30.8	2.57	89.7	9.1	74.46	10.22	15.33	0.0016	4.20	5.56	5.17	0.93
GA1	+Manure	33.4430	-83.5060	8.6	8.71	0.72	0.078	1.06	7.55	48.8	5.6	39.3	2.06	88.1	7.7	72.03	12.71	15.25	0.2132	8.06	7.29	6.65	1.40
KS	+Manure	39.0483	-96.9207	8.9	11.75	3.25	0.295	1.23	7.53	104.0	10.6	97.0	5.92	201.0	16.5	45.45	22.08	32.47	0.0683	6.38	5.90	5.30	0.62
MS	+Manure	33.4019	-88.7422	8.5	8.85	0.28	0.096	1.38	4.61	55.9	6.8	11.5	0.86	67.4	7.7	48.88	11.50	39.62	0.0063	7.77	6.67	6.08	2.40
NH	+Manure	43.1473	-70.9521	6.9	2.94	0.04	0.001	0.14	0.36	13.3	1.3	2.0	0.25	15.3	1.6	59.73	8.81	31.46	0.3853	6.73	5.85	5.82	0.42
VT	+Manure	44.5800	-73.1402	8.4	8.26	0.63	0.071	0.64	3.55	51.9	5.6	79.5	2.43	131.4	8.0	47.88	17.80	34.32	0.4196	7.01	6.81	6.88	0.54
VA	+Manure	37.2145	-80.4252	7.8	9.12	0.88	0.133	0.47	8.08	50.7	5.5	44.9	2.79	95.6	8.3	26.12	28.02	45.85	0.2808	7.86	6.63	6.57	1.46
WA	+Manure	46.6933	-117.2406	7.6	10.1	0.35	0.056	0.28	1.56	49.6	5.1	28.5	2.03	78.1	7.1	21.02	20.38	58.60	0.1481	6.56	6.19	5.64	2.75
WV	+Manure	39.6630	-79.9330	7.4	10.17	0.28	0.028	0.19	1.3	38.3	4.1	11.1	0.85	49.5	5.0	18.74	49.52	31.74	0.0575	6.40	6.14	5.81	1.49
NY	Reference	42.4407	-76.2454	7.1	10.46	0.5	0.074	0.17	1.68	64.2	7.3	24.9	2.11	89.1	9.4	43.58	12.82	43.60	0.0008	4.57	5.76	5.53	1.94
FL	Reference	29.7765	-82.4166	7.4	2.82	0.06	0.015	0.18	0.34	6.8	0.6	6.3	0.68	13.1	1.3	97.49	2.51	0.00	0.2620	5.40	6.18	4.72	0.57
GA2	Reference	33.9130	-83.2452	5.1	5.62	0.16	0.035	0.2	0.52	27.5	3.2	10.1	0.76	37.6	4.0	49.41	22.76	27.82	0.0010	5.12	6.04	5.45	1.36
GA1	Reference	33.4430	-83.5060	5.6	6.53	0.17	0.023	0.24	0.91	27.1	3.2	16.1	1.28	43.2	4.4	62.18	20.17	17.65	0.8353	5.74	6.16	5.63	1.02
KS	Reference	39.0483	-96.9207	7.9	6.39	0.65	0.06	0.41	1.66	81.1	7.8	43.4	2.97	124.6	10.7	41.22	12.78	46.00	0.0060	6.31	6.24	5.29	0.81
MS	Reference	33.4019	-88.7422	7.6	5.98	0.34	0.014	0.21	1.58	19.2	2.2	57.4	4.44	76.6	6.7	67.30	12.58	20.12	0.0160	5.40	6.17	5.54	1.51
NH	Reference	43.1473	-70.9521	6.9	1.82	0.03	0.012	0.08	0.16	11.6	1.1	1.1	0.10	12.8	1.2	54.84	17.56	27.60	0.3644	3.18	6.26	5.53	0.76
VT	Reference	44.5800	-73.1402	8.1	2.93	0.11	0.012	0.24	1.24	21.9	1.8	10.5	0.33	32.4	2.1	1.66	21.43	76.91	0.0127	5.73	6.21	5.83	0.59
VA	Reference	37.2145	-80.4252	7.9	5.36	0.31	0.052	0.39	1.63	46.8	4.0	47.4	1.22	94.2	5.2	33.87	25.43	40.69	0.0101	6.47	5.99	5.71	0.53
WA	Reference	46.6933	-117.2406	6.9	4.53	0.07	0.002	0.11	0.46	19.2	1.4	5.0	0.28	24.3	1.7	9.04	25.27	65.69	0.1307	6.12	6.29	5.46	1.96
WV	Reference	39.6630	-79.9330	7.4	5.2	0.13	0.013	0.1	0.33	24.0	2.5	3.1	0.21	27.1	2.7	16.45	37.98	45.57	0.1284	4.97	6.21	5.48	1.35

Fig. S1. Location of the 11 sites.

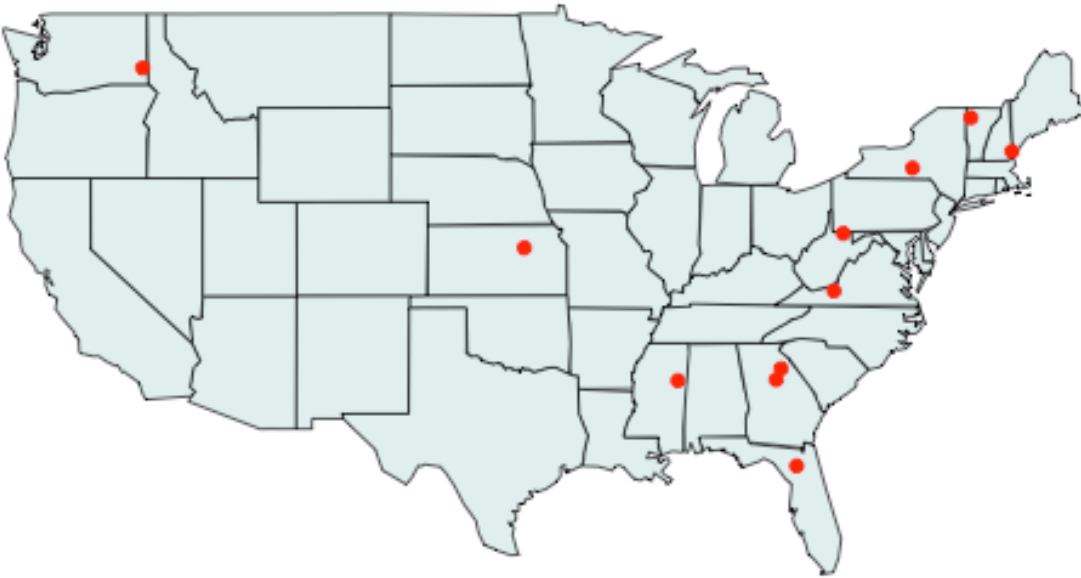


Fig. S2. The fungal genus *Preussia* showing (A) the significant change in relative abundance between manure-exposed (+Manure) and reference sites ($F_{1,10} = 8.0$; $P < 0.05$); and (B) the relationship between the relative abundance of *Preussia* and *ampC* abundance.

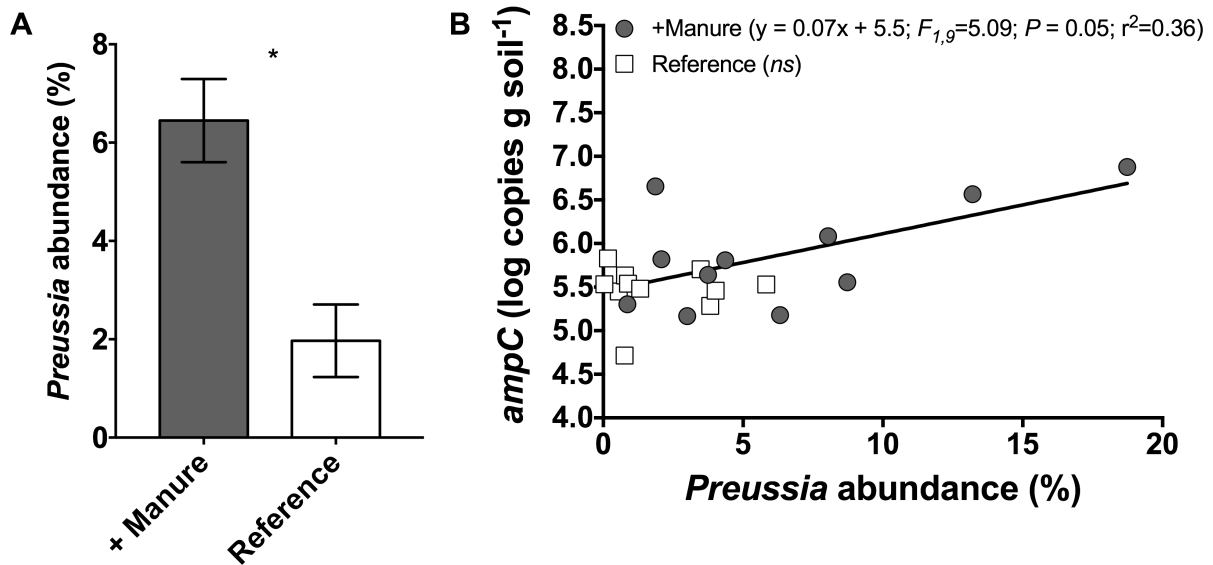


Fig. S3. The relationship between the change in the microbial community and change in total ARG abundance (the sum of the ARGs: *ampC*, *tetO*, *tetW*, and *ermB*). **A)** Relationship between Bray-Curtis dissimilarity between manure-exposed and reference plots located at the same geographic location and change in total ARG abundance expressed as relative percent change from the reference. **B)** Change in the relative abundance of the phylum Firmicutes and the change in total ARG abundance from reference to manure-exposed sites. Change is expressed as relative percent change from the reference.

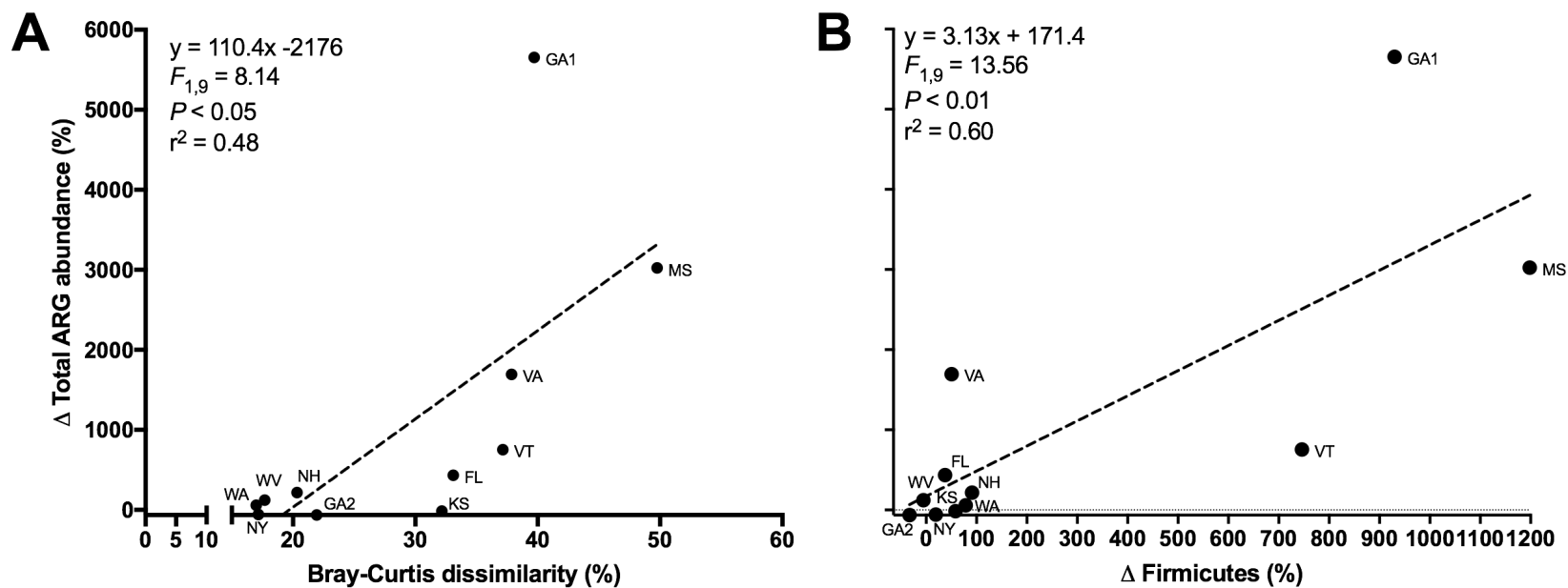


Fig. S4. Relative parameter importance for predicting qCO₂ as determined via model averaging of top models ($\Delta AICc < 4$). For further details see statistical analyses below.

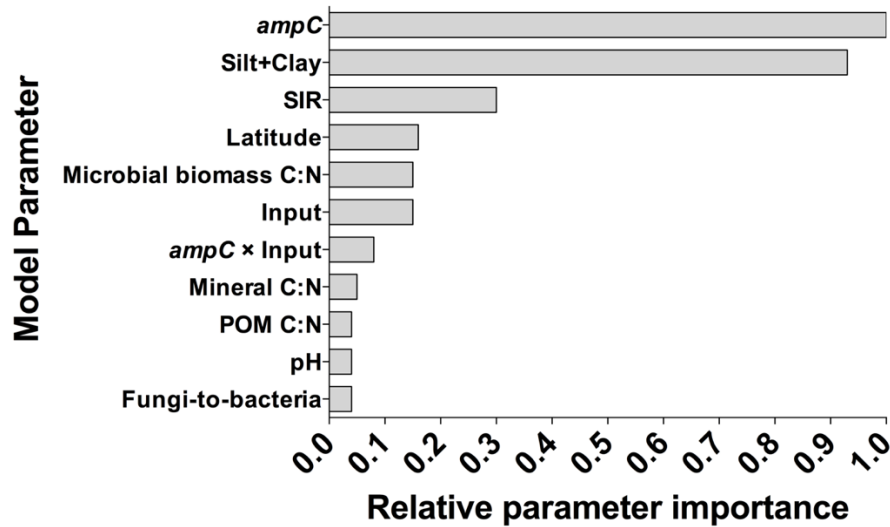


Table S2. Top models ($\Delta AICc < 2$) determined via multi-model inference.

Model	log(L)	AICc	$\Delta AICc$	ω_i	r^2	Rank
$qCO_2 = ampC + (Silt+Clay)$	14.6	-18.9	0.00	0.10	0.60	1
$qCO_2 = ampC + (Silt+Clay) + SIR$	16.0	-18.2	0.75	0.07	0.64	2
$qCO_2 = ampC \times Input + (Silt+Clay)$	17.3	-17.0	1.96	0.04	0.68	3

Table S3. Model-averaged coefficients using a subset of top models ($\Delta AICc < 4$).

Parameter	Estimate	Adjusted SE	z-value	P-value
Intercept	-0.86	0.44	1.94	0.05
<i>ampC</i>	0.28	0.07	4.10	<0.001
Silt+Clay	-0.004	0.001	2.93	<0.01
SIR	-0.01	0.01	1.50	ns
Input (Low)	1.02	1.15	0.89	ns
<i>ampC</i> × Input (Low)	-0.32	0.18	1.76	0.08
Microbial biomass C:N	-0.004	0.003	1.13	ns
Latitude	-0.01	0.01	1.19	ns
Mineral C:N	0.01	0.02	0.43	ns
pH	0.01	0.04	0.39	ns
Fungal-to-bacterial dominance	0.03	0.17	0.18	ns
POM C:N	-0.0001	0.004	0.03	ns

Fig. S5. Relationship between Silt + Clay (%) and $q\text{CO}_2$, an indicator of microbial stress. Grey circles indicate manure-exposed sites and open squares indicate reference sites. A significant relationship was observed for reference ($y = -0.003x + 0.59$; $F_{1,9} = 11.75$; $P < 0.01$; $r^2 = 0.57$) but not manure-exposed. Additionally, multi-model inference indicates that Silt + Clay (%) is an independent variable of high importance when considering microbial stress (**Fig. S4** and **Table S3**).

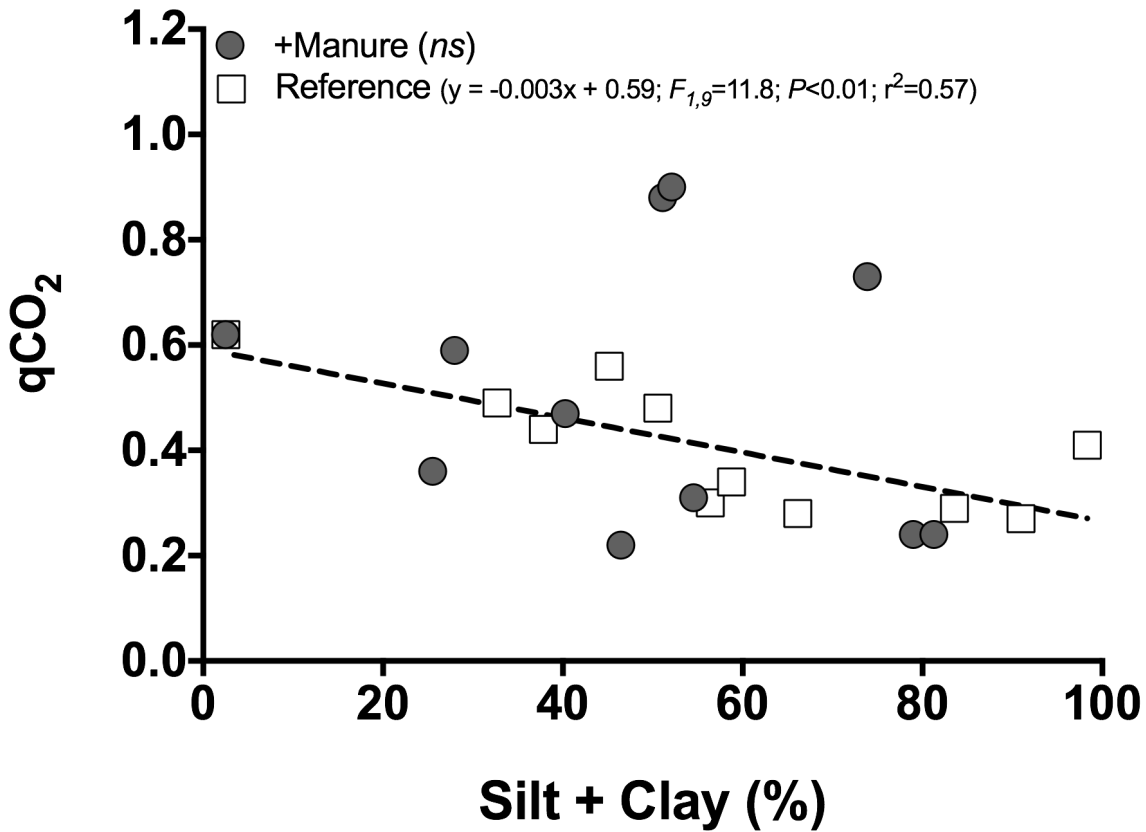


Fig. S6. The relationship between total ARG abundance (the sum of the ARGs: *ampC*, *tetO*, *tetW*, and *ermB*) and microbial mass specific respiration for reference and manure-exposed sites. Grey circles indicate manure-exposed sites and open squares indicate reference sites.

