

1 **S1. Potential enzyme activity method**

2 Potential activity of seven enzymes associated with C, N and P mineralisation were determined for bulk and
3 rhizosphere soil respectively. For this we used fluorometrically labelled substrates, following the method of Bell
4 et al., 2013. Two g frozen soil was mixed to a slurry (1:33 w:v) with MilliQ water in a laboratory blender for one
5 minute. The slurry was pipetted into 96 well plates with three technical replicates and given fluorescent substrates
6 (4-methylumbelliferone; MUB and 7-amino-4-methylcoumarin: MUC) in accordance with the Bell et al. protocol
7 (2013). The samples were then incubated at 25 °C for three hours and analysed for fluorescence with a
8 CLARIOstar plate reader (BMG LABTECH GmbH, Germany). Four enzymes (α -D-glucopyranoside (AG), β -D-
9 glucopyranoside (BG), β -D-cellobioside (CB), and β -D-xylopyranoside (XYL)) targeted C-rich compounds
10 (sugar, cellulose, hemicellulose), two enzymes (L-Leucine-7-aminopeptidase (LAP) and N-acetyl- β -D-
11 glucosamine (NAG)) targeted N-rich compounds (proteins and chitin), and acid phosphatase (PHOS) targeted
12 organic compounds with P. These enzymes are considered representative of the total enzyme pool active in the
13 soil.

14 **S2. Enzymatic activity results**

15 Enzyme activities decreased significantly with depth but did not differ significantly between soil or CO₂ treatment
16 (Table S2.3 and S2.4). One exception to the general trend was CB (β -D-cellobioside) that did not decrease with
17 depth and was significantly higher in rhizosphere soil compared to bulk soil. Notable is the difference in
18 magnitude for N targeting and P targeting enzymes where P enzymes were twice as abundant than N. The two
19 to one pattern was maintained as the enzyme activity declined with soil depth.

21 **Table S1.** Potential enzyme activity and stoichiometry of enzymes targeting C, N and P compounds ($\mu\text{mol h}^{-1} \text{g}^{-1}$) for bulk and rhizosphere soil of a mature Eucalyptus forest
 22 soil exposed to ambient and elevated CO_2 for three depths (0 to 10 cm, 10 to 30 cm, transition), with standard error in parenthesis. Four enzymes (α -D-glucopyranoside (AG),
 23 β -D-glucopyranoside (BG), β -D-cellobioside (CB), and β -D-xylopyranoside (XYL)) targeted C-rich compounds (sugar, cellulose, hemicellulose), two enzymes (L-Leucine-7-
 24 aminopeptidase (LAP) and N-acetyl- β -D-glucosamine (NAG)) targeted N-rich compounds (proteins and chitin), and acid phosphatase (PHOS) targeted organic compounds
 25 with P.

Layer	Enzyme							Sum			Stoichiometry			pH
	AG	BG	CB	XYL	LAP	NAG	PHOS	C	N	P	C:N	C:P	N:P	
Bulk Ambient														
<i>0-10</i>	5.3 (1)	38.9 (7.9)	16.4 (3.3)	23.5 (5.1)	33.8 (11.5)	32.1 (5.3)	121.9 (27.3)	84 (14.1)	65.9 (12.5)	121.9 (27.3)	1.5 (0.3)	0.8 (0.1)	0.7 (0.2)	5.8 (0.1)
<i>10-30</i>	3.5 (1)	9.5 (1.7)	4.1 (1)	6.6 (1.2)	16.3 (4.3)	10.4 (0.8)	47.6 (10.2)	23.6 (4)	26.8 (4.6)	47.6 (10.2)	1.2 (0.4)	0.8 (0.3)	0.6 (0)	6 (0.1)
<i>transition</i>	1.6 (0.6)	2.5 (1)	1.1 (0.4)	1.4 (0.5)	9.3 (1.8)	5.2 (1.4)	25 (6.3)	6.6 (2.3)	14.5 (2.7)	25 (6.3)	0.7 (0.3)	0.3 (0.1)	0.7 (0.2)	5.8 (0.1)
Bulk Elevated														
<i>0-10</i>	5.3 (1.3)	35.8 (11.3)	12.5 (3.9)	20.9 (6.7)	23.8 (7.5)	31.7 (10.1)	139.5 (52)	74.5 (22.3)	55.5 (15.4)	139.5 (52)	1.4 (0.2)	0.7 (0.2)	0.5 (0.1)	5.7 (0.2)
<i>10-30</i>	5.8 (1.6)	15.4 (5.7)	6.9 (2)	11.1 (2.7)	13.7 (3.3)	17 (4)	65.9 (18)	39.2 (10.5)	30.7 (5.8)	65.9 (18)	1.4 (0.3)	0.8 (0.3)	0.6 (0.1)	5.9 (0.1)
<i>transition</i>	4.6 (1.3)	7.3 (1.8)	4.7 (1.2)	5.2 (1.2)	3.4 (1.1)	16.1 (9.3)	23.6 (5.2)	21.7 (4.5)	19.5 (10.1)	23.6 (5.2)	2 (0.5)	1.1 (0.3)	0.7 (0.2)	6.1 (0.2)
Rhizosphere Ambient														
<i>0-10</i>	5.2 (1.7)	52.4 (17.7)	16.3 (3.1)	21.8 (6.6)	33.6 (13.4)	35.6 (9)	119.9 (33.4)	95.7 (26.8)	69.2 (14.1)	119.9 (33.4)	1.6 (0.4)	0.8 (0.1)	0.7 (0.2)	5.9 (0.1)
<i>10-30</i>	5.3 (1.3)	12.5 (1.4)	7.7 (1.6)	9.9 (1.3)	16.5 (4.4)	13.5 (1.8)	61.4 (13)	35.5 (4.4)	30 (4.9)	61.4 (13)	1.4 (0.3)	0.9 (0.3)	0.5 (0.1)	5.9 (0.1)
<i>transition</i>	4.3 (1.6)	12.3 (6.1)	6.5 (3.4)	9.4 (4.1)	13.3 (2.4)	19.7 (10.2)	56.2 (13.9)	32.4 (14.5)	33 (11.6)	56.2 (13.9)	1 (0.3)	0.5 (0.1)	0.6 (0.1)	5.7 (0.1)
Rhizosphere Elevated														

Layer	Enzyme							Sum			Stoichiometry			pH
	AG	BG	CB	XYL	LAP	NAG	PHOS	C	N	P	C:N	C:P	N:P	
<i>0-10</i>	3.9 (1.2)	34.4 (8.1)	12.4 (3.5)	20.1 (4.3)	25.1 (7.4)	29.7 (6.9)	126.1 (40.6)	70.8 (16.3)	54.8 (12.9)	126.1 (40.6)	1.3 (0.1)	0.7 (0.1)	0.5 (0.1)	5.7 (0.2)
<i>10-30</i>	6.6 (2.1)	17.8 (3.2)	6.8 (1)	11.4 (1.4)	16 (2.6)	23.9 (4)	97.1 (24.6)	42.6 (4.3)	40 (5.7)	97.1 (24.6)	1.2 (0.2)	0.7 (0.2)	0.5 (0.1)	5.8 (0.1)
<i>transition</i>	4.5 (1.3)	17.2 (3.8)	10.4 (3.5)	6.3 (1.5)	5.4 (1.1)	32.1 (15.5)	53.1 (16.8)	38.3 (5.2)	37.5 (15.8)	53.1 (16.8)	1.4 (0.3)	0.9 (0.2)	0.8 (0.3)	6 (0.3)

27 **Table S2:** Model F statistic and significance levels for potential enzyme activity. Significance of P values are in
 28 bold and as indicated: *** indicate P < 0.001; ** indicate P < 0.01 and * indicates P < 0.05.

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	AG	BG	CB	XYL	LAP	NAG	PHOS	sum			stoichiometry			
								C	N	P	C:N	C:P	N:P	pH
CO ₂	0.98	0	0.01	0.03	0.8	1.55	0.19	0.02	0	0.19	1.53	0.72	0.14	0.03
depth	1.45	23.28 ***	18.44 ***	22.84 ***	11.96 ***	6.37 **	17.62 ***	24.2 ***	14.41 ***	17.62 ***	0.51	0.48	0.73	0.67
soil	0.9	2.42	3.05 (.)	0.83	0.22	2.59	1.48	2.43	2.03	1.48	0	0	0	0.17
CO ₂ :depth	1.25	1.77	2.81 (.)	0.57	0.42	1.16	0.42	1.83	1.13	0.42	3.3 *	4.42 *	1.03	2.94 (.)
CO ₂ :soil	1.01	0.38	0.15	0.43	0.01	0	0.01	0.51	0	0.01	1.84	1.13	0.04	0.04
depth:soil	1.02	0.27	1.56	0.81	0.06	1.01	0.96	0.59	0.74	0.96	0.03	0	0.05	0.38
CO ₂ :depth:soil	0.07	0.41	0.29	0.25	0.02	0.12	0.12	0.04	0.06	0.12	0.34	0.22	0.07	0

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