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Research article

Nitrous oxide emissions from trees planted on a closed landfill site

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Supplementary methods

Site and tree characteristic measurements

For each tree flux measured the GPS location, tree species and diameter at breast height (DBH) was recorded. Air temperature and air pressure at each location were measured with a Comet C4141 Thermo-hygro barometer (air temperature accuracy ± 0.4 °C; air pressure accuracy ± 2 hPa at 23 °C). Tree stem surface temperature was recorded using an infrared thermometer (RS Pro RS1327k) (accuracy ± 0.1%). At each soil and tree location, soil temperature (Thermapen soil temperature probe; accuracy ± 0.4 °C) and soil moisture (Delta-T Devices HH2 moisture metre with ThetaProbe type ML2x; accuracy ± 1%) were measured at 10 and 6 cm depth, respectively. Soil cores were taken to determine bulk density and pH (Thermo Scientific Orion Versa Star Advanced Electrochemistry metre with Orion 8157 BNUMD ROSS Ultra pH ATC Triode; accuracy ± 0.002).

Gas chromatography

Samples taken from the stem and soil chambers were analysed using gas chromatography. An Agilent 7890A Gas Chromatograph (GC) fitted with a micro electron capture detector (μECD) and autosampler was used to introduce 2 ml samples into a 1 ml loop (Sgouridis, 2021). A splitless injection technique was used due to the low concentration of N_2O in the samples. A 1/8-in stainless steel packed column (packed with Porapak 80/100) and a Ar5%CH_4 carrier gas (flow rate 2 mL min^{-1}) were used during this analysis. The oven and μECD temperatures were 60 °C and 350°C, respectively. The N_2O

concentration in each sample was determined by integrating peak area (OpenLab CDS software, version 3.5) followed by linear regression comparison to known standards (0 ppm, 1 ppm and 6 ppm; $R^2 = 0.999$). Dilutions of a 6 ± 0.1 ppm calibration standard (CK Isotopes) were used for this analysis. The minimum detectable concentration difference for N_2O using this method was 4.2 ppb.

Vial leakage tests and corrections

Corrections were calculated for each set of monthly samples based on the difference between average N_2O concentrations of the stored standards and the standards that were not stored. A percentage correction was calculated for each sampling month and the N_2O concentration values for that month were all increased by this percentage (Table S1). These revised N2O concentration values were then carried forward to further analysis.

Table S1. Corrections applied to the samples from each month depending on the amount of time vials were stored.

Month sampled	Weeks stored	N_2O concentration (ppm)	% Correction
April	30	0.9753	15.3
May	26	0.9683	15.91
June	21	1.0593	8.01
July	17	1.0972	4.71
-	0	1.1515	-

2. Supplementary results

Table S2. Details of the statistical tests used for each data set including normality of data, the tests used, and the resultant test and *P* values.

Data	Parametric/Non-parametric	Statistical Test	Test Statistic	*P* value
Monthly comparison of N_2O stem fluxes from landfill site (all stem heights)	Non-parametric	Friedman test (post-hoc: Wilcoxon signed-rank test)	64.81	$P < 0.01$
Monthly comparison of N_2O stem fluxes from landfill site (30 cm stem height)	Parametric (After sqrt transform)	Repeated measures ANOVA (post-hoc: paired t-test)	28.80	$P < 0.01$
Monthly comparison of N_2O stem fluxes from landfill site (90 cm stem height)	Parametric	Repeated measures ANOVA (post-hoc: paired t-test)	11.44	$P < 0.01$
Monthly comparison of N_2O stem fluxes from landfill site (150 cm stem height)	Non-parametric	Friedman test (post-hoc: Wilcoxon signed-rank test)	30.44	$P < 0.01$
Monthly comparison of N_2O soil fluxes from landfill site	Parametric (After sqrt transform)	Repeated measures ANOVA (post-hoc: paired t-test)	31.51	$P < 0.01$
Comparison of N_2O fluxes at different stem heights from the landfill site (all months)	Non-parametric	Kruskal-Wallis test	4.68	$P = 0.092$
Comparison of N_2O fluxes at different stem heights from the landfill site (April 2021)	Parametric	One-way ANOVA	0.25	$P > 0.05$
Comparison of N_2O fluxes at different stem heights from the landfill site (May 2021)	Parametric	One-way ANOVA	0.28	$P = 0.070$
Comparison of N_2O fluxes at different stem heights from the landfill site (June 2021)	Parametric (After sqrt transform)	One-way ANOVA	0.39	$P > 0.05$
Comparison of N_2O fluxes at different stem heights from the landfill site (July 2021)	Parametric (After sqrt transform)	One-way ANOVA	0.67	$P > 0.05$
Comparison of N_2O stem fluxes between the landfill and non-landfill sites (all stem heights)	Non-parametric	Mann-Whitney U test	11545.00	$P < 0.01$
Comparison of N_2O stem fluxes between the landfill and non-landfill sites (30 cm stem height)	Parametric (After sqrt transform)	t-test	2.42	$P < 0.05$
Comparison of N_2O stem fluxes between the landfill and non-landfill sites (90 cm stem height)	Non-parametric	Mann-Whitney U test	1136.00	$P < 0.01$
Comparison of N_2O stem fluxes between the landfill and non-landfill sites (150 cm stem height)	Parametric	t-test	1.66	$P = 0.071$
Comparison of N_2O soil fluxes between the landfill and non-landfill sites	Non-parametric	Mann-Whitney U test	485.00	$P < 0.01$

Table S3. Summary of N_2O fluxes from tree stem and soil chambers on closed landfill site between April and July 2021. SE is standard error and n is the number of measurements.

		N_2O (μg m^{-2} h^{-1})			
	Month	Average	SE	Range	n
Tree Stem (all heights)	Apr-21	0.31	0.12	3.43	45
	May-21	0.50	0.11	3.66	45
	Jun-21	0.80	0.15	3.98	45
	Jul-21	0.90	0.11	3.80	45
Tree Stem (30 cm measurement height)	Apr-21	0.41	0.21	2.37	15
	May-21	0.83	0.25	3.46	15
	Jun-21	0.95	0.26	3.64	15
	Jul-21	1.06	0.22	3.34	15
Tree Stem (90 cm measurement height)	Apr-21	0.32	0.16	2.43	15
	May-21	0.43	0.10	1.37	15
	Jun-21	0.89	0.28	3.35	15
	Jul-21	0.81	0.21	2.76	15
Tree Stem (150 cm measurement height)	Apr-21	0.21	0.24	3.43	15
	May-21	0.24	0.14	1.83	15
	Jun-21	0.57	0.22	3.61	15
	Jul-21	0.82	0.12	1.65	15
Soil	Apr-21	18.36	4.03	44.17	15
	May-21	45.12	11.68	150.21	15
	Jun-21	17.10	3.83	54.42	15
	Jul-21	21.30	5.39	69.28	15

Environmental controls on N_2O fluxes

At the 30 cm measurement height, none of the measured ancillary variables significantly accounted for the variance in N_2O flux (Supplementary Table 4). Stem N_2O fluxes at 90 and 150 cm were best explained by air temperature, which accounted for *c.*6% of the variation (Supplementary Table 4). Variation in soil N_2O fluxes at the landfill site was best explained by air pressure (12%). The averages and ranges of the measured environmental variables for the landfill and non-landfill site are in Supplementary Table 5. Average rainfall and precipitation patterns at the field site are shown in Supplementary Table 6.

Table S4. Results of stepwise regression analysis showing the relationships between N_2O fluxes and environmental variables at the landfill site.

Sampling location	Adjusted R^2	Explanatory variables	*P* value	Standardised β coefficient
Stem 30 cm	-	-	-	-
Stem 90 cm	0.06	Air temperature	$P < 0.05$	0.275
Stem 150 cm	0.057	Air temperature	$P < 0.05$	0.269
Soil	0.121	Air pressure	$P < 0.01$	–0.369

Table S5. Average, minimum, maximum and ranges of measured environmental variables at the landfill and non-landfill sites.

Field site	Environmental variable	Average	Minimum	Maximum	Range
Landfill	Air temperature (°C)	18.1	9.7	26.5	16.8
	Air pressure (hPa)	1014	999.7	1022.4	22.7
	Stem temperature (°C)	16.9	10.3	26.5	16.3
	Soil temperature (°C)	13.2	7.1	19.4	12.3
	Soil moisture (%)	20.9	7.1	42.8	35.7
	Stem DBH (cm)	18.3	11.1	40.5	29.4
Non-landfill	Air temperature (°C)	17	6.5	28.8	22.3
	Air pressure (hPa)	1007.3	989.3	1099.1	109.8
	Stem temperature (°C)	15.3	6.8	26.2	19.4
	Soil temperature (°C)	12.3	6.6	18.7	12.1
	Soil moisture (%)	32	10.2	48.6	38.4
	Stem DBH (cm)	16.1	12.7	21.4	8.7

Table S6. Average monthly UK rainfall and rainfall patterns at site 1A. Average soil moisture and air temperature at site 1A for each month in the 2021 measurement period.

Month	UK rainfall (mm)	Soil moisture (%)	Air temperature (°C)	Rainfall at field site
April	5	20.1	15	No rain for 8 days preceding measurements
May	112	32.8	15	Rainfall every day for 5 days preceding measurements (including high rainfall event 5 days before)
June	48	14.9	22.6	No rain for 10 days preceding measurements
July	78	15.5	19.7	No rain for 3 days preceding measurements

Table S7. Summary of N_2O fluxes from a closed landfill and a comparable non-landfill area. SE is standard error and n is the number of measurements.

		N_2O (μg m^{-2} h^{-1})			
	Measurement Type	Average	SE	Range	n
Closed landfill site	Tree stem all heights	0.627	0.062	4.82	180
	Tree stem 30cm	0.811	0.119	4.39	60
	Tree stem 90cm	0.612	0.101	3.35	60
	Tree stem 150cm	0.457	0.097	4.56	60
	Soil	25.48	3.72	153.7	60
Non-landfill site	Tree stem all heights	0.264	0.052	5.17	180
	Tree stem 30cm	0.402	0.103	4.21	60
	Tree stem 90cm	0.16	0.087	3.49	60
	Tree stem 150cm	0.229	0.078	3.57	60
	Soil	3.53	0.583	23.43	60

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