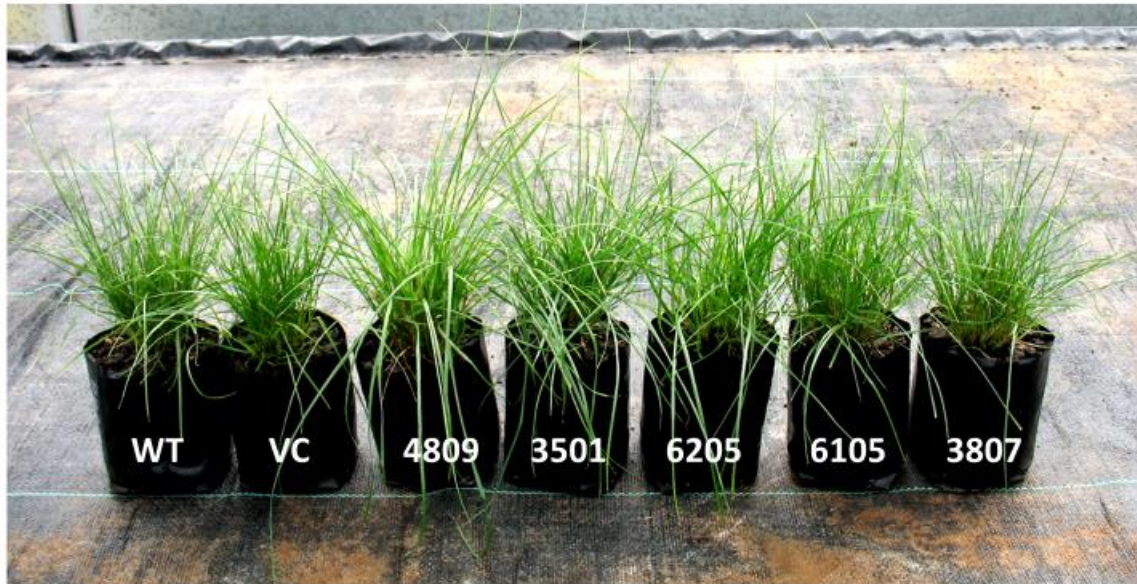
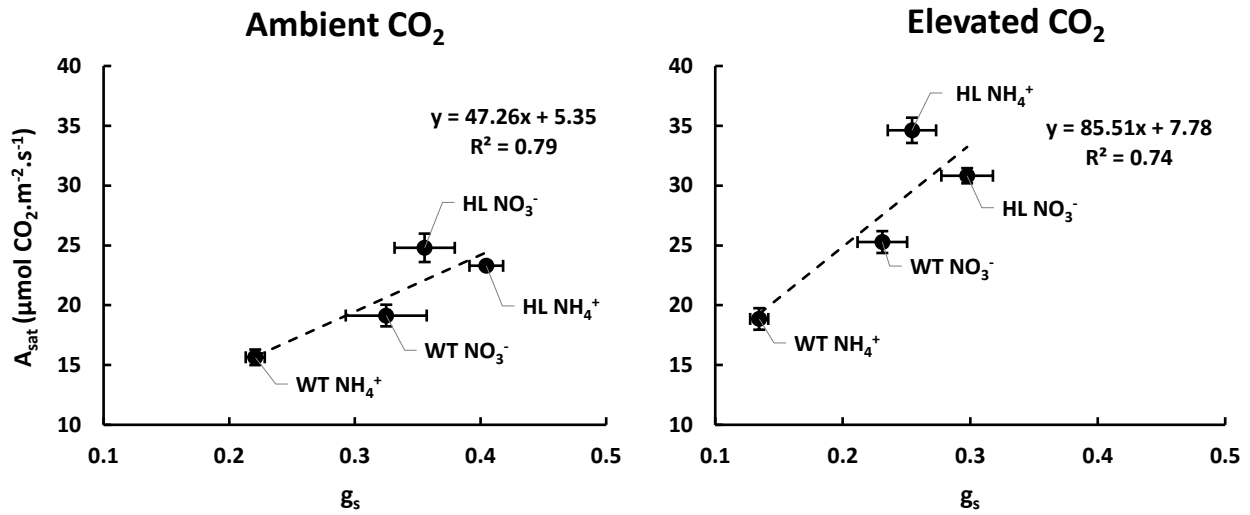


Supplementary Figure S1. sheath and root DW of a defoliated clonal cys-OLE/DGAT ryegrass transformant (HL) and a wild type control (WT) genotype. Plants were established from 3-4 tillers for 23 days at 2mM NO_3^- supply at ambient CO_2 . Bars represent the average for each genotype ($n=5$) \pm S.E. * = denotes a significant difference at the $p<0.05$ level in DW, according to student's t test.



Supplemental Figure S2. visual comparison of shoot regrowth of *cys-OLE/DGAT* transformants with a WT and VC genotype. Ramets consisting of 5 tillers were placed in pots and trimmed to an even height every 3 weeks for 3 months.



Supplementary Figure S3. relationship between light saturated photosynthetic rate per unit leaf area (A_{sat}) and stomatal conductance (g_s) of a clonal *cys*-OLE/DGAT ryegrass transformant (HL) and a wild type control (WT) genotype. Plants were regrown for 28-29 days after defoliation at 7.5 mM N supply at either ambient (400 ppm) and elevated CO₂ (760 ppm). Data points represent the averages of plants grown under NO₃⁻ or NH₄⁺ ($n=5$) \pm S.E.

A)	Γ^* experiment		B)	F_v/F_m experiment		
	Γ^* (ppm)	R_i ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)		F_v/F_m	$\Phi \text{ PSII}$	A_{area} ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)
WT	27.87 ± 1.48	0.71 ± 0.07	0.79 ± 0.003	0.48 ± 0.01	17.83 ± 0.40	
HL	35.50 ± 1.11	0.41 ± 0.05	0.80 ± 0.002	0.51 ± 0.01	20.63 ± 0.89	
p value	**	**	*	**	*	

Supplementary Table S1. CO₂ compensation point in the absence of dark respiration in the light (Γ^*) and F_v/F_m measured alongside the quantum efficiency of photosystem II ($\Phi \text{ PSII}$) and photosynthesis (A_{area}) of a clonal cys-OLE/DGAT ryegrass transformant (HL) and a wild type control (WT) genotype. Measurements were taken in two separate regrowth experiments, approximately 3 weeks after the defoliation of vegetative clones grown in potting mix. Values represent raw averages ± S.E. ($n=6$ for Γ^* experiment, $n=10$ for F_v/F_m experiment). p values are from Welch two sample t-tests or Wilcoxon rank sum test. * = $p < 0.05$, ** = $p < 0.01$.

Genotype	Total leaf FA (%DW)	Leaf TAG (%DW)	Total root FA (%DW)
WT	3.49 ± 0.07 A	0.18 ± 0.03 A	0.66 ± 0.01 A
VC	3.50 ± 0.13 A	0.23 ± 0.02 A	0.73 ± 0.01 B
3501	5.56 ± 0.06 B	2.20 ± 0.06 B	0.99 ± 0.01 C
3807	6.69 ± 0.07 C	2.47 ± 0.06 C	0.97 ± 0.01 C
p value	**	***	***

Supplementary Table S2. total leaf fatty acid (FA), leaf triacylglycerol (TAG) and total root FA of a wild type (WT) and vector control (VC) and two independent *cys-OLE/DGAT* ryegrass transformants, from leaves 3 weeks after cutting and roots 3 months after propagation. Values are expressed on a %DW basis and represent raw averages ± S.E. ($n=4-8$). ** = $p < 0.01$, *** = $p < 0.001$ effect significant according to Kruskal-Wallis test. Different letters indicate statistically significant differences in predicted means obtained from Wilcoxon rank sum test, with p values adjusted according to BH method.

CO ₂	Genotype	V _{cmax} ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)	J ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)	g _m ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}\cdot\text{pa}^{-1}$)	V _{cmax} /J	Model fit
Ambient	WT	161.78 ± 9.77 A	178.08 ± 8.92 A	1.39 ± 0.17 A	0.91 ± 0.03 A	0.41 ± 0.10
	HL	112.86 ± 7.46 BC	171.73 ± 6.07 A	4.29 ± 0.51 B	0.65 ± 0.02 C	0.51 ± 0.13
Elevated	WT	122.48 ± 4.74 B	160.76 ± 3.26 A	1.30 ± 0.09 A	0.76 ± 0.02 B	1.56 ± 0.24
	HL	97.46 ± 5.24 C	165.66 ± 4.71 A	5.17 ± 1.18 B	0.59 ± 0.02 C	2.39 ± 0.68
ANOVA	G	***	-	***	***	
	CO ₂	**	-	-	**	
	GxCO ₂	-	-	-	-	

Supplementary Table S3. maximum velocity of rubisco carboxylation (V_{c,max}), rate of electron transport (J), and mesophyll conductance to CO₂ (g_m) derived from modelling the response of net photosynthesis per unit leaf area (A) to intracellular CO₂ concentration (C_i) of a clonal cys-OLE/DGAT ryegrass transformant (HL) and a wild type control (WT) genotype. Plants were regrown at 5mM NO₃⁻ supply under ambient CO₂ (400 ppm) and at 7.5mM NO₃⁻ supply under elevated CO₂ (760 ppm). Values represent raw averages (n=5) ± S.E. G = genotype effect, CO₂ = CO₂ effect significant in a two-way ANOVA. * = p<0.05, ** = p<0.01, *** = p<0.001. Different letters indicate statistically significant differences in predicted means obtained from two-way ANOVA, with p values adjusted according to BH method.

CO ₂	Genotype	SLA (cm ² .g.DW ⁻¹)	A _{sat} (μmol CO ₂ .m ⁻² .s ⁻¹)	Leaf FA (mg/gDW)	Leaf WSC (mg/gDW)
Ambient	WT	249 ± 15 BC	19.8 ± 1.5 C	24.8 ± 0.76 C	156.2 ± 18.2 B
	3501	310 ± 24 AB	26.0 ± 0.6 B	41.1 ± 2.02 AB	ND
	6205 (HL)	369 ± 12 A	27.1 ± 2.3 B	46.2 ± 0.56 A	86.9 ± 13.8 C
Elevated	WT	196 ± 9 C	22.3 ± 1.9 BC	22.7 ± 0.07 C	236.1 ± 10.3 A
	3501	266 ± 22 B	36.4 ± 1.5 A	38.8 ± 2.41 B	ND
	6205 (HL)	300 ± 8 B	37.0 ± 1.6 A	43.6 ± 1.34 AB	97.2 ± 5.6 C
ANOVA	G	***	***	***	***
	CO ₂	**	***	-	**
	GxCO ₂	-	*	-	*

Supplementary Table S4. specific leaf area (SLA), light saturated photosynthetic rate per unit leaf area (A_{sat}), leaf fatty acid (FA) and leaf water soluble carbohydrates (WSC) of two clonal cys-OLE/DGAT ryegrass lines; 3501 and 6205 (HL) and a wild type control (WT) genotype. Plants were regrown for 33-34 days after defoliation at 5 mM NO₃⁻ supply at either ambient (400 ppm) or elevated CO₂ (760 ppm). Data points represent raw averages ($n=4-6$) ± S.E. G = genotype effect, CO₂ = CO₂ effect significant in a two-way ANOVA. * = p<0.05, ** = p<0.01, *** = p<0.001. Different letters indicate statistically significant differences in predicted means obtained from two-way ANOVA, with p values adjusted according to BH method.