

Appendix 4- Raw Data

CHAPTER 3

Construct G⁴⁴⁸ A Transformant 1103 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
31/01/08C											
T454	0	0.398	0.393	0.396	0.396	791	199	9.93	37.5	0.75	13.2
	20'	0.297	0.297	0.295	0.296	593					
T110	0	0.342	0.339	0.342	0.341	682	-1	-0.03	18.4	0.368	-0.1
	20'	0.338	0.341	0.345	0.341	683					
G448A	0	0.372	0.378	0.371	0.374	747	62	3.10	23.3	0.466	6.7
	20'	0.345	0.341	0.342	0.343	685					
21/07/08A											
T454	0	0.382	0.387	0.392	0.387	645	168	8.42	39.4	0.788	10.7
	20'	0.287	0.287	0.284	0.286	477					
T110	0	0.437	0.444	0.435	0.439	731	1	0.06	62.2	1.244	0.0
	20'	0.440	0.447	0.427	0.438	730					
G448A	0	0.439	0.438	0.436	0.438	729	91	4.53	29.8	0.596	7.6
	20'	0.386	0.384	0.380	0.383	639					
13/08/08											
T454	0	0.313	0.300	0.300	0.304	507	109	5.47	19.8	0.396	13.8
	20'	0.235	0.241	0.240	0.239	398					
T110	0	0.317	0.310	0.319	0.315	526	-3	-0.17	11.4	0.228	-0.7
	20'	0.307	0.317	0.328	0.317	529					
G448A	0	0.396	0.398	0.394	0.396	792	76	3.80	24	0.48	7.9
	20'	0.357	0.359	0.358	0.358	716					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 31/01/08C	T454	T110	G448A
Weight of filter	0.1273	0.11	0.112
Dry weight of mycelium	0.0898	0.092	0.0887
Difference	0.0375	0.018	0.0233
Difference in μg	37.5	18.4	23.3
Date of experiment: 21/07/08A	T454	T110	G448A
Weight of filter	0.13	0.1534	0.1203
Dry weight of mycelium	0.0906	0.0912	0.0905
Difference	0.0394	0.0622	0.0298
Difference in μg	39.4	62.2	29.8
Date of experiment: 13/08/08	T454	T110	G448A
Weight of filter	0.1383	0.1935	0.1157
Dry weight of mycelium	0.1185	0.1823	0.0917
Difference	0.0198	0.0112	0.024
Difference in μg	19.8	11.2	24

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

G448A uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average G448A uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
6.7	7.6	7.9	7.4 ±0.66	13.2	10.7	13.8	12.6 ±1.67	-0.1	0.0	-0.7	-0.3 ±0.41
Comparison with the respective wild type (T454) result (%)											
50.0	71.0	57.0	Average: 60.0%								
Growth test^a											
G448A			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct G⁴⁵²A Transformant 3022 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
13/02/08B											
T454	0	0.374	0.374	0.377	0.375	750	120	6.00	25.7	0.514	11.7
	20'	0.312	0.318	0.315	0.315	630					
T110	0	0.389	0.372	0.381	0.381	761	-1	-0.07	32.7	0.654	-0.1
	20'	0.387	0.391	0.366	0.381	763					
G452A	0	0.455	0.435	0.444	0.445	889	83	4.13	34.8	0.696	5.9
	20'	0.402	0.403	0.405	0.403	807					
04/08/08B											
T454	0	0.312	0.310	0.303	0.308	514	104	5.19	25.1	0.502	10.3
	20'	0.243	0.247	0.248	0.246	410					
T110	0	0.316	0.322	0.316	0.318	530	-13	-0.64	43.2	0.864	-0.7
	20'	0.330	0.337	0.310	0.326	543					
G452A	0	0.330	0.334	0.335	0.333	555	43	2.14	17.2	0.344	6.2
	20'	0.307	0.310	0.305	0.307	512					
13/08/08											
T454	0	0.383	0.382	0.381	0.382	637	191	9.53	36.2	0.724	13.2
	20'	0.261	0.268	0.274	0.268	446					
T110	0	0.317	0.310	0.319	0.315	526	-3	-0.17	11.2	0.224	-0.7
	20'	0.307	0.317	0.328	0.317	529					
G452A	0	0.461	0.465	0.456	0.461	921	88	4.4	39.1	0.782	5.6
	20'	0.418	0.420	0.412	0.417	833					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 13/02/08B	T454	T110	G452A
Weight of filter	0.1156	0.123	0.1244
Dry weight of mycelium	0.0899	0.0903	0.0896
Difference	0.0257	0.0327	0.0348
Difference in μg	25.7	32.7	34.8
Date of experiment: 04/08/08B	T454	T110	G452A
Weight of filter	0.1177	0.1341	0.1069
Dry weight of mycelium	0.0926	0.0909	0.0897
Difference	0.0251	0.0432	0.0172
Difference in μg	25.1	43.2	17.2
Date of experiment: 13/08/08	T454	T110	G452A
Weight of filter	0.127	0.1935	0.1303
Dry weight of mycelium	0.0908	0.1823	0.0912
Difference	0.0362	0.0112	0.0391
Difference in μg	36.2	11.2	39.1

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

G452A uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average G452A uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
5.9	6.2	5.6	5.9 ±0.3	11.7	10.3	13.2	11.7 ±1.41	-0.1	-0.7	-0.7	-0.5 ±0.37
Comparison with the respective wild type (T454) result (%)											
51.0	60.0	43.0	Average: 51.0%								
Growth test^a											
G452A			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct G⁴⁵⁵A Transformant 2656 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
07/09/07A											
T454	0	0.329	0.323	0.329	0.327	654	211	10.53	46.4	0.928	11.4
	20'	0.224	0.220	0.221	0.222	443					
T110	0	0.356	0.354	0.355	0.355	710	3	0.13	68.9	1.378	0.1
	20'	0.352	0.358	0.351	0.354	707					
G455A	0	0.357	0.356	0.353	0.355	592	37	1.83	27.6	0.552	3.3
	20'	0.337	0.332	0.331	0.333	556					
07/09/07B											
T454	0	0.310	0.309	0.305	0.308	616	265	13.23	51.2	1.024	12.9
	20'	0.178	0.178	0.171	0.176	351					
T110	0	0.341	0.329	0.330	0.333	667	-2	-0.10	72.9	1.458	-0.1
	20'	0.345	0.321	0.337	0.334	669					
G455A	0	0.376	0.377	0.378	0.377	754	85	4.27	59.6	1.192	3.6
	20'	0.336	0.336	0.331	0.334	669					
08/02/08A											
T454	0	0.367	0.364	0.368	0.366	733	137	6.87	28.2	0.564	12.2
	20'	0.296	0.296	0.301	0.298	595					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
G455A	0	0.363	0.357	0.364	0.361	723	31	1.6	23	0.46	3.4
	20'	0.341	0.347	0.349	0.346	691					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 07/09/07A	T454	T110	G455A
Weight of filter	0.1382	0.16	0.119
Dry weight of mycelium	0.0918	0.091	0.091
Difference	0.0464	0.069	0.028
Difference in μg	46.4	68.9	27.6
Date of experiment: 07/09/07B	T454	T110	G455A
Weight of filter	0.144	0.1643	0.1518
Dry weight of mycelium	0.092	0.0914	0.0922
Difference	0.051	0.0729	0.0596
Difference in μg	51.2	72.9	59.6
Date of experiment:08/02/08A	T454	T110	G455A
Weight of filter	0.1196	0.1204	0.1145
Dry weight of mycelium	0.0914	0.0925	0.0915
Difference	0.0282	0.0279	0.023
Difference in μg	28.2	27.9	23

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

G455A uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average G455A uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
3.3	3.6	3.4	3.4 ±0.13	11.4	12.9	12.2	12.1 ±0.79	0.1	-0.1	-0.5	-0.1 ±0.3
Comparison with the respective wild type (T454) result (%)											
29.0	28.0	28.0	Average: 28.0%								
Growth test^a											
G455A			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct G⁴⁵⁶A Transformant 2761 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
31/01/08C											
05/10/07A											
T454	0	0.305	0.297	0.297	0.300	499	89	4.44	18.2	0.364	12.2
	20'	0.245	0.240	0.254	0.246	411					
T110	0	0.310	0.310	0.315	0.312	519	-8	-0.42	22.7	0.454	-0.9
	20'	0.315	0.310	0.325	0.317	528					
G456A	0	0.443	0.422	0.435	0.433	867	12	0.60	29.7	0.594	1.0
	20'	0.433	0.420	0.429	0.427	855					
05/10/07B											
T454	0	0.433	0.432	0.430	0.432	863	139	6.97	35.4	0.708	9.8
	20'	0.365	0.361	0.360	0.362	724					
T110	0	0.444	0.441	0.442	0.442	885	0	0.00	26.1	0.522	0.0
	20'	0.445	0.439	0.443	0.442	885					
G456A	0	0.431	0.433	0.431	0.432	863	18	0.90	29.6	0.592	1.5
	20'	0.423	0.424	0.421	0.423	845					
15/10/07											
T454	0	0.374	0.364	0.368	0.369	737	142	7.10	28.2	0.564	12.6
	20'	0.296	0.296	0.301	0.298	595					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
G456A	0	0.323	0.324	0.328	0.325	542	18	0.89	29.6	0.592	1.5

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 05/10/07A	T454	T110	G456A
Weight of filter	0.109	0.114	0.124
Dry weight of mycelium	0.0908	0.092	0.094
Difference	0.0182	0.023	0.03
Difference in μg	18.2	22.7	29.7
Date of experiment: 05/10/07B	T454	T110	G456A
Weight of filter	0.1293	0.1207	0.1216
Dry weight of mycelium	0.0939	0.0946	0.092
Difference	0.0354	0.0261	0.0296
Difference in μg	35.4	26.1	29.6
Date of experiment: 08/02/08C	T454	T110	G456A
Weight of filter	0.1196	0.1204	0.1216
Dry weight of mycelium	0.0914	0.0925	0.092
Difference	0.0282	0.0279	0.0296
Difference in μg	28.2	27.9	29.6

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

G456A uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average G456A uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
1.0	1.5	1.5	1.3 ±0.29	12.2	9.8	12.6	11.5 ±1.49	-0.9	0.0	-0.5	-0.5 ±0.46
Comparison with the respective wild type (T454) result (%)											
8.0	15.0	12.0	Average: 12.0%								
Growth test^a											
G456A			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F⁴⁵⁷A Transformant 4661 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
05/02/09											
T454	0	0.858	0.862	0.885	0.868	1737	177	8.83	32.4	0.648	13.6
	20'	0.778	0.782	0.78	0.780	1560					
T110	0	0.317	0.319	0.318	0.318	636	1	0.07	22.4	0.448	0.1
	20'	0.315	0.320	0.317	0.317	635					
F457A	0	0.915	0.915	0.915	0.915	1830	150	7.50	36.4	0.728	10.3
	20'	0.87	0.82	0.83	0.840	1680					
05/02/09											
T454	0	0.298	0.288	0.285	0.290	484	117	5.86	25.7	0.514	11.4
	20'	0.223	0.223	0.214	0.220	367					
T110	0	0.286	0.327	0.304	0.306	509	-8	-0.42	21	0.42	-1.0
	20'	0.297	0.314	0.321	0.311	518					
F457A	0	0.854	0.871	0.843	0.856	1712	97	4.87	25.4	0.508	9.6
	20'	0.783	0.819	0.82	0.807	1615					
05/02/09											
T454	0	0.287	0.298	0.303	0.296	493	117	5.83	25.1	0.502	11.6
	20'	0.223	0.227	0.228	0.226	377					
T110	0	0.416	0.402	0.399	0.406	676	-12	-0.58	43.2	0.864	-0.7
	20'	0.400	0.416	0.422	0.413	688					
F457A	0	0.86	0.881	0.852	0.864	1729	114	5.70	25.4	0.508	11.2
	20'	0.783	0.819	0.82	0.807	1615					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 05/02/09	T454	T110	F457A
Weight of filter	0.107	0.1119	0.1005
Dry weight of mycelium	0.0746	0.0755	0.0751
Difference	0.0324	0.0364	0.0254
Difference in μg	32.4	36.4	25.4
Date of experiment: 05/02/09	T454	T110	F457A
Weight of filter	0.1166	0.1124	0.1142
Dry weight of mycelium	0.0909	0.0914	0.0907
Difference	0.0257	0.021	0.0235
Difference in μg	25.7	21	23.5
Date of experiment: 05/02/09	T454	T110	F457A
Weight of filter	0.1177	0.1341	0.1205
Dry weight of mycelium	0.0926	0.0909	0.0902
Difference	0.0251	0.0432	0.0303
Difference in μg	25.1	43.2	30.3

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F457A uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F457A uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
10.3	11.2	9.6	10.3 ±0.82	13.6	11.4	11.6	12.2 ±0.95	0.1	-1.0	-0.7	-0.5 ±0.59
Comparison with the respective wild type (T454) result (%)											
5.0	9.0	3.0	Average: 84.0%								
Growth test ^a											
F457A			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct G⁴⁵⁸A Transformant 10 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
13/06/07B											
T454	0	0.341	0.353	0.351	0.348	697	189	9.47	45	0.9	10.5
	20'	0.250	0.256	0.255	0.254	507					
T110	0	0.366	0.359	0.358	0.361	722	4	0.20	46.9	0.938	0.2
	20'	0.363	0.356	0.358	0.359	718					
G458A	0	0.360	0.360	0.358	0.359	719	47	2.37	21	0.42	5.6
	20'	0.333	0.335	0.339	0.336	671					
30/10/07A											
T454	0	0.340	0.335	0.336	0.337	674	181	9.03	42	0.84	10.8
	20'	0.252	0.245	0.243	0.247	493					
T110	0	0.342	0.347	0.343	0.344	688	-9	-0.43	28	0.56	-0.8
	20'	0.345	0.347	0.353	0.348	697					
G458A	0	0.400	0.407	0.402	0.403	672	48	2.42	21.3	0.426	5.7
	20'	0.372	0.375	0.375	0.374	623					
30/10/07B											
T454	0	0.394	0.401	0.405	0.400	800	153	7.67	42	0.84	9.1
	20'	0.322	0.325	0.323	0.323	647					
T110	0	0.342	0.347	0.343	0.344	688	-9	-0.43	28	0.56	-0.8
	20'	0.345	0.347	0.353	0.348	697					
G458A	0	0.305	0.307	0.307	0.306	511	51	2.56	26.4	0.528	4.8
	20'	0.271	0.278	0.278	0.276	459					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 13/06/07B	T454	T110	G458A
Weight of filter	0.1971	0.228	0.186
Dry weight of mycelium	0.1521	0.181	0.165
Difference	0.045	0.047	0.021
Difference in μg	45	46.9	21
Date of experiment: 30/10/07A	T454	T110	G458A
Weight of filter	0.134	0.1205	0.1207
Dry weight of mycelium	0.092	0.0925	0.0994
Difference	0.042	0.028	0.0213
Difference in μg	42	28	21.3
Date of experiment: 30/10/07B	T454	T110	G458A
Weight of filter	0.1341	0.1205	0.1179
Dry weight of mycelium	0.0921	0.0925	0.0915
Difference	0.042	0.028	0.0264
Difference in μg	42	28	26.4

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

G458A uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average G458A uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
5.6	5.7	4.8	5.4 ±0.47	10.5	10.8	9.1	10.1 ±0.88	0.2	-0.8	-0.8	-0.4 ±0.57
Comparison with the respective wild type (T454) result (%)											
54.0	53.0	53.0	Average: 53.0%								
Growth test^a											
G458A			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct L⁴⁶⁰ A Transformant 4568 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
06/02/09											
T454	0	0.858	0.862	0.885	0.868	1737	177	8.83	32.4	0.648	13.6
	20'	0.778	0.782	0.78	0.780	1560					
T110	0	0.317	0.319	0.318	0.318	636	1	0.07	22.4	0.448	0.1
	20'	0.315	0.320	0.317	0.317	635					
F460A	0	0.87	0.833	0.811	0.838	1676	53	2.63	29.8	0.596	4.4
	20'	0.82	0.802	0.813	0.812	1623					
06/02/09											
T454	0	0.298	0.288	0.285	0.290	484	117	5.86	25.7	0.514	11.4
	20'	0.223	0.223	0.214	0.220	367					
T110	0	0.286	0.327	0.304	0.306	509	-8	-0.42	21	0.42	-1.0
	20'	0.297	0.314	0.321	0.311	518					
F460A	0	0.928	0.864	0.849	0.880	1761	75	3.77	41.4	0.828	4.5
	20'	0.859	0.834	0.835	0.843	1685					
06/02/09											
T454	0	0.287	0.298	0.303	0.296	493	117	5.83	25.1	0.502	11.6
	20'	0.223	0.227	0.228	0.226	377					
T110	0	0.416	0.402	0.399	0.406	676	-12	-0.58	43.2	0.864	-0.7
	20'	0.400	0.416	0.422	0.413	688					
F460A	0	0.924	0.861	0.845	0.877	1753	68	3.40	41.4	0.828	4.1
	20'	0.859	0.834	0.835	0.843	1685					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 06/02/09	T454	T110	F460A
Weight of filter	0.107	0.1119	0.1172
Dry weight of mycelium	0.0746	0.0755	0.0758
Difference	0.0324	0.0364	0.0414
Difference in µg	32.4	36.4	41.4
Date of experiment: 06/02/09	T454	T110	F460A
Weight of filter	0.1166	0.1124	0.105
Dry weight of mycelium	0.0909	0.0914	0.0752
Difference	0.0257	0.021	0.0298
Difference in µg	25.7	21	29.8
Date of experiment: 06/02/09	T454	T110	F460A
Weight of filter	0.1177	0.1341	0.1205
Dry weight of mycelium	0.0926	0.0909	0.0902
Difference	0.0251	0.0432	0.0303
Difference in µg	25.1	43.2	30.3

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F460A uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F460A uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
4.5	4.4	4.1	4.3 ±0.22	13.6	11.4	11.6	12.2 ±0.95	0.1	-1.0	-0.7	-0.5 ±0.59
Comparison with the respective wild type (T454) result (%)											
5.0	9.0	3.0	Average: 35.0%								
Growth test^a											
F460A			T454				T110				
++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct G⁴⁶¹A Transformant 42 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
04/06/07A											
T454	0	0.339	0.348	0.346	0.344	689	177	8.87	48.2	0.964	9.2
	20'	0.251	0.259	0.257	0.256	511					
T110	0	0.347	0.377	0.359	0.361	722	-13	-0.63	53.6	1.072	-0.6
	20'	0.362	0.372	0.368	0.367	735					
G461A	0	0.410	0.409	0.412	0.410	821	-7	-0.33	66.5	1.33	-0.3
	20'	0.415	0.416	0.410	0.414	827					
04/06/07B											
T454	0	0.312	0.301	0.317	0.310	620	217	10.83	57.8	1.156	9.4
	20'	0.205	0.199	0.201	0.202	403					
T110	0	0.312	0.309	0.315	0.312	624	1	0.07	49.3	0.986	0.1
	20'	0.318	0.303	0.313	0.311	623					
G461A	0	0.338	0.327	0.332	0.332	665	17	0.87	78.7	1.574	0.6
	20'	0.327	0.324	0.320	0.324	647					
08/02/08B											
T454	0	0.340	0.364	0.368	0.357	715	119	5.97	28.2	0.564	10.6
	20'	0.296	0.296	0.301	0.298	595					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
G461A	0	0.365	0.369	0.362	0.365	731	2	0.1	21	0.42	0.2
	20'	0.361	0.364	0.368	0.364	729					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 04/06/07A	T454	T110	G461A
Weight of filter	0.2563	0.2331	0.277
Dry weight of mycelium	0.2081	0.1795	0.211
Difference	0.0482	0.0536	0.067
Difference in μg	48.2	53.6	66.5
Date of experiment: 04/06/07B	T454	T110	G461A
Weight of filter	0.269	0.2303	0.3155
Dry weight of mycelium	0.211	0.181	0.2368
Difference	0.058	0.0493	0.0787
Difference in μg	57.8	49.3	78.7
Date of experiment: 08/02/08B	T454	T110	G461A
Weight of filter	0.1196	0.1204	0.1139
Dry weight of mycelium	0.0914	0.0925	0.0929
Difference	0.0282	0.0279	0.021
Difference in μg	28.2	27.9	21

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

G461A uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average G461A uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
-0.3	0.6	0.2	0.2 ±0.4	9.2	9.4	10.6	9.7 ±0.75	-0.6	0.1	-0.5	-0.3 ±0.35
Comparison with the respective wild type (T454) result (%)											
0.0	5.9	2.3	Average: 2.7%								
Growth test^a											
G461A			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct G⁴⁶²A Transformant 2676 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
07/09/07A											
T454	0	0.329	0.333	0.339	0.334	667	224	11.20	46.4	0.928	12.1
	20'	0.224	0.220	0.221	0.222	443					
T110	0	0.376	0.384	0.378	0.379	759	0	0.00	68.9	1.378	0.0
	20'	0.382	0.381	0.375	0.379	759					
G462A	0	0.328	0.322	0.325	0.325	650	71	3.57	60.9	1.218	2.9
	20'	0.291	0.291	0.286	0.289	579					
07/09/07B											
T454	0	0.315	0.319	0.305	0.313	626	275	13.73	51.2	1.024	13.4
	20'	0.178	0.178	0.171	0.176	351					
T110	0	0.341	0.349	0.330	0.340	680	2	0.10	72.9	1.458	0.1
	20'	0.340	0.349	0.328	0.339	678					
G462A	0	0.393	0.392	0.388	0.391	782	89	4.43	83.4	1.668	2.7
	20'	0.349	0.348	0.343	0.347	693					
31/01/08A											
T454	0	0.359	0.353	0.360	0.357	715	149	7.43	37.5	0.75	9.9
	20'	0.287	0.277	0.285	0.283	566					
T110	0	0.338	0.337	0.336	0.337	562	3	0.17	43.2	0.864	0.2
	20'	0.336	0.335	0.334	0.335	558					
G462A	0	0.349	0.351	0.352	0.351	584	22	1.11	17.1	0.342	3.2
	20'	0.341	0.333	0.338	0.337	562					

Table containing the raw data obtained on three independent net nitrate uptake assay

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 07/09/07A	T454	T110	G462A
Weight of filter	0.1382	0.16	0.153
Dry weight of mycelium	0.0918	0.091	0.092
Difference	0.0464	0.069	0.061
Difference in μg	46.4	68.9	60.9
Date of experiment: 07/09/07B	T454	T110	G462A
Weight of filter	0.144	0.1643	0.1739
Dry weight of mycelium	0.092	0.0914	0.0905
Difference	0.051	0.0729	0.0834
Difference in μg	51.2	72.9	83.4
Date of experiment: 31/01/08A	T454	T110	G462A
Weight of filter	0.1273	0.1341	0.161
Dry weight of mycelium	0.0898	0.0909	0.1439
Difference	0.0375	0.0432	0.0171
Difference in μg	37.5	43.2	17.1

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

G462A uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average G462A uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
2.9	2.7	3.2	2.9 ±0.3	12.1	13.4	9.9	11.8 ±1.7	0.0	0.1	0.2	0.1 ±0.1
Comparison with the respective wild type (T454) result (%)											
24.0	20.0	33.0	Average: 26.0%								
Growth test^a											
G462A			T454			T110					
+++			+++			-					

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F⁴⁵⁷G Transformant 1334 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
29/10/07A											
T454	0	0.337	0.333	0.335	0.335	670	145	7.27	28	0.56	13.0
	20'	0.264	0.258	0.265	0.262	525					
T110	0	0.317	0.319	0.318	0.318	636	1	0.07	22.4	0.448	0.1
	20'	0.315	0.320	0.317	0.317	635					
F457G	0	0.276	0.272	0.279	0.276	551	17	0.83	62.5	1.25	0.7
	20'	0.266	0.272	0.264	0.267	535					
22/07/08A											
T454	0	0.298	0.288	0.285	0.290	484	117	5.86	25.7	0.514	11.4
	20'	0.223	0.223	0.214	0.220	367					
T110	0	0.286	0.327	0.304	0.306	509	-8	-0.42	21	0.42	-1.0
	20'	0.297	0.314	0.321	0.311	518					
F457G	0	0.368	0.370	0.371	0.370	739	9	0.5	23.5	0.47	1.0
	20'	0.364	0.362	0.369	0.365	730					
04/08/08B											
T454	0	0.287	0.298	0.303	0.296	493	117	5.83	25.1	0.502	11.6
	20'	0.223	0.227	0.228	0.226	377					
T110	0	0.416	0.402	0.399	0.406	676	-12	-0.58	43.2	0.864	-0.7
	20'	0.400	0.416	0.422	0.413	688					
F457G	0	0.319	0.314	0.317	0.317	528	4	0.19	30.3	0.606	0.3
	20'	0.311	0.317	0.315	0.314	524					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 29/10/07A	T454	T110	F457G
Weight of filter	0.1219	0.1154	0.1556
Dry weight of mycelium	0.0939	0.093	0.0931
Difference	0.028	0.0224	0.0625
Difference in µg	28	22.4	62.5
Date of experiment: 22/07/08A	T454	T110	F457G
Weight of filter	0.1166	0.1124	0.1142
Dry weight of mycelium	0.0909	0.0914	0.0907
Difference	0.0257	0.021	0.0235
Difference in µg	25.7	21	23.5
Date of experiment: 04/08/08B	T454	T110	F457G
Weight of filter	0.1177	0.1341	0.1205
Dry weight of mycelium	0.0926	0.0909	0.0902
Difference	0.0251	0.0432	0.0303
Difference in µg	25.1	43.2	30.3

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F457G uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F457G uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.7	1.0	0.3	0.7 ±0.34	13.0	11.4	11.6	12.0 ±0.85	0.1	-1.0	-0.7	-0.5 ±0.59
Comparison with the respective wild type (T454) result (%)											
5.0	9.0	3.0	Average: 5.0%								
Growth test^a											
F457G			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F⁴⁵⁷M Transformant 1832 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
30/10/07A											
T454	0	0.340	0.335	0.336	0.337	674	181	9.03	42	0.84	10.8
	20'	0.252	0.245	0.243	0.247	493					
T110	0	0.342	0.347	0.343	0.344	688	-9	-0.43	28	0.56	-0.8
	20'	0.345	0.347	0.353	0.348	697					
F457M	0	0.378	0.379	0.376	0.378	755	21	1.0	57.2	1.144	0.9
	20'	0.368	0.367	0.367	0.367	735					
08/02/08C											
T454	0	0.340	0.364	0.368	0.357	715	119	5.97	28.2	0.564	10.6
	20'	0.296	0.296	0.301	0.298	595					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
F457M	0	0.297	0.300	0.299	0.299	597	21	1.03	40.4	0.808	1.3
	20'	0.294	0.289	0.282	0.288	577					
02/08/08B											
T454	0	0.432	0.434	0.439	0.435	725	273	13.67	50.3	1.006	13.6
	20'	0.276	0.275	0.262	0.271	452					
T110	0	0.402	0.397	0.406	0.402	669	7	0.36	36.5	0.73	0.5
	20'	0.397	0.392	0.403	0.397	662					
F457M	0	0.375	0.378	0.372	0.375	750	25	1.23	50.5	1.01	1.2
	20'	0.354	0.365	0.369	0.363	725					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 30/10/07A	T454	T110	F457M
Weight of filter	0.1341	0.1205	0.1477
Dry weight of mycelium	0.0921	0.0925	0.0905
Difference	0.042	0.028	0.0572
Difference in μg	42	28	57.2
Date of experiment: 08/02/08C	T454	T110	F457M
Weight of filter	0.1196	0.1204	0.1332
Dry weight of mycelium	0.0914	0.0925	0.0928
Difference	0.0282	0.0279	0.0404
Difference in μg	28.2	27.9	40.4
Date of experiment: 02/08/08B	T454	T110	F457M
Weight of filter	0.1409	0.1278	0.142
Dry weight of mycelium	0.0906	0.0913	0.0915
Difference	0.0503	0.0365	0.0505
Difference in μg	50.3	36.5	50.5

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F457M uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average F457M uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
0.9	1.3	1.2	1.1 ±0.2	10.8	10.6	13.6	11.6 ±1.69	-0.8	-0.5	0.5	-0.3 ±0.66
Comparison with the respective wild type (T454) result (%)											
8.4	12.1	9.0	Average: 8.4%								
Growth test^a											
F457M			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct L⁴⁶⁰ F Transformant 1379 Colony 3 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
03/10/07A											
T454	0	0.365	0.357	0.362	0.361	723	164	8.20	39.5	0.79	10.4
	20'	0.286	0.279	0.273	0.279	559					
T110	0	0.395	0.380	0.384	0.386	773	-12	-0.60	37.6	0.752	-0.8
	20'	0.398	0.398	0.381	0.392	785					
L460F	0	0.367	0.367	0.365	0.366	733	69	3.43	38.4	0.768	4.5
	20'	0.334	0.331	0.331	0.332	664					
03/10/07B											
T454	0	0.369	0.356	0.367	0.364	728	77	3.87	17.9	0.358	10.8
	20'	0.320	0.335	0.321	0.325	651					
T110	0	0.350	0.350	0.349	0.350	699	-3	-0.17	37.7	0.754	-0.2
	20'	0.355	0.351	0.348	0.351	703					
L460F	0	0.387	0.380	0.384	0.384	767	67	3.33	34.6	0.692	4.8
	20'	0.348	0.351	0.352	0.350	701					
10/10/07A											
T454	0	0.305	0.303	0.307	0.305	610	246	12.30	61.9	1.238	9.9
	20'	0.180	0.182	0.184	0.182	364					
T110	0	0.304	0.309	0.301	0.305	609	3	0.13	44	0.88	0.2
	20'	0.308	0.301	0.301	0.303	607					
L460F	0	0.310	0.313	0.317	0.313	627	41	2.07	27.7	0.554	3.7
	20'	0.291	0.293	0.294	0.293	585					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 03/10/07A	T454	T110	L460F
Weight of filter	0.1341	0.133	0.132
Dry weight of mycelium	0.0946	0.095	0.094
Difference	0.0395	0.038	0.038
Difference in μg	39.5	37.6	38.4
Date of experiment: 03/10/07B	T454	T110	L460F
Weight of filter	0.112	0.1316	0.1287
Dry weight of mycelium	0.094	0.0939	0.0941
Difference	0.018	0.0377	0.0346
Difference in μg	17.9	37.7	34.6
Date of experiment: 10/10/07A	T454	T110	L460F
Weight of filter	0.1547	0.1361	0.1202
Dry weight of mycelium	0.0928	0.0921	0.0925
Difference	0.0619	0.044	0.0277
Difference in μg	61.9	44	27.7

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

L460F uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average L460F uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
4.5	4.8	3.7	4.3 ±0.56	10.4	10.8	9.9	10.4 ±4.3	-0.8	-0.2	0.2	-0.3 ±0.48
Comparison with the respective wild type (T454) result (%)											
43.0	45.0	38.0	Average: 43.0%								
Growth test ^a											
L460F			T454			T110					
+++			+++			-					

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Chapter 4

Construct L⁸⁴F Transformant 2607 Colony 1 with both controls: T454 (wild type) positive and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
20/08/07A											
T454	0	0.345	0.341	0.348	0.345	689	309	15.43	80.5	1.61	9.6
	20'	0.190	0.192	0.189	0.190	381					
T110	0	0.431	0.431	0.429	0.430	861	-3	-0.17	61.3	1.226	-0.1
	20'	0.428	0.433	0.435	0.432	864					
L84F	0	0.324	0.320	0.317	0.320	641	61	3.03	68.3	1.366	2.2
	20'	0.284	0.296	0.290	0.290	580					
29/10/07A											
T454	0	0.310	0.314	0.315	0.313	626	135	6.73	28	0.56	12.0
	20'	0.244	0.248	0.245	0.246	491					
T110	0	0.317	0.319	0.318	0.318	636	1	0.07	22.4	0.448	0.1
	20'	0.315	0.320	0.317	0.317	635					
L84F	0	0.341	0.344	0.349	0.345	689	51	2.6	69.4	1.388	1.8
	20'	0.322	0.317	0.318	0.319	638					
11/08/08											
T454	0	0.305	0.297	0.297	0.300	499	83	4.17	18.2	0.364	11.4
	20'	0.245	0.240	0.264	0.250	416					
T110	0	0.310	0.310	0.335	0.318	531	3	0.14	22.7	0.454	0.3
	20'	0.315	0.310	0.325	0.317	528					
L84F	0	0.299	0.298	0.302	0.300	499	22	1.08	21.3	0.426	2.5
	20'	0.289	0.282	0.289	0.287	478					

Table containing the raw data obtained on three independent net nitrate uptake assays

Calculating the Mycelial Weight			
Date of experiment: 20/08/07A	T454	T110	L84F
Weight of filter	0.1725	0.152	0.16
Dry weight of mycelium	0.092	0.091	0.092
Difference	0.0805	0.061	0.068
Difference in μg	80.5	61.3	68.3
Date of experiment: 29/10/07A	T454	T110	L84F
Weight of filter	0.122	0.1154	0.1625
Dry weight of mycelium	0.094	0.093	0.0931
Difference	0.028	0.0224	0.0694
Difference in μg	28	22.4	69.4
Date of experiment: 11/08/08	T454	T110	L84F
Weight of filter	0.109	0.1142	0.1135
Dry weight of mycelium	0.0908	0.0915	0.0922
Difference	0.0182	0.0227	0.0213
Difference in μg	18.2	22.7	21.3

L84F uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average L84F uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T454 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T454 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T110 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T110 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)
2.2	1.8	2.5	2.2 \pm 0.35	9.6	12.0	11.4	11.0 \pm 1.27	-0.1	0.1	0.3	0.1 \pm 0.22
Comparison with the respective wild type (T454) result (%)											
23.0	15.0	22.0	Average: 20.0%								
Growth test^a											
L84F			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct L⁸⁸F Transformant 2613 Colony 1 Controls: T454 (wild type) positive and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
29/10/07B											
T454	0	0.319	0.320	0.333	0.324	648	161	8.07	28	0.56	14.4
	20'	0.249	0.242	0.239	0.243	487					
T110	0	0.301	0.302	0.301	0.301	603	1	0.03	22.4	0.448	0.1
	20'	0.299	0.305	0.299	0.301	602					
L88F	0	0.333	0.336	0.338	0.336	671	21	1.07	50.2	1.004	1.1
	20'	0.325	0.324	0.326	0.325	650					
21/08/08											
T454	0	0.314	0.311	0.312	0.312	625	191	9.57	32.8	0.656	14.6
	20'	0.211	0.219	0.220	0.217	433					
T110	0	0.342	0.347	0.343	0.344	688	-9	-0.43	28	0.56	-0.8
	20'	0.345	0.347	0.353	0.348	697					
L88F	0	0.294	0.291	0.288	0.291	582	51	2.53	71.8	1.436	1.8
	20'	0.265	0.263	0.269	0.266	531					
11/08/08											
T454	0	0.305	0.297	0.297	0.300	499	94	4.72	18.2	0.364	13.0
	20'	0.245	0.240	0.244	0.243	405					
T110	0	0.310	0.310	0.335	0.318	531	3	0.14	22.7	0.454	0.3
	20'	0.315	0.310	0.325	0.317	528					
L88F	0	0.362	0.362	0.367	0.364	727	22	1.10	28	0.56	2.0
	20'	0.350	0.356	0.352	0.353	705					

Table containing the raw data obtained on three independent net nitrate uptake assays

Calculating the Mycelial Weight			
Date of experiment: 29/10/07B	T454	T110	L88F
Weight of filter	0.1219	0.115	0.1433
Dry weight of mycelium	0.0939	0.093	0.0931
Difference	0.028	0.022	0.0502
Difference in μg	28	22.4	50.2
Date of experiment: 21/08/08	T454	T110	L88F
Weight of filter	0.1254	0.1205	0.1636
Dry weight of mycelium	0.0926	0.0925	0.0918
Difference	0.0328	0.028	0.0718
Difference in μg	32.8	28	71.8
Date of experiment: 11/08/08	T454	T110	L88F
Weight of filter	0.109	0.1142	0.1183
Dry weight of mycelium	0.0908	0.0915	0.0903
Difference	0.0182	0.0227	0.028
Difference in μg	18.2	22.7	28

L88F uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average L88F uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T454 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T454 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T110 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T110 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)
1.1	1.8	2.0	1.6 \pm 0.47	14.4	14.6	13.0	14.0 \pm 0.88	0.1	-0.8	0.3	-0.1 \pm 0.57
Comparison with the respective wild type (T454) result (%)											
7.0	12.0	15.0	Average: 12.0%								
Growth test^a											
L88F			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct W¹⁹⁷Y Transformant 3766 Colony 1 Controls: T454 (wild type) positive and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
31/07/08C											
T454	0	0.415	0.410	0.412	0.412	687	147	7.36	34.7	0.694	10.6
	20'	0.324	0.327	0.321	0.324	540					
T110	0	0.425	0.425	0.418	0.423	704	-6	-0.28	50.3	1.006	-0.3
	20'	0.429	0.423	0.426	0.426	710					
W197Y	0	0.484	0.474	0.476	0.478	797	6	0.28	49.1	0.982	0.3
	20'	0.472	0.478	0.474	0.475	791					
31/07/08C											
T454	0	0.415	0.410	0.412	0.412	687	147	7.36	34.7	0.694	10.6
	20'	0.324	0.327	0.321	0.324	540					
T110	0	0.425	0.425	0.418	0.423	704	-6	-0.28	50.3	1.006	-0.3
	20'	0.429	0.423	0.426	0.426	710					
W197Y	0	0.450	0.451	0.442	0.448	746	2	0.11	34.6	0.692	0.2
	20'	0.449	0.447	0.443	0.446	744					
01/08/08C											
T454	0	0.420	0.424	0.429	0.424	707	261	13.06	50.5	1.01	12.9
	20'	0.276	0.265	0.262	0.268	446					
T110	0	0.407	0.377	0.406	0.397	661	-2	-0.08	37.2	0.744	-0.1
	20'	0.401	0.392	0.400	0.398	663					
W197Y	0	0.379	0.381	0.388	0.383	638	11	0.56	42.3	0.846	0.7
	20'	0.373	0.376	0.379	0.376	627					

Table containing the raw data obtained on three independent net nitrate uptake assays

Calculating the Mycelial Weight			
Date of experiment: 31/07/08C	T454	T110	W197Y
Weight of filter	0.1255	0.141	0.1398
Dry weight of mycelium	0.0908	0.091	0.0907
Difference	0.0347	0.05	0.0491
Difference in μg	34.7	50.3	49.1
Date of experiment: 31/07/08C	T454	T110	W197Y
Weight of filter	0.1255	0.141	0.1262
Dry weight of mycelium	0.0908	0.091	0.0916
Difference	0.0347	0.05	0.0346
Difference in μg	34.7	50.3	34.6
Date of experiment: 01/08/08C	T454	T110	W197Y
Weight of filter	0.1409	0.1278	0.1325
Dry weight of mycelium	0.0904	0.0906	0.0902
Difference	0.0505	0.0372	0.0423
Difference in μg	50.5	37.2	42.3

W197Y uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average W197Y uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T454 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T454 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T110 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T110 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)
0.3	0.2	0.7	0.4 \pm 0.26	10.6	10.6	12.9	11.8 \pm 1.6	-0.3	-0.3	-0.1	-0.2 \pm 0.12
Comparison with the respective wild type (T454) result (%)											
2.7	1.5	5.1	Average: 3.1%								
Growth test^a											
W197Y			T454				T110				
+-			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct C³⁵⁷F Transformant 3146 Colony 1 Controls: T454 (wild type) positive and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
20/08/07											
T454	0	0.441	0.434	0.424	0.433	866	112	5.6	23.2	0.464	12.1
	20'	0.378	0.376	0.377	0.377	754					
T110	0	0.365	0.366	0.365	0.365	731	-3	-0.2	25.2	0.504	-0.3
	20'	0.369	0.362	0.370	0.367	734					
C357F	0	0.368	0.366	0.356	0.363	727	7	0.4	26.4	0.528	0.7
	20'	0.359	0.361	0.359	0.360	719					
11/02/08											
T454	0	0.360	0.354	0.368	0.361	721	141	7.0	25.7	0.514	13.7
	20'	0.294	0.289	0.288	0.290	581					
T110	0	0.389	0.372	0.381	0.381	761	1	0.0	32.7	0.654	0.1
	20'	0.385	0.380	0.376	0.380	761					
C357F	0	0.477	0.479	0.481	0.479	958	12	0.6	32.3	0.646	0.9
	20'	0.473	0.475	0.471	0.473	946					
13/02/08											
T454	0	0.320	0.325	0.321	0.322	537	112	5.6	24.8	0.496	11.3
	20'	0.252	0.255	0.258	0.255	425					
T110	0	0.313	0.317	0.314	0.315	524	0	0.0	11.2	0.224	0.0
	20'	0.314	0.321	0.309	0.315	524					
C357F	0	0.287	0.289	0.305	0.294	587	7	0.3	52.6	1.052	0.3
	20'	0.298	0.281	0.292	0.290	581					

Calculating the Mycelial Weight			
Date of experiment: 20/08/07	T454	T110	C357F
Weight of filter	0.1126	0.1157	0.1168
Dry weight of mycelium	0.0894	0.0905	0.0904
Difference	0.0232	0.0252	0.0264
Difference in μg	23.2	25.2	26.4
Date of experiment: 11/02/08	T454	T110	C357F
Weight of filter	0.1156	0.123	0.123
Dry weight of mycelium	0.0899	0.0903	0.0907
Difference	0.0257	0.0327	0.0323
Difference in μg	25.7	32.7	32.3
Date of experiment: 13/02/08	T454	T110	C357F
Weight of filter	0.1383	0.1935	0.1451
Dry weight of mycelium	0.1135	0.1823	0.0925
Difference	0.0248	0.0112	0.0526
Difference in μg	24.8	11.2	52.6

C357F uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average C357F uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T454 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T454 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T110 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T110 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)
0.7	0.9	0.3	0.6 ± 0.31	12.1	13.7	11.3	12.3 ± 1.24	-0.3	0.1	0.0	-0.1 ± 0.21
Comparison with the respective wild type (T454) result (%)											
5.8	6.6	2.7	Average: 5.0%								
Growth test^a											
C357F			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO_3 and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Chapter 5

Construct F³⁶C Transformant 51 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
04/06/07A											
T454	0	0.358	0.342	0.352	0.351	701	198	9.90	30.8	0.616	16.1
	20'	0.255	0.249	0.251	0.252	503					
T110	0	0.301	0.347	0.336	0.328	656	-14	-0.70	33.7	0.674	-1.0
	20'	0.340	0.326	0.339	0.335	670					
F36C	0	0.321	0.326	0.326	0.324	649	18	0.90	57.6	1.152	0.8
	20'	0.311	0.317	0.318	0.315	631					
04/06/07B											
T454	0	0.358	0.342	0.352	0.351	701	198	9.90	30.8	0.616	16.1
	20'	0.255	0.249	0.251	0.252	503					
T110	0	0.301	0.347	0.336	0.328	656	-14	-0.70	33.7	0.674	-1.0
	20'	0.340	0.326	0.339	0.335	670					
F36C	0	0.376	0.379	0.376	0.377	754	8	0.40	34.3	0.686	0.6
	20'	0.371	0.376	0.372	0.373	746					
15/10/07											
T454	0	0.419	0.428	0.426	0.424	849	266	13.30	48.2	0.964	13.8
	20'	0.289	0.289	0.296	0.291	583					
T110	0	0.347	0.347	0.359	0.351	702	-13	-0.63	53.6	1.072	-0.6
	20'	0.362	0.352	0.358	0.357	715					
F36C	0	0.373	0.375	0.375	0.374	749	4	0.2	36.4	0.728	0.3
	20'	0.371	0.372	0.374	0.372	745					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 20/08/07	T454	T110	F36C
Weight of filter	0.1245	0.124	0.1504
Dry weight of mycelium	0.0937	0.09	0.0928
Difference	0.0308	0.034	0.0576
Difference in μg	30.8	33.7	57.6
Date of experiment: 11/02/08	T454	T110	F36C
Weight of filter	0.1245	0.124	0.1278
Dry weight of mycelium	0.0937	0.09	0.0935
Difference	0.0308	0.034	0.0343
Difference in μg	30.8	33.7	34.3
Date of experiment: 13/02/08	T454	T110	F36C
Weight of filter	0.2563	0.2331	0.1285
Dry weight of mycelium	0.2081	0.1795	0.0921
Difference	0.0482	0.0536	0.0364
Difference in μg	48.2	53.6	36.4

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F36C uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F36C uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.8	0.6	0.3	0.5 ±0.3	16.1	16.1	13.8	15.3 ±1.3	13.8	-1.0	-0.6	-0.9 ±0.3
Comparison with the respective wild type (T454) result (%)											
4.9	3.6	2.0	Average: 3.5%								
Growth test^a											
F36C			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F³⁶ Y Transformant 17 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
30/10/07B											
T454	0	0.341	0.338	0.337	0.339	677	153	7.67	42	0.84	9.1
	20'	0.262	0.261	0.263	0.262	524					
T110	0	0.349	0.352	0.354	0.352	703	-3	-0.13	28	0.56	-0.2
	20'	0.361	0.347	0.351	0.353	706					
F36Y	0	0.338	0.335	0.335	0.336	672	43	2.13	73	1.46	1.5
	20'	0.314	0.314	0.316	0.315	629					
31/01/08B											
T454	0	0.358	0.353	0.360	0.357	714	121	6.07	37.5	0.75	8.1
	20'	0.297	0.297	0.295	0.296	593					
T110	0	0.378	0.360	0.362	0.367	733	-4	-0.20	18.4	0.368	-0.5
	20'	0.367	0.369	0.370	0.369	737					
F36Y	0	0.349	0.343	0.348	0.347	693	9	0.43	18	0.36	1.2
	20'	0.339	0.349	0.339	0.342	685					
14/08/08											
T454	0	0.331	0.325	0.331	0.329	548	90	4.50	19.4	0.388	11.6
	20'	0.282	0.275	0.268	0.275	458					
T110	0	0.333	0.337	0.324	0.331	552	-6	-0.28	11.2	0.224	-1.2
	20'	0.334	0.331	0.339	0.335	558					
F36Y	0	0.345	0.347	0.347	0.346	693	26	1.30	37.6	0.752	1.7
	20'	0.330	0.335	0.335	0.333	667					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 30/10/07B	T454	T110	F36Y
Weight of filter	0.1341	0.121	0.165
Dry weight of mycelium	0.0921	0.093	0.092
Difference	0.042	0.028	0.073
Difference in μg	42	28	73
Date of experiment: 31/01/08B	T454	T110	F36Y
Weight of filter	0.1273	0.1101	0.1088
Dry weight of mycelium	0.0898	0.0917	0.0908
Difference	0.0375	0.0184	0.018
Difference in μg	37.5	18.4	18
Date of experiment: 14/08/08	T454	T110	F36Y
Weight of filter	0.1393	0.1935	0.1292
Dry weight of mycelium	0.1199	0.1823	0.0916
Difference	0.0194	0.0112	0.0376
Difference in μg	19.4	11.2	37.6

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F36Y uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average F36Y uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
1.5	1.2	1.7	1.5 ±0.26	9.1	8.1	11.6	9.6 ±1.8	-0.2	-0.5	-1.2	-0.7 ±0.51
Comparison with the respective wild type (T454) result (%)											
16.0	14.9	14.9	Average: 16.0%								
Growth test^a											
F36Y			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F³⁶ L Transformant 116 Colony 3 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
15/10/07A											
T454	0	0.340	0.348	0.380	0.356	712	206	10.30	30.6	0.612	16.8
	20'	0.267	0.255	0.237	0.253	506					
T110	0	0.306	0.323	0.313	0.314	628	-17	-0.87	27.5	0.55	-1.6
	20'	0.318	0.323	0.327	0.323	645					
F36L	0	0.364	0.369	0.361	0.365	729	5	0.27	23.7	0.474	0.6
	20'	0.362	0.361	0.363	0.362	724					
15/10/07B											
T454	0	0.340	0.342	0.341	0.341	682	211	10.53	40.6	0.812	13.0
	20'	0.237	0.235	0.235	0.236	471					
T110	0	0.366	0.363	0.363	0.364	728	-13	-0.63	23.2	0.464	-1.4
	20'	0.368	0.373	0.370	0.370	741					
F36L	0	0.405	0.408	0.400	0.404	809	9	0.47	30.3	0.606	0.8
	20'	0.401	0.397	0.401	0.400	799					
15/10/07C											
T454	0	0.351	0.348	0.357	0.352	704	169	8.47	25.6	0.512	16.5
	20'	0.267	0.275	0.260	0.267	535					
T110	0	0.306	0.303	0.315	0.308	616	-3	-0.13	28.9	0.578	-0.2
	20'	0.308	0.313	0.307	0.309	619					
F36L	0	0.386	0.384	0.384	0.385	769	1	0.1	22.6	0.452	0.1
	20'	0.385	0.380	0.387	0.384	768					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 15/10/07A	T454	T110	F36L
Weight of filter	0.123	0.121	0.1153
Dry weight of mycelium	0.0924	0.093	0.0916
Difference	0.0306	0.028	0.0237
Difference in µg	30.6	27.5	23.7
Date of experiment: 15/10/07B	T454	T110	F36L
Weight of filter	0.133	0.1206	0.1243
Dry weight of mycelium	0.0924	0.0974	0.094
Difference	0.0406	0.0232	0.0303
Difference in µg	40.6	23.2	30.3
Date of experiment: 15/10/07C	T454	T110	F36L
Weight of filter	0.123	0.1276	0.1127
Dry weight of mycelium	0.0974	0.0987	0.0901
Difference	0.0256	0.0289	0.0226
Difference in µg	25.6	28.9	22.6

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F36L uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F36L uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.6	0.8	0.1	0.5 ±0.32	16.8	13.0	16.5	15.4 ±2.15	-1.6	-1.4	-0.2	-1.1 ±0.72
Comparison with the respective wild type (T454) result (%)											
3.3	5.9	0.9	Average: 3.3%								
Growth test^a											
F36L			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct W⁴⁰ F Transformant 48 Colony 3 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
29/10/07B											
T454	0	0.364	0.364	0.368	0.365	731	135	6.77	28.2	0.564	12.0
	20'	0.296	0.296	0.301	0.298	595					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
W40F	0	0.358	0.360	0.357	0.358	717	39	1.93	29.5	0.59	3.3
	20'	0.342	0.338	0.337	0.339	678					
08/02/08A											
T454	0	0.319	0.320	0.313	0.317	635	148	7.40	28	0.56	13.2
	20'	0.249	0.242	0.239	0.243	487					
T110	0	0.317	0.319	0.318	0.318	636	1	0.07	22.4	0.448	0.1
	20'	0.315	0.320	0.317	0.317	635					
W40F	0	0.349	0.349	0.364	0.354	708	68	3.4	46.9	0.938	3.6
	20'	0.319	0.321	0.320	0.320	640					
14/08/08											
T454	0	0.381	0.385	0.371	0.379	632	40	2.00	10	0.2	10.0
	20'	0.352	0.355	0.358	0.355	592					
T110	0	0.313	0.307	0.314	0.311	519	0	0.00	11.2	0.224	0.0
	20'	0.314	0.311	0.309	0.311	519					
W40F	0	0.367	0.359	0.362	0.363	725	34	1.7	23.1	0.462	3.7
	20'	0.345	0.347	0.345	0.346	691					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 29/10/07B	T454	T110	W40F
Weight of filter	0.1196	0.12	0.1225
Dry weight of mycelium	0.0914	0.093	0.093
Difference	0.0282	0.028	0.0295
Difference in μg	28.2	27.9	29.5
Date of experiment: 08/02/08A	T454	T110	W40F
Weight of filter	0.1219	0.1154	0.1406
Dry weight of mycelium	0.0939	0.093	0.0937
Difference	0.028	0.0224	0.0469
Difference in μg	28	22.4	46.9
Date of experiment: 14/08/08	T454	T110	W40F
Weight of filter	0.1383	0.1935	0.1157
Dry weight of mycelium	0.1283	0.1823	0.0926
Difference	0.01	0.0112	0.0231
Difference in μg	10	11.2	23.1

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

W40F uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average W40F uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
3.3	3.6	3.7	3.5 ±0.22	12.0	13.2	10.0	11.7 ±1.62	-0.5	0.1	0.0	-0.1 ±0.33
Comparison with the respective wild type (T454) result (%)											
27.3	27.4	36.8	Average: 30.5%								
Growth test^a											
W40F			T454				T110				
++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct W⁴⁰ L Transformant 3068 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
11/02/08B											
T454	0	0.381	0.391	0.385	0.386	771	129	6.47	32.3	0.646	10.0
	20'	0.323	0.319	0.321	0.321	642					
T110	0	0.365	0.366	0.365	0.365	731	-3	-0.17	25.2	0.504	-0.3
	20'	0.369	0.362	0.370	0.367	734					
W40L	0	0.362	0.362	0.365	0.363	726	9	0.5	36.3	0.726	0.6
	20'	0.356	0.359	0.360	0.358	717					
13/02/08A											
T454	0	0.344	0.345	0.346	0.345	690	99	4.93	25.7	0.514	9.6
	20'	0.294	0.299	0.294	0.296	591					
T110	0	0.389	0.399	0.381	0.390	779	-16	-0.80	32.7	0.654	-1.2
	20'	0.395	0.402	0.396	0.398	795					
W40L	0	0.365	0.367	0.361	0.364	729	18	0.90	39.8	0.796	1.1
	20'	0.357	0.357	0.352	0.355	711					
20/02/08B											
T454	0	0.331	0.335	0.331	0.332	554	129	6.44	26.3	0.526	12.3
	20'	0.252	0.255	0.258	0.255	425					
T110	0	0.313	0.317	0.314	0.315	524	0	0.00	11.2	0.224	0.0
	20'	0.314	0.321	0.309	0.315	524					
W40L	0	0.351	0.354	0.362	0.356	711	5	0.2	23.4	0.468	0.5
	20'	0.352	0.356	0.352	0.353	707					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 11/02/08B	T454	T110	W40L
Weight of filter	0.123	0.1157	0.1264
Dry weight of mycelium	0.0907	0.0905	0.0901
Difference	0.0323	0.0252	0.0363
Difference in μg	32.3	25.2	36.3
Date of experiment: 13/02/08A	T454	T110	W40L
Weight of filter	0.1156	0.123	0.1306
Dry weight of mycelium	0.0899	0.0903	0.0908
Difference	0.0257	0.0327	0.0398
Difference in μg	25.7	32.7	39.8
Date of experiment: 20/02/08B	T454	T110	W40L
Weight of filter	0.1178	0.1935	0.1134
Dry weight of mycelium	0.0915	0.1823	0.09
Difference	0.0263	0.0112	0.0234
Difference in μg	26.3	11.2	23.4

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

W40L uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average W40L uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.6	1.1	0.5	0.8 ±0.3	10.0	9.6	12.3	10.6 ±1.4	-0.3	-1.2	0.0	-0.5 ±0.63
Comparison with the respective wild type (T454) result (%)											
6.4	11.8	4.1	Average: 7.4%								
Growth test^a											
W40L			T454				T110				
+			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct W⁴⁰Y Transformant 2705 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
03/10/07B											
T454	0	0.364	0.366	0.367	0.366	731	81	4.03	17.9	0.358	11.3
	20'	0.320	0.335	0.321	0.325	651					
T110	0	0.350	0.350	0.349	0.350	699	-3	-0.13	37.7	0.754	-0.2
	20'	0.358	0.347	0.348	0.351	702					
W40Y	0	0.228	0.228	0.224	0.227	453	49	2.47	24.6	0.492	5.0
	20'	0.205	0.201	0.200	0.202	404					
04/10/07A											
T454	0	0.232	0.240	0.235	0.236	471	130	6.50	23.2	0.464	14.0
	20'	0.173	0.171	0.168	0.171	341					
T110	0	0.220	0.224	0.212	0.219	437	-3	-0.13	33	0.66	-0.2
	20'	0.225	0.218	0.217	0.220	440					
W40Y	0	0.238	0.234	0.233	0.235	470	35	1.73	20.6	0.412	4.2
	20'	0.220	0.218	0.215	0.218	435					
30/10/07A											
T454	0	0.349	0.335	0.336	0.340	680	187	9.33	42	0.84	11.1
	20'	0.252	0.245	0.243	0.247	493					
T110	0	0.378	0.360	0.362	0.367	733	-4	-0.20	18.4	0.368	-0.5
	20'	0.367	0.369	0.370	0.369	737					
W40Y	0	0.376	0.371	0.376	0.374	749	37	1.9	18.3	0.366	5.1
	20'	0.354	0.354	0.359	0.356	711					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 03/10/07B	T454	T110	W40Y
Weight of filter	0.1116	0.132	0.119
Dry weight of mycelium	0.0937	0.094	0.0944
Difference	0.0179	0.038	0.0246
Difference in μg	17.9	37.7	24.6
Date of experiment: 04/10/07A	T454	T110	W40Y
Weight of filter	0.1181	0.1281	0.114
Dry weight of mycelium	0.0949	0.0951	0.0934
Difference	0.0232	0.033	0.0206
Difference in μg	23.2	33	20.6
Date of experiment: 30/10/07A	T454	T110	W40Y
Weight of filter	0.1341	0.1101	0.1086
Dry weight of mycelium	0.0921	0.0917	0.0903
Difference	0.042	0.0184	0.0183
Difference in μg	42	18.4	18.3

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

W40Y uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average W40Y uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
5.0	4.2	5.1	4.8 ±0.49	11.3	14.0	11.1	12.1 ±0.11	-0.2	-0.2	-0.5	-0.3 ±0.2
Comparison with the respective wild type (T454) result (%)											
44.5	30.0	46.0	Average: 40.1%								
Growth test^a											
W40Y			T454				T110				
++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F⁴³ W Transformant 370 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
08/02/08A											
T454	0	0.364	0.364	0.368	0.365	731	138	6.90	28.2	0.564	12.2
	20'	0.296	0.296	0.297	0.296	593					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
F43W	0	0.365	0.367	0.368	0.367	733	49	2.47	16.9	0.338	7.3
	20'	0.342	0.343	0.341	0.342	684					
08/02/08B											
T454	0	0.349	0.364	0.368	0.360	721	123	6.17	28.2	0.564	10.9
	20'	0.299	0.296	0.301	0.299	597					
T110	0	0.383	0.385	0.379	0.382	765	-14	-0.70	27.9	0.558	-1.3
	20'	0.384	0.395	0.389	0.389	779					
F43W	0	0.396	0.397	0.398	0.397	794	58	2.9	21	0.42	6.9
	20'	0.367	0.369	0.368	0.368	736					
08/02/08D											
T454	0	0.394	0.391	0.388	0.391	782	129	6.43	28.2	0.564	11.4
	20'	0.325	0.326	0.329	0.327	653					
T110	0	0.321	0.322	0.318	0.320	641	-5	-0.27	27.9	0.558	-0.5
	20'	0.315	0.330	0.324	0.323	646					
F43W	0	0.401	0.397	0.403	0.400	801	73	3.63	26.9	0.538	6.8
	20'	0.364	0.362	0.366	0.364	728					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 08/02/08A	T454	T110	F43W
Weight of filter	0.1196	0.1204	0.1092
Dry weight of mycelium	0.0914	0.0925	0.0923
Difference	0.0282	0.0279	0.0169
Difference in μg	28.2	27.9	16.9
Date of experiment: 08/02/08B	T454	T110	F43W
Weight of filter	0.1196	0.1204	0.1129
Dry weight of mycelium	0.0914	0.0925	0.0919
Difference	0.0282	0.0279	0.021
Difference in μg	28.2	27.9	21
Date of experiment: 08/02/08D	T454	T110	F43W
Weight of filter	0.1196	0.1204	0.1196
Dry weight of mycelium	0.0914	0.0925	0.0927
Difference	0.0282	0.0279	0.0269
Difference in μg	28.2	27.9	26.9

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F43W uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F43W uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
7.3	6.9	6.8	7.0 ±0.28	12.2	10.9	11.4	11.5 ±0.66	-0.5	-1.3	-0.5	-0.7 ±0.45
Comparison with the respective wild type (T454) result (%)											
60.0	63.0	59.0	Average: 60.0%								
Growth test^a											
F43W			T454				T110				
+-			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F⁴³ L Transformant 2126 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
10/10/07B											
T454	0	0.301	0.303	0.307	0.304	607	243	12.13	62.4	1.248	9.7
	20'	0.182	0.183	0.182	0.182	365					
T110	0	0.301	0.302	0.300	0.301	602	-2	-0.10	44.6	0.892	-0.1
	20'	0.299	0.303	0.304	0.302	604					
F43L	0	0.310	0.312	0.314	0.312	624	53	2.63	34.3	0.686	3.8
	20'	0.287	0.287	0.283	0.286	571					
30/10/07B											
T454	0	0.340	0.335	0.336	0.337	674	181	9.03	42	0.84	10.8
	20'	0.252	0.245	0.243	0.247	493					
T110	0	0.342	0.347	0.343	0.344	688	-9	-0.43	28	0.56	-0.8
	20'	0.345	0.347	0.353	0.348	697					
F43L	0	0.327	0.342	0.336	0.335	670	67	3.33	51.5	1.03	3.2
	20'	0.300	0.311	0.294	0.302	603					
21/08/08											
T454	0	0.314	0.305	0.320	0.313	626	212	10.60	32.8	0.656	16.2
	20'	0.211	0.205	0.205	0.207	414					
T110	0	0.289	0.285	0.284	0.286	572	-13	-0.67	27.2	0.544	-1.2
	20'	0.289	0.296	0.293	0.293	585					
F43L	0	0.387	0.389	0.388	0.388	776	41	2.07	25.2	0.504	4.1
	20'	0.369	0.368	0.365	0.367	735					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 10/10/07B	T454	T110	F43L
Weight of filter	0.1545	0.137	0.1278
Dry weight of mycelium	0.0921	0.0924	0.0935
Difference	0.0624	0.0446	0.0343
Difference in µg	62.4	44.6	34.3
Date of experiment: 30/10/07B	T454	T110	F43L
Weight of filter	0.1341	0.1205	0.1439
Dry weight of mycelium	0.0921	0.0925	0.0924
Difference	0.042	0.028	0.0515
Difference in µg	42	28	51.5
Date of experiment: 21/08/08	T454	T110	F43L
Weight of filter	0.1254	0.1182	0.1179
Dry weight of mycelium	0.0926	0.091	0.0927
Difference	0.0328	0.0272	0.0252
Difference in µg	32.8	27.2	25.2

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F43L uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F43L uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
3.8	3.2	4.1	3.7 ±0.44	9.7	10.8	16.2	12.2 ±3.46	-0.1	-0.8	-1.2	-0.7 ±0.56
Comparison with the respective wild type (T454) result (%)											
39.45	30.1	25.4	Average: 39.5%								
Growth test^a											
F43L			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F⁴⁷ L Transformant 3866 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
04/06/07A											
T454	0	0.358	0.342	0.352	0.351	701	198	9.90	30.8	0.616	16.1
	20'	0.255	0.249	0.251	0.252	503					
T110	0	0.301	0.347	0.336	0.328	656	-14	-0.70	33.7	0.674	-1.0
	20'	0.340	0.326	0.339	0.335	670					
F47K	0	0.321	0.326	0.326	0.324	649	18	0.90	57.6	1.152	0.8
	20'	0.311	0.317	0.318	0.315	631					
04/06/07B											
T454	0	0.358	0.342	0.352	0.351	701	198	9.90	30.8	0.616	16.1
	20'	0.255	0.249	0.251	0.252	503					
T110	0	0.301	0.347	0.336	0.328	656	-14	-0.70	33.7	0.674	-1.0
	20'	0.340	0.326	0.339	0.335	670					
F47K	0	0.376	0.379	0.376	0.377	754	8	0.40	34.3	0.686	0.6
	20'	0.371	0.376	0.372	0.373	746					
15/10/07											
T454	0	0.419	0.428	0.426	0.424	849	266	13.30	48.2	0.964	13.8
	20'	0.289	0.289	0.296	0.291	583					
T110	0	0.347	0.347	0.359	0.351	702	-13	-0.63	53.6	1.072	-0.6
	20'	0.362	0.352	0.358	0.357	715					
F47K	0	0.373	0.375	0.375	0.374	749	4	0.2	36.4	0.728	0.3
	20'	0.371	0.372	0.374	0.372	745					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 20/08/07	T454	T110	F47K
Weight of filter	0.1245	0.124	0.1504
Dry weight of mycelium	0.0937	0.09	0.0928
Difference	0.0308	0.034	0.0576
Difference in μg	30.8	33.7	57.6
Date of experiment: 11/02/08	T454	T110	F47K
Weight of filter	0.1245	0.124	0.1278
Dry weight of mycelium	0.0937	0.09	0.0935
Difference	0.0308	0.034	0.0343
Difference in μg	30.8	33.7	34.3
Date of experiment: 13/02/08	T454	T110	F47K
Weight of filter	0.2563	0.2331	0.1285
Dry weight of mycelium	0.2081	0.1795	0.0921
Difference	0.0482	0.0536	0.0364
Difference in μg	48.2	53.6	36.4

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F47K uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F47K uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.8	0.6	0.3	0.5 ±0.3	16.1	16.1	13.8	15.3 ±1.3	-1.0	-1.0	-0.6	-0.9 ±0.3
Comparison with the respective wild type (T454) result (%)											
4.9	3.6	2.0	Average: 3.5%								
Growth test^a											
F47K			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F⁴⁷ W Transformant 447 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
31/01/08A											
T454	0	0.388	0.381	0.386	0.385	770	177	8.87	37.5	0.75	11.8
	20'	0.297	0.297	0.295	0.296	593					
T110	0	0.342	0.350	0.342	0.345	689	-2	-0.10	18.4	0.368	-0.3
	20'	0.341	0.353	0.343	0.346	691					
F47W	0	0.401	0.405	0.399	0.402	803	15	0.77	29.8	0.596	1.3
	20'	0.392	0.398	0.392	0.394	788					
08/02/08C											
T454	0	0.340	0.344	0.348	0.344	688	129	6.43	28.2	0.564	11.4
	20'	0.276	0.279	0.284	0.280	559					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
F47W	0	0.361	0.358	0.362	0.360	721	6	0.30	19.3	0.386	0.8
	20'	0.364	0.351	0.357	0.357	715					
21/08/08											
T454	0	0.334	0.341	0.340	0.338	677	173	8.67	32.8	0.656	13.2
	20'	0.249	0.255	0.251	0.252	503					
T110	0	0.288	0.275	0.274	0.279	558	-5	-0.23	27.2	0.544	-0.4
	20'	0.279	0.284	0.281	0.281	563					
F47W	0	0.305	0.318	0.310	0.311	622	16	0.80	26.1	0.522	1.5
	20'	0.305	0.318	0.286	0.303	606					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 31/01/08A	T454	T110	F47W
Weight of filter	0.1273	0.11	0.1203
Dry weight of mycelium	0.0898	0.092	0.0905
Difference	0.0375	0.018	0.0298
Difference in μg	37.5	18.4	29.8
Date of experiment: 08/02/08C	T454	T110	F47W
Weight of filter	0.1196	0.1204	0.1085
Dry weight of mycelium	0.0914	0.0925	0.0892
Difference	0.0282	0.0279	0.0193
Difference in μg	28.2	27.9	19.3
Date of experiment: 21/08/08	T454	T110	F47W
Weight of filter	0.1254	0.1182	0.1178
Dry weight of mycelium	0.0926	0.091	0.0917
Difference	0.0328	0.0272	0.0261
Difference in μg	32.8	27.2	26.1

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F47W uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F47W uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
1.3	0.8	1.5	1.2 ±0.39	11.8	11.4	13.2	12.1 ±0.95	-0.3	-0.5	-0.4	-0.4 ±0.11
Comparison with the respective wild type (T454) result (%)											
11.0	7.0	12.0	Average: 11.0%								
Growth test^a											
F47W			T454				T110				
+			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F⁴⁷ Y Transformant 2635 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
19/09/07B											
T454	0	0.309	0.301	0.300	0.303	607	193	9.63	49.3	0.986	9.8
	20'	0.210	0.205	0.206	0.207	414					
T110	0	0.351	0.357	0.346	0.351	703	-14	-0.70	62.4	1.248	-0.6
	20'	0.358	0.362	0.355	0.358	717					
F47Y	0	0.312	0.306	0.311	0.310	619	26	1.30	60	1.2	1.1
	20'	0.298	0.290	0.302	0.297	593					
10/10/07A											
T454	0	0.305	0.303	0.307	0.305	610	246	12.30	61.9	1.238	9.9
	20'	0.180	0.182	0.184	0.182	364					
T110	0	0.304	0.309	0.301	0.305	609	11	0.53	44	0.88	0.6
	20'	0.299	0.301	0.298	0.299	599					
F47Y	0	0.311	0.313	0.307	0.310	621	7	0.33	47.3	0.946	0.4
	20'	0.302	0.309	0.310	0.307	614					
31/01/08B											
T454	0	0.358	0.353	0.360	0.357	714	121	6.07	37.5	0.75	8.1
	20'	0.297	0.297	0.295	0.296	593					
T110	0	0.378	0.360	0.362	0.367	733	-4	-0.20	18.4	0.368	-0.5
	20'	0.367	0.369	0.370	0.369	737					
F47Y	0	0.387	0.390	0.402	0.393	786	12	0.6	20.9	0.418	1.4
	20'	0.386	0.382	0.393	0.387	774					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 19/09/07B	T454	T110	F47Y
Weight of filter	0.1392	0.155	0.149
Dry weight of mycelium	0.0899	0.093	0.089
Difference	0.0493	0.062	0.06
Difference in µg	49.3	62.4	60
Date of experiment: 10/10/07A	T454	T110	F47Y
Weight of filter	0.1547	0.1361	0.1405
Dry weight of mycelium	0.0928	0.0921	0.0932
Difference	0.0619	0.044	0.0473
Difference in µg	61.9	44	47.3
Date of experiment: 31/01/08B	T454	T110	F47Y
Weight of filter	0.1273	0.1101	0.1105
Dry weight of mycelium	0.0898	0.0917	0.0896
Difference	0.0375	0.0184	0.0209
Difference in µg	37.5	18.4	20.9

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F47Y uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F47Y uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
1.1	0.4	1.4	1.0±0.55	9.8	9.9	8.1	9.3 ±1.02	-0.6	0.6	-0.5	-0.2±0.67
Comparison with the respective wild type (T454) result (%)											
11.1	3.5	17.7	Average: 11.1%								
Growth test^a											
F47Y			T454				T110				
+			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct Y⁵¹ L Transformant 3016 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
11/02/08A											
T454	0	0.371	0.388	0.385	0.381	763	146	7.30	32.3	0.646	11.3
	20'	0.311	0.310	0.304	0.308	617					
T110	0	0.375	0.356	0.365	0.365	731	-3	-0.17	25.2	0.504	-0.3
	20'	0.369	0.362	0.370	0.367	734					
Y51L	0	0.361	0.368	0.363	0.364	728	23	1.2	22.7	0.454	2.6
	20'	0.347	0.349	0.361	0.352	705					
20/02/08B											
T454	0	0.374	0.371	0.375	0.373	747	137	6.87	25.7	0.514	13.4
	20'	0.310	0.305	0.299	0.305	609					
T110	0	0.369	0.372	0.371	0.371	741	-7	-0.37	32.7	0.654	-0.6
	20'	0.365	0.382	0.376	0.374	749					
Y51L	0	0.356	0.360	0.361	0.359	718	29	1.4	28.9	0.578	2.5
	20'	0.346	0.342	0.346	0.345	689					
14/08/08											
T454	0	0.310	0.325	0.331	0.322	537	112	5.58	25.8	0.516	10.8
	20'	0.252	0.255	0.258	0.255	425					
T110	0	0.313	0.317	0.314	0.315	524	0	0.00	11.2	0.224	0.0
	20'	0.314	0.321	0.309	0.315	524					
Y51L	0	0.381	0.390	0.381	0.384	640	19	0.97	16.2	0.324	3.0
	20'	0.374	0.365	0.378	0.372	621					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 11/02/08A	T454	T110	Y51L
Weight of filter	0.123	0.1157	0.113
Dry weight of mycelium	0.0907	0.0905	0.0903
Difference	0.0323	0.0252	0.0227
Difference in µg	32.3	25.2	22.7
Date of experiment: 20/02/08B	T454	T110	Y51L
Weight of filter	0.1156	0.123	0.1193
Dry weight of mycelium	0.0899	0.0903	0.0904
Difference	0.0257	0.0327	0.0289
Difference in µg	25.7	32.7	28.9
Date of experiment: 14/08/08	T454	T110	Y51L
Weight of filter	0.1224	0.1935	0.1967
Dry weight of mycelium	0.0966	0.1823	0.1805
Difference	0.0258	0.0112	0.0162
Difference in µg	25.8	11.2	16.2

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

Y51L uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average Y51L uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
2.6	2.5	3.0	2.7 ±0.28	11.3	13.4	10.8	11.8 ±1.35	-0.3	-0.6	0.0	-0.3 ±0.28
Comparison with the respective wild type (T454) result (%)											
22.7	18.7	27.8	Average: 23.1%								
Growth test^a											
Y51L			T454				T110				
+			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct Y⁵¹ W Transformant 1226 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
23/10/07a											
T454	0	0.373	0.374	0.375	0.374	748	237	11.83	65.6	1.312	9.0
	20'	0.243	0.257	0.267	0.256	511					
T110	0	0.315	0.318	0.319	0.317	635	1	0.07	26.6	0.532	0.1
	20'	0.314	0.319	0.317	0.317	633					
Y51W	0	0.327	0.334	0.334	0.332	663	-1	-0.03	51.9	1.038	0.0
	20'	0.332	0.330	0.334	0.332	664					
13/02/08a											
T454	0	0.356	0.354	0.354	0.355	709	124	6.20	25.7	0.514	12.1
	20'	0.294	0.289	0.295	0.293	585					
T110	0	0.389	0.372	0.381	0.381	761	2	0.10	32.7	0.654	0.2
	20'	0.385	0.388	0.366	0.380	759					
Y51W	0	0.481	0.487	0.487	0.485	970	-2	-0.10	36	0.72	-0.1
	20'	0.484	0.482	0.492	0.486	972					
11/08/08											
T454	0	0.305	0.297	0.297	0.300	499	83	4.17	18.2	0.364	11.4
	20'	0.245	0.240	0.264	0.250	416					
T110	0	0.310	0.310	0.315	0.312	519	1	0.03	22.7	0.454	0.1
	20'	0.315	0.310	0.309	0.311	519					
Y51W	0	0.362	0.363	0.369	0.365	729	7	0.3	32.7	0.654	0.5
	20'	0.364	0.359	0.361	0.361	723					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 23/10/07a	T454	T110	Y51W
Weight of filter	0.1574	0.118	0.1452
Dry weight of mycelium	0.0918	0.091	0.0933
Difference	0.0656	0.027	0.0519
Difference in μg	65.6	26.6	51.9
Date of experiment: 13/02/08a	T454	T110	Y51W
Weight of filter	0.1156	0.123	0.1265
Dry weight of mycelium	0.0899	0.0903	0.0905
Difference	0.0257	0.0327	0.036
Difference in μg	25.7	32.7	36
Date of experiment: 11/08/08	T454	T110	Y51W
Weight of filter	0.109	0.1142	0.1233
Dry weight of mycelium	0.0908	0.0915	0.0906
Difference	0.0182	0.0227	0.0327
Difference in μg	18.2	22.7	32.7

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

Y51W uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average Y51W uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.0	-0.1	0.5	0.1 ±0.35	9.0	12.1	11.4	10.8 ±1.6	0.1	0.2	0.1	0.1 ±0.05
Comparison with the respective wild type (T454) result (%)											
0.0	-0.8	4.4	Average: 1.2%								
Growth test^a											
Y51W			T454				T110				
+			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct Y⁵¹F Transformant 85 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
04/06/07D											
T454	0	0.292	0.297	0.296	0.295	590	337	16.83	63	1.26	13.4
	20'	0.134	0.119	0.127	0.127	253					
T110	0	0.312	0.309	0.301	0.307	615	12	0.60	46.9	0.938	0.64
	20'	0.306	0.296	0.302	0.301	603					
Y51F	0	0.362	0.360	0.357	0.360	719	76	3.80	56.9	1.138	3.3
	20'	0.322	0.320	0.323	0.322	643					
13/06/07D											
T454	0	0.393	0.399	0.391	0.394	789	177	8.83	34.9	0.698	12.7
	20'	0.302	0.302	0.314	0.306	612					
T110	0	0.331	0.334	0.334	0.333	666	-7	-0.33	44.5	0.89	-0.4
	20'	0.330	0.336	0.343	0.336	673					
Y51F	0	0.381	0.388	0.385	0.385	769	86	4.30	54.7	1.094	3.9
	20'	0.344	0.345	0.336	0.342	683					
14/06/07											
T454	0	0.301	0.303	0.308	0.304	608	325	16.23	57.4	1.148	14.1
	20'	0.144	0.146	0.135	0.142	283					
T110	0	0.331	0.334	0.353	0.339	679	6	0.30	51	1.02	0.3
	20'	0.330	0.336	0.343	0.336	673					
Y51F	0	0.349	0.353	0.351	0.351	702	77	3.87	60.8	1.216	3.2
	20'	0.311	0.311	0.315	0.312	625					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 04/06/07D	T454	T110	Y51F
Weight of filter	0.2199	0.2254	0.2417
Dry weight of mycelium	0.1569	0.1785	0.1848
Difference	0.063	0.0469	0.0569
Difference in μg	63	46.9	56.9
Date of experiment: 13/06/07D	T454	T110	Y51F
Weight of filter	0.2636	0.2017	0.2384
Dry weight of mycelium	0.2287	0.1572	0.1837
Difference	0.0349	0.0445	0.0547
Difference in μg	34.9	44.5	54.7
Date of experiment: 14/06/07	T454	T110	Y51F
Weight of filter	0.2199	0.2254	0.2744
Dry weight of mycelium	0.1625	0.1744	0.2136
Difference	0.0574	0.051	0.0608
Difference in μg	57.4	51	60.8

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

Y51F uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average Y51F uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
3.3	3.9	3.2	3.5 ±0.40	13.4	12.7	14.1	13.4 ±0.74	0.6	-0.4	0.3	0.2 ±0.52
Comparison with the respective wild type (T454) result (%)											
25.0	31.0	22.0	Average: 26.0%								
Growth test^a											
Y51F			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F¹⁴⁰ L Transformant 1941 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
20/08/07B											
T454	0	0.339	0.337	0.334	0.337	673	245	12.23	67.4	1.348	9.1
	20'	0.216	0.217	0.210	0.214	429					
T110	0	0.366	0.386	0.371	0.374	749	-7	-0.33	47.7	0.954	-0.3
	20'	0.365	0.387	0.381	0.378	755					
F140L	0	0.292	0.287	0.288	0.289	578	14	0.70	55.1	1.102	0.6
	20'	0.289	0.280	0.277	0.282	564					
11/02/08B											
T454	0	0.361	0.375	0.365	0.367	734	123	6.17	32.3	0.646	9.5
	20'	0.311	0.304	0.301	0.305	611					
T110	0	0.375	0.356	0.375	0.369	737	-1	-0.03	25.2	0.504	-0.1
	20'	0.369	0.368	0.370	0.369	738					
F140L	0	0.381	0.388	0.380	0.383	766	16	0.80	34.7	0.694	1.2
	20'	0.372	0.375	0.378	0.375	750					
13/08/08											
T454	0	0.313	0.300	0.300	0.304	507	109	5.47	30.9	0.618	8.9
	20'	0.235	0.241	0.240	0.239	398					
T110	0	0.317	0.310	0.319	0.315	526	-3	-0.17	11.2	0.224	-0.7
	20'	0.307	0.317	0.328	0.317	529					
F140L	0	0.310	0.312	0.312	0.311	519	9	0.44	26.3	0.526	0.8
	20'	0.307	0.305	0.306	0.306	510					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 20/08/07B	T454	T110	F140L
Weight of filter	0.1589	0.1394	0.1464
Dry weight of mycelium	0.0915	0.0917	0.0913
Difference	0.0674	0.0477	0.0551
Difference in μg	67.4	47.7	55.1
Date of experiment: 11/02/08B	T454	T110	F140L
Weight of filter	0.123	0.1157	0.1249
Dry weight of mycelium	0.0907	0.0905	0.0902
Difference	0.0323	0.0252	0.0347
Difference in μg	32.3	25.2	34.7
Date of experiment: 13/08/08	T454	T110	F140L
Weight of filter	0.1201	0.1935	0.1178
Dry weight of mycelium	0.0892	0.1823	0.0915
Difference	0.0309	0.0112	0.0263
Difference in μg	30.9	11.2	26.3

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F140L uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F140L uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.6	1.2	0.8	0.9 ±0.26	9.1	9.5	8.9	9.2 ±0.35	-0.3	-0.1	-0.7	-0.4 ±0.34
Comparison with the respective wild type (T454) result (%)											
7.0	12.1	9.5	Average: 7.0%								
Growth test^a											
F140L			T454				T110				
++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F¹⁵¹L Transformant 1757 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
23/10/07A											
T454	0	0.373	0.374	0.375	0.374	748	257	12.87	65.6	1.312	9.8
	20'	0.243	0.247	0.246	0.245	491					
T110	0	0.315	0.318	0.319	0.317	635	1	0.07	26.6	0.532	0.1
	20'	0.314	0.319	0.317	0.317	633					
F151L	0	0.352	0.349	0.349	0.350	700	11	0.5	71.6	1.432	0.4
	20'	0.344	0.343	0.347	0.345	689					
23/10/07B											
T454	0	0.377	0.380	0.376	0.378	755	239	11.97	65.5	1.31	9.1
	20'	0.258	0.256	0.260	0.258	516					
T110	0	0.315	0.315	0.311	0.314	627	-3	-0.13	22.4	0.448	-0.3
	20'	0.312	0.316	0.317	0.315	630					
F151L	0	0.358	0.356	0.359	0.358	715	35	1.73	61.6	1.232	1.4
	20'	0.341	0.339	0.341	0.340	681					
23/10/07B											
T454	0	0.377	0.380	0.376	0.378	755	239	11.97	65.5	1.31	9.1
	20'	0.258	0.256	0.260	0.258	516					
T110	0	0.315	0.315	0.311	0.314	627	-3	-0.13	22.4	0.448	-0.3
	20'	0.312	0.316	0.317	0.315	630					
F151L	0	0.345	0.346	0.344	0.345	690	17	0.87	65.6	1.312	0.7
	20'	0.336	0.338	0.335	0.336	673					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 23/10/07A	T454	T110	F151L
Weight of filter	0.1574	0.118	0.1638
Dry weight of mycelium	0.0918	0.091	0.0922
Difference	0.0656	0.027	0.0716
Difference in µg	65.6	26.6	71.6
Date of experiment: 23/10/07B	T454	T110	F151L
Weight of filter	0.157	0.1154	0.1545
Dry weight of mycelium	0.0915	0.093	0.0929
Difference	0.0655	0.0224	0.0616
Difference in µg	65.5	22.4	61.6
Date of experiment: 23/10/07B	T454	T110	F151L
Weight of filter	0.157	0.1154	0.1575
Dry weight of mycelium	0.0915	0.093	0.0919
Difference	0.0655	0.0224	0.0656
Difference in µg	65.5	22.4	65.6

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F151L uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F151L uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.4	1.4	0.7	0.8 ±0.53	9.8	9.1	9.1	9.4 ±0.39	0.1	-0.3	-0.3	-0.2 ±0.24
Comparison with the respective wild type (T454) result (%)											
4.0	15.0	7.0	Average: 4.0%								
Growth test ^a											
F151L			T454				T110				
++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F¹⁵¹S Transformant 1914 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
23/10/07A											
T454	0	0.377	0.380	0.376	0.378	755	239	11.97	65.5	1.31	9.1
	20'	0.258	0.256	0.260	0.258	516					
T110	0	0.315	0.315	0.311	0.314	627	-3	-0.13	22.4	0.448	-0.3
	20'	0.312	0.316	0.317	0.315	630					
F151S	0	0.343	0.349	0.341	0.344	689	168	8.40	57.4	1.148	7.3
	20'	0.257	0.259	0.265	0.260	521					
23/10/07B											
T454	0	0.389	0.387	0.391	0.389	778	281	14.07	74.3	1.486	9.5
	20'	0.245	0.241	0.259	0.248	497					
T110	0	0.301	0.302	0.303	0.302	604	1	0.07	16.7	0.334	0.2
	20'	0.300	0.299	0.305	0.301	603					
F151S	0	0.344	0.348	0.348	0.347	693	166	8.3	51.7	1.034	8.0
	20'	0.260	0.268	0.263	0.264	527					
23/10/07C											
T454	0	0.410	0.415	0.421	0.415	831	317	15.87	64.3	1.286	12.3
	20'	0.254	0.256	0.260	0.257	513					
T110	0	0.300	0.315	0.311	0.309	617	-13	-0.67	34.8	0.696	-1.0
	20'	0.311	0.316	0.319	0.315	631					
F151S	0	0.348	0.346	0.345	0.346	693	167	8.4	56.7	1.134	7.4
	20'	0.264	0.263	0.261	0.263	525					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 23/10/07A	T454	T110	F151S
Weight of filter	0.157	0.1154	0.1497
Dry weight of mycelium	0.092	0.093	0.0923
Difference	0.066	0.0224	0.0574
Difference in µg	65.5	22.4	57.4
Date of experiment: 23/10/07B	T454	T110	F151S
Weight of filter	0.1658	0.1154	0.1445
Dry weight of mycelium	0.0915	0.0987	0.0928
Difference	0.0743	0.0167	0.0517
Difference in µg	74.3	16.7	51.7
Date of experiment: 23/10/07C	T454	T110	F151S
Weight of filter	0.1547	0.1247	0.1467
Dry weight of mycelium	0.0904	0.0899	0.09
Difference	0.0643	0.0348	0.0567
Difference in µg	64.3	34.8	56.7

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F151S uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F151S uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
7.3	8.0	7.4	7.6 ±0.39	9.1	9.5	12.3	10.3 ±1.76	-0.3	0.2	-1.0	-0.4 ±0.58
Comparison with the respective wild type (T454) result (%)											
80.0	85.0	60.0	Average: 80.0%								
Growth test^a											
F151S			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F¹⁵¹W Transformant 1779 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
10/10/07B											
T454	0	0.301	0.303	0.307	0.304	607	243	12.13	62.4	1.248	9.7
	20'	0.182	0.183	0.182	0.182	365					
T110	0	0.301	0.302	0.300	0.301	602	-2	-0.10	44.6	0.892	-0.1
	20'	0.299	0.303	0.304	0.302	604					
F151W	0	0.309	0.310	0.308	0.309	618	19	0.93	57.6	1.152	0.8
	20'	0.299	0.301	0.299	0.300	599					
16/10/07B											
T454	0	0.319	0.320	0.313	0.317	635	148	7.40	28	0.56	13.2
	20'	0.249	0.242	0.239	0.243	487					
T110	0	0.317	0.319	0.318	0.318	636	1	0.07	22.4	0.448	0.1
	20'	0.315	0.320	0.317	0.317	635					
F151W	0	0.317	0.316	0.319	0.317	635	7	0.33	24.4	0.488	0.7
	20'	0.314	0.316	0.312	0.314	628					
30/10/07C											
T454	0	0.341	0.338	0.337	0.339	677	147	7.33	42	0.84	8.7
	20'	0.262	0.271	0.263	0.265	531					
T110	0	0.349	0.352	0.354	0.352	703	-3	-0.13	28	0.56	-0.2
	20'	0.361	0.347	0.351	0.353	706					
F151W	0	0.327	0.330	0.329	0.329	657	23	1.13	64.2	1.284	0.9
	20'	0.319	0.317	0.316	0.317	635					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 10/10/07B	T454	T110	F151W
Weight of filter	0.1545	0.137	0.1504
Dry weight of mycelium	0.0921	0.092	0.0928
Difference	0.0624	0.045	0.0576
Difference in μg	62.4	44.6	57.6
Date of experiment: 16/10/07B	T454	T110	F151W
Weight of filter	0.1219	0.1154	0.1166
Dry weight of mycelium	0.0939	0.093	0.0922
Difference	0.028	0.0224	0.0244
Difference in μg	28	22.4	24.4
Date of experiment: 30/10/07C	T454	T110	F151W
Weight of filter	0.1341	0.1205	0.156
Dry weight of mycelium	0.0921	0.0925	0.0918
Difference	0.042	0.028	0.0642
Difference in μg	42	28	64.2

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F151W uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F151W uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.8	0.7	0.9	0.8 ±0.10	9.7	13.2	8.7	10.6 ±2.36	-0.1	0.1	-0.2	-0.1 ±0.20
Comparison with the respective wild type (T454) result (%)											
8.0	5.0	10.0	Average: 8.0%								
Growth test^a											
F151W			T454			T110					
+++			+++			-					

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct F¹⁵¹Y Transformant 18 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
04/06/07C											
T454	0	0.346	0.552	0.360	0.419	839	312	15.60	54.6	1.092	14.3
	20'	0.264	0.266	0.260	0.263	527					
T110	0	0.340	0.345	0.349	0.345	689	-10	-0.50	63.5	1.27	-0.39
	20'	0.348	0.350	0.351	0.350	699					
F151Y	0	0.375	0.376	0.381	0.377	755	95	4.73	33.9	0.678	7.0
	20'	0.326	0.327	0.337	0.330	660					
13/06/07C											
T454	0	0.405	0.419	0.422	0.415	831	305	15.27	47.3	0.946	16.1
	20'	0.269	0.261	0.258	0.263	525					
T110	0	0.368	0.368	0.368	0.368	736	-19	-0.93	49.5	0.99	-0.9
	20'	0.367	0.375	0.390	0.377	755					
F151Y	0	0.385	0.388	0.386	0.386	773	67	3.33	27.4	0.548	6.1
	20'	0.360	0.346	0.353	0.353	706					
28/06/07B											
T454	0	0.520	0.515	0.513	0.516	1032	282	14.10	84.6	1.692	8.3
	20'	0.377	0.372	0.376	0.375	750					
T110	0	0.411	0.410	0.399	0.407	813	-2	-0.10	74	1.48	-0.1
	20'	0.408	0.405	0.410	0.408	815					
F151Y	0	0.571	0.599	0.583	0.584	1169	143	7.13	52	1.04	6.9
	20'	0.529	0.510	0.500	0.513	1026					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 04/06/07C	T454	T110	F151Y
Weight of filter	0.291	0.2474	0.233
Dry weight of mycelium	0.237	0.1839	0.199
Difference	0.055	0.0635	0.034
Difference in μg	54.6	63.5	33.9
Date of experiment: 13/06/07C	T454	T110	F151Y
Weight of filter	0.194	0.2021	0.21
Dry weight of mycelium	0.146	0.1526	0.19
Difference	0.047	0.0495	0.03
Difference in μg	47.3	49.5	27.4
Date of experiment: 28/06/07B	T454	T110	F151Y
Weight of filter	0.175	0.165	0.145
Dry weight of mycelium	0.091	0.091	0.093
Difference	0.085	0.074	0.052
Difference in μg	84.6	74	52

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

F151Y uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average F151Y uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
7.0	6.1	6.9	6.6 ±0.49	14.3	16.1	8.3	12.9 ±4.0	-0.39	-0.9	-0.1	-0.46 ±0.40
Comparison with the respective wild type (T454) result (%)											
49.0	37.8	83.0	Average: 56.0%								
Growth test^a											
F151Y			T454				T110				
+++			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct W¹⁹⁷Y Transformant 3766 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
31/07/08C											
T454	0	0.415	0.410	0.412	0.412	687	147	7.36	34.7	0.694	10.6
	20'	0.324	0.327	0.321	0.324	540					
T110	0	0.425	0.425	0.418	0.423	704	-6	-0.28	50.3	1.006	-0.3
	20'	0.429	0.423	0.426	0.426	710					
W197Y	0	0.484	0.474	0.476	0.478	797	6	0.28	49.1	0.982	0.3
	20'	0.472	0.478	0.474	0.475	791					
31/07/08C											
T454	0	0.415	0.410	0.412	0.412	687	147	7.36	34.7	0.694	10.6
	20'	0.324	0.327	0.321	0.324	540					
T110	0	0.425	0.425	0.418	0.423	704	-6	-0.28	50.3	1.006	-0.3
	20'	0.429	0.423	0.426	0.426	710					
W197Y	0	0.450	0.451	0.442	0.448	746	2	0.11	34.6	0.692	0.2
	20'	0.449	0.447	0.443	0.446	744					
01/08/08C											
T454	0	0.420	0.424	0.429	0.424	707	261	13.06	50.5	1.01	12.9
	20'	0.276	0.265	0.262	0.268	446					
T110	0	0.407	0.377	0.406	0.397	661	-2	-0.08	37.2	0.744	-0.1
	20'	0.401	0.392	0.400	0.398	663					
W197Y	0	0.379	0.381	0.388	0.383	638	11	0.56	42.3	0.846	0.7
	20'	0.373	0.376	0.379	0.376	627					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 31/07/08C	T454	T110	W197Y
Weight of filter	0.1255	0.141	0.1398
Dry weight of mycelium	0.0908	0.091	0.0907
Difference	0.0347	0.05	0.0491
Difference in μg	34.7	50.3	49.1
Date of experiment: 31/07/08C	T454	T110	W197Y
Weight of filter	0.1255	0.141	0.1262
Dry weight of mycelium	0.0908	0.091	0.0916
Difference	0.0347	0.05	0.0346
Difference in μg	34.7	50.3	34.6
Date of experiment: 01/08/08C	T454	T110	W197Y
Weight of filter	0.1409	0.1278	0.1325
Dry weight of mycelium	0.0904	0.0906	0.0902
Difference	0.0505	0.0372	0.0423
Difference in μg	50.5	37.2	42.3

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

W197Y uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average W197Y uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
0.3	0.2	0.7	0.4 ± 0.26	10.6	10.6	12.9	11.8 ± 1.6	-0.3	-0.3	-0.1	-0.2 ± 0.12
Comparison with the respective wild type (T454) result (%)											
2.7	1.5	5.1	Average: 3.1%								
Growth test^a											
W197Y			T454				T110				
+-			+++				-				

The Phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Chapter 6

Construct N³⁶⁴A Transformant 1814 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
08/02/08D											
T454	0	0.374	0.364	0.368	0.369	737	142	7.10	28.2	0.564	12.6
	20'	0.296	0.296	0.301	0.298	595					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
N364A	0	0.353	0.359	0.355	0.356	711	45	2.23	32	0.64	3.5
	20'	0.330	0.335	0.335	0.333	667					
11/08/08											
T454	0	0.305	0.297	0.297	0.300	499	94	4.72	18.2	0.364	13.0
	20'	0.245	0.240	0.244	0.243	405					
T110	0	0.310	0.310	0.335	0.318	531	3	0.14	22.7	0.454	0.3
	20'	0.315	0.310	0.325	0.317	528					
N364A	0	0.300	0.299	0.304	0.301	502	32	1.61	20.5	0.41	3.9
	20'	0.284	0.280	0.281	0.282	469					
21/08/08											
T454	0	0.314	0.305	0.310	0.310	619	195	9.73	32.8	0.656	14.8
	20'	0.211	0.215	0.211	0.212	425					
T110	0	0.280	0.275	0.274	0.276	553	-9	-0.47	27.2	0.544	-0.9
	20'	0.283	0.279	0.281	0.281	562					
N364A	0	0.394	0.399	0.388	0.394	787	35	1.73	21.4	0.428	4.0
	20'	0.382	0.376	0.371	0.376	753					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 08/02/08D	T454	T110	N364A
Weight of filter	0.1196	0.1204	0.1253
Dry weight of mycelium	0.0914	0.0925	0.0933
Difference	0.0282	0.0279	0.032
Difference in µg	28.2	27.9	32
Date of experiment: 11/08/08	T454	T110	N364A
Weight of filter	0.109	0.1142	0.1118
Dry weight of mycelium	0.0908	0.0915	0.0913
Difference	0.0182	0.0227	0.0205
Difference in µg	18.2	22.7	20.5
Date of experiment: 21/08/08	T454	T110	N364A
Weight of filter	0.1254	0.1182	0.112
Dry weight of mycelium	0.0926	0.091	0.0906
Difference	0.0328	0.0272	0.0214
Difference in µg	32.8	27.2	21.4

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

N364A uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average N364A uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
3.5	3.9	4.0	3.8 ±0.29	12.6	13.0	14.8	13.5 ±1.2	-0.5	0.3	-0.9	-0.3 ±0.59
Comparison with the respective wild type (T454) result (%)											
28.0	30.0	27.0	Average: 28.0%								
Growth test^a											
N364A			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct N³⁶⁴Q transformant 3902 Colony 1 with both controls: T454 (wild type) and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
05/08/08A											
T454	0	0.399	0.395	0.394	0.396	660	197	9.86	44.3	0.886	11.1
	20'	0.274	0.280	0.279	0.278	463					
T110	0	0.382	0.389	0.387	0.386	643	0	0.00	29.8	0.596	0.0
	20'	0.388	0.389	0.381	0.386	643					
N364Q	0	0.377	0.376	0.371	0.375	624	22	1.08	22	0.44	2.5
	20'	0.357	0.367	0.361	0.362	603					
05/08/08B											
T454	0	0.374	0.363	0.379	0.372	620	151	7.53	31.9	0.638	11.8
	20'	0.287	0.281	0.277	0.282	469					
T110	0	0.362	0.378	0.381	0.374	623	-25	-1.25	41.3	0.826	-1.5
	20'	0.381	0.392	0.393	0.389	648					
N364Q	0	0.382	0.384	0.376	0.381	634	27	1.33	28.6	0.572	2.3
	20'	0.365	0.366	0.363	0.365	608					
07/08/08B											
T454	0	0.338	0.337	0.343	0.339	566	103	5.17	20.1	0.402	12.9
	20'	0.273	0.276	0.283	0.277	462					
T110	0	0.334	0.330	0.327	0.330	551	-3	-0.17	17	0.34	-0.5
	20'	0.329	0.331	0.337	0.332	554					
N364Q	0	0.354	0.350	0.356	0.353	589	16	0.81	18.1	0.362	2.2
	20'	0.346	0.340	0.345	0.344	573					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 05/08/08A	T454	T110	N³⁶⁴Q
Weight of filter	0.1365	0.1215	0.1123
Dry weight of mycelium	0.0922	0.0917	0.0903
Difference	0.0443	0.0298	0.022
Difference in µg	44.3	29.8	22
Date of experiment: 05/08/08B	T454	T110	N³⁶⁴Q
Weight of filter	0.1236	0.133	0.1194
Dry weight of mycelium	0.0917	0.0917	0.0908
Difference	0.0319	0.0413	0.0286
Difference in µg	31.9	41.3	28.6
Date of experiment: 07/08/08B	T454	T110	N³⁶⁴Q
Weight of filter	0.1117	0.1088	0.1098
Dry weight of mycelium	0.0916	0.0918	0.0917
Difference	0.0201	0.017	0.0181
Difference in µg	20.1	17	18.1

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

N364 Q uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average N364 Q uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
2.5	2.3	2.2	2.3 ±0.12	11.1	11.8	12.9	11.9 ±0.87	0.0	-1.5	-0.5	-0.7 ±0.77
Comparison with the respective wild type (T454) result (%)											
22.0	20.0	17.0	Average: 20.0%								
Growth test^a											
N³⁶⁴ Q			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Chapter 7

Construct K¹⁹L Transformant 3854 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
05/08/08											
T454	0	0.146	0.148	0.134	0.143	285	104	5.20	23.7	0.474	11.0
	20'	0.091	0.081	0.100	0.091	181					
T110	0	0.145	0.147	0.151	0.148	295	1	0.03	27.6	0.552	0.1
	20'	0.147	0.146	0.149	0.147	295					
K19L	0	0.406	0.406	0.402	0.405	674	99	4.97	32.2	0.644	7.7
	20'	0.344	0.345	0.346	0.345	575					
06/08/08A											
T454	0	0.353	0.366	0.364	0.361	602	132	6.61	33.9	0.678	9.8
	20'	0.286	0.288	0.271	0.282	469					
T110	0	0.352	0.363	0.362	0.359	598	-9	-0.47	24.4	0.488	-1.0
	20'	0.358	0.367	0.369	0.365	608					
K19L	0	0.401	0.400	0.399	0.400	667	106	5.31	30	0.6	8.8
	20'	0.334	0.336	0.339	0.336	561					
08/08/08											
T454	0	0.291	0.313	0.311	0.305	508	91	4.56	20.4	0.408	11.2
	20'	0.243	0.255	0.253	0.250	417					
T110	0	0.328	0.329	0.327	0.328	547	2	0.11	22	0.44	0.3
	20'	0.329	0.326	0.325	0.327	544					
K19L	0	0.349	0.343	0.345	0.346	576	69	3.44	24	0.48	7.2
	20'	0.303	0.307	0.303	0.304	507					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 05/08/08	T454	T110	K19L
Weight of filter	0.114	0.1282	0.1244
Dry weight of mycelium	0.0903	0.1006	0.0922
Difference	0.0237	0.0276	0.0322
Difference in μg	23.7	27.6	32.2
Date of experiment: 06/08/08A	T454	T110	K19L
Weight of filter	0.1253	0.1162	0.1227
Dry weight of mycelium	0.0914	0.0918	0.0927
Difference	0.0339	0.0244	0.03
Difference in μg	33.9	24.4	30
Date of experiment: 08/08/08	T454	T110	K19L
Weight of filter	0.1121	0.1132	0.1156
Dry weight of mycelium	0.0917	0.0912	0.0916
Difference	0.0204	0.022	0.024
Difference in μg	20.4	22	24

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

K19L uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average K19L uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
7.7	8.8	7.2	7.9 ± 0.85	11.0	9.8	11.2	10.6 ± 0.77	0.1	-1.0	0.3	-0.2 ± 0.66
Comparison with the respective wild type (T454) result (%)											
70.0	91.0	64.0	Average: 70.0%								
Growth test^a											
K19L			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct R¹³⁰K Transformant 2401 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
20/08/07C											
T454	0	0.466	0.467	0.467	0.467	933	289	14.47	86.2	1.724	8.4
	20'	0.325	0.322	0.319	0.322	644					
T110	0	0.391	0.370	0.388	0.383	766	-21	-1.07	68.4	1.368	-0.8
	20'	0.386	0.394	0.401	0.394	787					
R130K	0	0.445	0.441	0.449	0.445	890	48	2.40	70.4	1.408	1.7
	20'	0.417	0.421	0.425	0.421	842					
04/10/07B											
T454	0	0.233	0.233	0.236	0.234	468	116	5.80	31.9	0.638	9.1
	20'	0.174	0.174	0.180	0.176	352					
T110	0	0.222	0.221	0.229	0.224	448	4	0.20	38.5	0.77	0.3
	20'	0.225	0.220	0.221	0.222	444					
R130K	0	0.243	0.249	0.257	0.250	499	26	1.30	35.2	0.704	1.8
	20'	0.238	0.233	0.239	0.237	473					
16/10/07B											
T454	0	0.301	0.302	0.305	0.303	605	119	5.93	30.8	0.616	9.6
	20'	0.234	0.249	0.247	0.243	487					
T110	0	0.316	0.315	0.319	0.317	633	4	0.20	24.6	0.492	0.4
	20'	0.312	0.318	0.314	0.315	629					
R130K	0	0.246	0.240	0.241	0.242	485	32	1.60	36.8	0.736	2.2
	20'	0.223	0.229	0.227	0.226	453					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 20/08/07C	T454	T110	R130K
Weight of filter	0.1772	0.16	0.162
Dry weight of mycelium	0.091	0.091	0.091
Difference	0.0862	0.068	0.07
Difference in μg	86.2	68.4	70.4
Date of experiment: 04/10/07B	T454	T110	R130K
Weight of filter	0.1279	0.1322	0.1302
Dry weight of mycelium	0.096	0.0937	0.095
Difference	0.0319	0.0385	0.0352
Difference in μg	31.9	38.5	35.2
Date of experiment: 16/10/07B	T454	T110	R130K
Weight of filter	0.1225	0.1176	0.1302
Dry weight of mycelium	0.0917	0.093	0.0934
Difference	0.0308	0.0246	0.0368
Difference in μg	30.8	24.6	36.8

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

R130K uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average R130K uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
1.7	1.8	2.2	1.9±0.24	8.4	9.1	9.6	9.0 ±0.62	-0.8	0.3	0.4	0.0 ±0.65
Comparison with the respective wild type (T454) result (%)											
20.3	20.3	22.6	Average: 21.1%								
Growth test^a											
R130K			T454				T110				
+-			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct R¹³⁰Q Transformant 2439 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
08/10/07B											
T454	0	0.305	0.303	0.307	0.305	610	246	12.30	61.9	1.238	9.9
	20'	0.180	0.182	0.184	0.182	364					
T110	0	0.304	0.309	0.301	0.305	609	11	0.53	44	0.88	0.6
	20'	0.299	0.301	0.298	0.299	599					
R130Q	0	0.285	0.289	0.284	0.286	572	-13	-0.67	33.5	0.67	-1.0
	20'	0.297	0.297	0.284	0.293	585					
10/10/07A											
T454	0	0.301	0.302	0.305	0.303	605	99	4.93	30.8	0.616	8.0
	20'	0.254	0.249	0.257	0.253	507					
T110	0	0.316	0.315	0.319	0.317	633	4	0.20	24.6	0.492	0.4
	20'	0.312	0.318	0.314	0.315	629					
R130Q	0	0.352	0.357	0.387	0.365	731	-5	-0.27	37.7	0.754	-0.4
	20'	0.375	0.365	0.364	0.368	736					
16/10/07B											
T454	0	0.320	0.319	0.321	0.320	640	215	10.77	47.1	0.942	11.4
	20'	0.216	0.210	0.211	0.212	425					
T110	0	0.332	0.330	0.336	0.333	665	-8	-0.40	22.7	0.454	-0.9
	20'	0.336	0.339	0.335	0.337	673					
R130Q	0	0.373	0.381	0.368	0.374	748	-5	-0.27	36.7	0.734	-0.4
	20'	0.376	0.382	0.372	0.377	753					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 08/10/07B	T454	T110	R130Q
Weight of filter	0.1547	0.136	0.1252
Dry weight of mycelium	0.0928	0.092	0.0917
Difference	0.0619	0.044	0.0335
Difference in μg	61.9	44	33.5
Date of experiment: 10/10/07A	T454	T110	R130Q
Weight of filter	0.1225	0.1176	0.1279
Dry weight of mycelium	0.0917	0.093	0.0902
Difference	0.0308	0.0246	0.0377
Difference in μg	30.8	24.6	37.7
Date of experiment: 16/10/07B	T454	T110	R130Q
Weight of filter	0.1441	0.1172	0.1309
Dry weight of mycelium	0.097	0.0945	0.0942
Difference	0.0471	0.0227	0.0367
Difference in μg	47.1	22.7	36.7

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

R130Q uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average R130Q uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T454 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T454 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)	T110 uptake in triplicates ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)			Average T110 uptake ($\text{nmol min}^{-1} \text{mg}^{-1} \text{DW}$)
-1.0	-0.4	-0.4	-0.6 ±0.37	9.9	8.0	11.4	9.8 ±1.7	0.6	0.4	-0.9	0.0 ±0.81
Comparison with the respective wild type (T454) result (%)											
0.0	0.0	0.0	Average: 0.0%								
Growth test^a											
R130Q			T454				T110				
+-			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct R¹⁹⁸Q Transformant 3055 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
20/02/08B											
T454	0	0.366	0.354	0.366	0.362	724	143	7.17	25.7	0.514	13.9
	20'	0.291	0.291	0.289	0.290	581					
T110	0	0.379	0.372	0.371	0.374	748	-7	-0.37	32.7	0.654	-0.6
	20'	0.381	0.378	0.374	0.378	755					
R198Q	0	0.345	0.352	0.354	0.350	701	39	1.93	22	0.44	4.4
	20'	0.331	0.330	0.332	0.331	662					
11/08/08B											
T454	0	0.350	0.354	0.358	0.354	708	131	6.53	28.2	0.564	11.6
	20'	0.286	0.286	0.294	0.289	577					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.23	27.9	0.558	-0.4
	20'	0.344	0.354	0.359	0.352	705					
R198Q	0	0.401	0.400	0.403	0.401	803	42	2.10	25.9	0.518	4.1
	20'	0.376	0.381	0.384	0.380	761					
08/08/08											
T454	0	0.291	0.313	0.311	0.305	508	91	4.56	20.4	0.408	11.2
	20'	0.243	0.255	0.253	0.250	417					
T110	0	0.328	0.329	0.327	0.328	547	-9	-0.44	22	0.44	-1.0
	20'	0.329	0.346	0.325	0.333	556					
R198Q	0	0.361	0.363	0.362	0.362	724	40	2.00	26.1	0.522	3.8
	20'	0.342	0.341	0.343	0.342	684					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 20/02/08B	T454	T110	R198Q
Weight of filter	0.1156	0.123	0.1131
Dry weight of mycelium	0.0899	0.0903	0.0911
Difference	0.0257	0.0327	0.022
Difference in μg	25.7	32.7	22
Date of experiment: 11/08/08B	T454	T110	R198Q
Weight of filter	0.1196	0.1204	0.1164
Dry weight of mycelium	0.0914	0.0925	0.0905
Difference	0.0282	0.0279	0.0259
Difference in μg	28.2	27.9	25.9
Date of experiment: 08/08/08	T454	T110	R198Q
Weight of filter	0.1121	0.1132	0.1158
Dry weight of mycelium	0.0917	0.0912	0.0897
Difference	0.0204	0.022	0.0261
Difference in μg	20.4	22	26.1

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

R198Q uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average R198Q uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
4.4	4.1	3.8	4.1 ±0.28	13.9	11.6	11.2	12.2 ±1.5	-0.6	-0.4	-1.0	-0.7 ±0.31
Comparison with the respective wild type (T454) result (%)											
31.5	35.0	34.3	Average: 33.6%								
Growth test^a											
R198Q			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct K³⁸⁸L Transformant 3121 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
13/02/08A											
T454	0	0.354	0.357	0.360	0.357	714	95	4.73	25.7	0.514	9.2
	20'	0.310	0.315	0.304	0.310	619					
T110	0	0.401	0.410	0.381	0.397	795	-7	-0.37	32.7	0.654	-0.6
	20'	0.415	0.402	0.386	0.401	802					
K388L	0	0.367	0.374	0.374	0.372	743	91	4.6	39.4	0.788	5.8
	20'	0.330	0.327	0.321	0.326	652					
13/02/08C											
T454	0	0.354	0.364	0.368	0.362	724	129	6.43	28.2	0.564	11.4
	20'	0.296	0.296	0.301	0.298	595					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
K388L	0	0.482	0.473	0.485	0.480	960	71	3.57	28.1	0.562	6.3
	20'	0.447	0.445	0.441	0.444	889					
11/08/08											
T454	0	0.289	0.297	0.297	0.294	491	87	4.36	18.2	0.364	12.0
	20'	0.245	0.240	0.241	0.242	403					
T110	0	0.310	0.310	0.335	0.318	531	3	0.14	22.7	0.454	0.3
	20'	0.315	0.310	0.325	0.317	528					
K388L	0	0.297	0.298	0.315	0.303	506	57	2.83	23.9	0.478	5.9
	20'	0.271	0.268	0.269	0.269	449					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 13/02/08A	T454	T110	K388L
Weight of filter	0.1156	0.123	0.1289
Dry weight of mycelium	0.0899	0.0903	0.0895
Difference	0.0257	0.0327	0.0394
Difference in μg	25.7	32.7	39.4
Date of experiment: 13/02/08C	T454	T110	K388L
Weight of filter	0.1196	0.1204	0.118
Dry weight of mycelium	0.0914	0.0925	0.0899
Difference	0.0282	0.0279	0.0281
Difference in μg	28.2	27.9	28.1
Date of experiment: 11/08/08	T454	T110	K388L
Weight of filter	0.109	0.1142	0.1164
Dry weight of mycelium	0.0908	0.0915	0.0925
Difference	0.0182	0.0227	0.0239
Difference in μg	18.2	22.7	23.9

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

K388L uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average K388L uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
5.8	6.3	5.9	6.0 ± 0.29	9.2	11.4	12.0	10.9 ± 1.46	-0.6	-0.5	0.3	-0.2 ± 0.48
Comparison with the respective wild type (T454) result (%)											
63.0	56.0	49.0	Average: 56.0%								
Growth test^a											
K388L			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct R¹³⁰Q/R¹⁹⁸Q Transformant 3827 Colony 1 with both controls: T454 (wild type) and T110 (negative)

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
20/08/07											
T454	0	0.399	0.395	0.394	0.396	660	197	9.86	44.3	0.886	11.1
	20'	0.274	0.280	0.279	0.278	463					
T110	0	0.382	0.389	0.387	0.386	643	0	0.00	29.8	0.596	0.0
	20'	0.388	0.389	0.381	0.386	643					
R130Q/R198Q	0	0.399	0.401	0.400	0.400	667	57	2.86	50.2	1.004	2.8
	20'	0.368	0.365	0.364	0.366	609					
11/02/08											
T454	0	0.374	0.373	0.379	0.375	626	138	6.92	31.9	0.638	10.8
	20'	0.291	0.292	0.294	0.292	487					
T110	0	0.362	0.378	0.381	0.374	623	-14	-0.69	41.3	0.826	-0.8
	20'	0.381	0.372	0.393	0.382	637					
R130Q/R198Q	0	0.394	0.393	0.392	0.393	655	48	2.42	44.4	0.888	2.7
	20'	0.368	0.364	0.360	0.364	607					
13/02/08											
T454	0	0.338	0.337	0.343	0.339	566	103	5.17	20.1	0.402	12.9
	20'	0.273	0.276	0.283	0.277	462					
T110	0	0.354	0.343	0.347	0.348	580	-11	-0.53	30.2	0.604	-0.9
	20'	0.353	0.356	0.354	0.354	591					
R130Q/R198Q	0	0.349	0.347	0.343	0.346	577	18	0.92	31.4	0.628	1.5
	20'	0.336	0.333	0.337	0.335	559					

Table containing the raw data obtained on three independent net nitrate uptake assays

Dry Mycelial Weight Calculations

Calculating the Mycelial Weight			
Date of experiment: 05/08/08A	T454	T110	R130Q/R198Q
Weight of filter	0.1365	0.1215	0.1411
Dry weight of mycelium	0.0922	0.0917	0.0909
Difference	0.0443	0.0298	0.0502
Difference in μg	44.3	29.8	50.2
Date of experiment: 05/08/08B	T454	T110	R130Q/R198Q
Weight of filter	0.1236	0.133	0.1361
Dry weight of mycelium	0.0917	0.0917	0.0917
Difference	0.0319	0.0413	0.0444
Difference in μg	31.9	41.3	44.4
Date of experiment: 07/08/08	T454	T110	R130Q/R198Q
Weight of filter	0.1117	0.1215	0.1225
Dry weight of mycelium	0.0916	0.0913	0.0911
Difference	0.0201	0.0302	0.0314
Difference in μg	20.1	30.2	31.4

Table containing the raw data obtained on three independent net nitrate uptake assays dry mycelial weight

Average of net nitrate uptake results of three independent experiments including both sets of controls

R130Q/R198Q uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average R130Q/R198Q uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
2.8	2.7	1.5	2.3 ±0.77	11.1	10.8	12.9	11.6 ±1.1	0.0	-0.8	-0.9	-0.6 ±0.5
Comparison with the respective wild type (T454) result (%)											
25.7	25.2	11.3	Average: 20.7%								
Growth test ^a											
R130Q/R198Q			T454				T110				
++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Chapter 8

Construct A²⁰C Transformant 3742 Colony 1 Controls: T454 (wild type) positive and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
31/07/08B											
T454	0	0.414	0.419	0.410	0.414	691	149	7.47	23.2	0.464	16.1
	20'	0.323	0.327	0.324	0.325	541					
T110	0	0.318	0.322	0.312	0.317	529	1	0.06	29.1	0.582	0.1
	20'	0.321	0.322	0.307	0.317	528					
A20C	0	0.422	0.411	0.421	0.418	697	189	9.44	40.4	0.808	11.7
	20'	0.304	0.301	0.309	0.305	508					
01/08/08											
T454	0	0.456	0.446	0.448	0.450	750	179	8.97	26.7	0.534	16.8
	20'	0.341	0.342	0.344	0.342	571					
T110	0	0.417	0.424	0.414	0.418	697	1	0.03	34.7	0.694	0.0
	20'	0.415	0.420	0.419	0.418	697					
A20C	0	0.434	0.429	0.435	0.433	721	185	9.25	42.9	0.858	10.8
	20'	0.326	0.321	0.318	0.322	536					
21/08/08											
T454	0	0.314	0.315	0.320	0.316	633	215	10.73	32.8	0.656	16.4
	20'	0.210	0.209	0.208	0.209	418					
T110	0	0.283	0.295	0.294	0.291	581	-4	-0.20	27.2	0.544	-0.4
	20'	0.289	0.296	0.293	0.293	585					
A20C	0	0.310	0.300	0.307	0.306	611	113	5.67	23.7	0.474	12.0
	20'	0.246	0.246	0.255	0.249	498					

Calculating the Mycelial Weight			
Date of experiment: 31/07/08b	T454	T110	A20C
Weight of filter	0.1145	0.1217	0.1326
Dry weight of mycelium	0.0913	0.0926	0.0922
Difference	0.0232	0.0291	0.0404
Difference in μg	23.2	29.1	40.4
Date of experiment: 01/08/08	T454	T110	A20C
Weight of filter	0.1178	0.1266	0.1337
Dry weight of mycelium	0.0911	0.0919	0.0908
Difference	0.0267	0.0347	0.0429
Difference in μg	26.7	34.7	42.9
Date of experiment: 21/08/08	T454	T110	A20C
Weight of filter	0.1254	0.1182	0.12
Dry weight of mycelium	0.0926	0.091	0.09
Difference	0.0328	0.0272	0.02
Difference in μg	32.8	27.2	23.7

A20C uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average A20C uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
10.8	12.0	11.7	11.5 ±0.62	16.8	16.4	16.1	16.4 ±0.35	0.0	-0.4	0.1	0.2 ±0.26
Comparison with the respective wild type (T454) result (%)											
64.3	73.2	72.7	Average: 70.0%								
Growth test^a											
A20C			T454				T110				
+++			+++				-				

This experiment was done four times and the result above was carried out using three set of data. The set of data highlighted was ignored. The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth

Construct A²⁰G Transformant 3751 Colony 1 Controls: T454 (wild type) positive and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
01/08/08C											
T454	0	0.420	0.414	0.409	0.414	691	253	12.64	50.5	1.01	12.5
	20'	0.276	0.250	0.262	0.263	438					
T110	0	0.391	0.399	0.412	0.401	668	-3	-0.17	37.7	0.754	-0.2
	20'	0.392	0.411	0.405	0.403	671					
A20G	0	0.415	0.402	0.398	0.405	675	52	2.61	39.2	0.784	3.3
	20'	0.372	0.377	0.372	0.374	623					
01/08/08C											
T454	0	0.420	0.414	0.409	0.414	691	253	12.64	50.5	1.01	12.5
	20'	0.276	0.250	0.262	0.263	438					
T110	0	0.391	0.399	0.412	0.401	668	-3	-0.17	37.7	0.754	-0.2
	20'	0.392	0.411	0.405	0.403	671					
A20G	0	0.454	0.452	0.450	0.452	753	56	2.81	33.2	0.664	4.2
	20'	0.421	0.419	0.415	0.418	697					
02/08/08											
T454	0	0.398	0.397	0.387	0.394	657	221	11.06	40.5	0.81	13.6
	20'	0.256	0.268	0.260	0.261	436					
T110	0	0.407	0.377	0.406	0.397	661	-2	-0.08	37.2	0.744	-0.1
	20'	0.401	0.392	0.400	0.398	663					
A20G	0	0.464	0.472	0.477	0.471	785	48	2.42	34	0.68	3.6
	20'	0.446	0.445	0.435	0.442	737					

Calculating the Mycelial Weight			
Date of experiment: 01/08/08C	T454	T110	A20G
Weight of filter	0.1409	0.1289	0.1316
Dry weight of mycelium	0.0904	0.0912	0.0924
Difference	0.0505	0.0377	0.0392
Difference in μg	50.5	37.7	39.2
Date of experiment: 01/08/08C	T454	T110	A20G
Weight of filter	0.1409	0.1289	0.1248
Dry weight of mycelium	0.0904	0.0912	0.0916
Difference	0.0505	0.0377	0.0332
Difference in μg	50.5	37.7	33.2
Date of experiment: 02/08/08	T454	T110	A20G
Weight of filter	0.1309	0.1278	0.1249
Dry weight of mycelium	0.0904	0.0906	0.0909
Difference	0.0405	0.0372	0.034
Difference in μg	40.5	37.2	34

A20G uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average A20G uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
3.3	4.2	3.6	3.7 ±0.46	12.5	12.5	13.6	13.05 ±0.64	-0.2	-0.2	-0.1	-0.15 ±0.06
Comparison with the respective wild type (T454) result (%)											
26.4	33.6	26.5	Average: 28.8%								
Growth test^a											
A20G			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct A²⁰G Transformant 3751 Colony 1 Controls: T454 (wild type) positive and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
01/08/08C											
T454	0	0.420	0.414	0.409	0.414	691	253	12.64	50.5	1.01	12.5
	20'	0.276	0.250	0.262	0.263	438					
T110	0	0.391	0.399	0.412	0.401	668	-3	-0.17	37.7	0.754	-0.2
	20'	0.392	0.411	0.405	0.403	671					
A20G	0	0.415	0.402	0.398	0.405	675	52	2.61	39.2	0.784	3.3
	20'	0.372	0.377	0.372	0.374	623					
01/08/08C											
T454	0	0.420	0.414	0.409	0.414	691	253	12.64	50.5	1.01	12.5
	20'	0.276	0.250	0.262	0.263	438					
T110	0	0.391	0.399	0.412	0.401	668	-3	-0.17	37.7	0.754	-0.2
	20'	0.392	0.411	0.405	0.403	671					
A20G	0	0.454	0.452	0.450	0.452	753	56	2.81	33.2	0.664	4.2
	20'	0.421	0.419	0.415	0.418	697					
02/08/08											
T454	0	0.398	0.397	0.387	0.394	657	221	11.06	40.5	0.81	13.6
	20'	0.256	0.268	0.260	0.261	436					
T110	0	0.407	0.377	0.406	0.397	661	-2	-0.08	37.2	0.744	-0.1
	20'	0.401	0.392	0.400	0.398	663					
A20G	0	0.464	0.472	0.477	0.471	785	48	2.42	34	0.68	3.6
	20'	0.446	0.445	0.435	0.442	737					

Calculating the Mycelial Weight			
Date of experiment: 01/08/08C	T454	T110	A20G
Weight of filter	0.1409	0.1289	0.1316
Dry weight of mycelium	0.0904	0.0912	0.0924
Difference	0.0505	0.0377	0.0392
Difference in μg	50.5	37.7	39.2
Date of experiment: 01/08/08C	T454	T110	A20G
Weight of filter	0.1409	0.1289	0.1248
Dry weight of mycelium	0.0904	0.0912	0.0916
Difference	0.0505	0.0377	0.0332
Difference in μg	50.5	37.7	33.2
Date of experiment: 02/08/08	T454	T110	A20G
Weight of filter	0.1309	0.1278	0.1249
Dry weight of mycelium	0.0904	0.0906	0.0909
Difference	0.0405	0.0372	0.034
Difference in μg	40.5	37.2	34

A20G uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average A20G uptake (nmol min⁻¹ mg⁻¹ DW)	T454 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T454 uptake (nmol min⁻¹ mg⁻¹ DW)	T110 uptake in triplicates (nmol min⁻¹ mg⁻¹ DW)			Average T110 uptake (nmol min⁻¹ mg⁻¹ DW)
3.3	4.2	3.6	3.7 ±0.46	12.5	12.5	13.6	13.05 ±0.64	-0.2	-0.2	-0.1	-0.15 ±0.06
Comparison with the respective wild type (T454) result (%)											
26.4	33.6	26.5	Average: 28.8%								
Growth test^a											
A20G			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.

Construct G³²⁸A Transformant 356 Colony 1 Controls: T454 (wild type) positive and T110 negative

Date of Experiment	Sampling Time (min)	Absorbance Readings (OD 204)			Average Absorbance Readings	NaNO ₃ (nmol)	delta	delta/min	µg	µg/ml	nmol/min ⁻¹ /mg ⁻¹ /DW
13/06/07B											
T454	0	0.371	0.373	0.371	0.372	743	233	11.67	45	0.9	13.0
	20'	0.250	0.260	0.255	0.255	510					
T110	0	0.366	0.352	0.358	0.359	717	0	0.00	46.9	0.938	0.0
	20'	0.353	0.365	0.358	0.359	717					
G328A	0	0.376	0.372	0.368	0.372	744	85	4.23	35.1	0.702	6.0
	20'	0.327	0.337	0.325	0.330	659					
13/06/07C											
T454	0	0.405	0.409	0.400	0.405	809	264	13.20	47.3	0.946	14.0
	20'	0.279	0.261	0.278	0.273	545					
T110	0	0.368	0.368	0.368	0.368	736	-19	-0.93	49.5	0.99	-0.9
	20'	0.367	0.375	0.390	0.377	755					
G328A	0	0.366	0.356	0.370	0.364	728	93	4.63	44.3	0.886	5.2
	20'	0.319	0.318	0.316	0.318	635					
08/02/08C											
T454	0	0.364	0.364	0.368	0.365	731	137	6.83	28.2	0.564	12.1
	20'	0.296	0.296	0.299	0.297	594					
T110	0	0.351	0.350	0.349	0.350	700	-5	-0.27	27.9	0.558	-0.5
	20'	0.344	0.355	0.359	0.353	705					
G328A	0	0.398	0.396	0.403	0.399	798	42	2.10	17.1	0.342	6.1
	20'	0.376	0.377	0.381	0.378	756					

Table containing the raw data obtained on three independent net nitrate uptake assays

Calculating the Mycelial Weight			
Date of experiment: 13/06/07B	T454	T110	G328A
Weight of filter	0.1971	0.228	0.214
Dry weight of mycelium	0.1521	0.1811	0.179
Difference	0.045	0.0469	0.035
Difference in μg	45	46.9	35.1
Date of experiment: 13/06/07C	T454	T110	G328A
Weight of filter	0.194	0.2021	0.219
Dry weight of mycelium	0.146	0.1526	0.1747
Difference	0.047	0.0495	0.0443
Difference in μg	47.3	49.5	44.3
Date of experiment: 08/02/08C	T454	T110	G328A
Weight of filter	0.1196	0.1204	0.1091
Dry weight of mycelium	0.0914	0.0925	0.092
Difference	0.0282	0.0279	0.0171
Difference in μg	28.2	27.9	17.1

Table containing the raw data obtained on three independent net nitrate uptake assay

G328A uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average G328A uptake (nmol min ⁻¹ mg ⁻¹ DW)	T454 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T454 uptake (nmol min ⁻¹ mg ⁻¹ DW)	T110 uptake in triplicates (nmol min ⁻¹ mg ⁻¹ DW)			Average T110 uptake (nmol min ⁻¹ mg ⁻¹ DW)
6.0	5.2	6.1	5.8 ±0.5	13.0	14.0	12.1	13.0 ±0.92	0.0	-0.9	-0.5	-0.5 ±0.47
Comparison with the respective wild type (T454) result (%)											
47.0	37.0	51.0	Average: 47.0%								
Growth test^a											
G328A			T454				T110				
+++			+++				-				

The phenotypical growth test ^a was carried out on minimal medium, supplemented with 100mM NaNO₃ and vitamins at 37°C. A score of – represents no growth, + represents poor growth, ++ represents moderate growth and +++ represents wild type growth.