



## Data in Brief

## Gene expression profiling of rice seedlings in response to glutamine treatment



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## ABSTRACT

Glutamine, the most abundant free amino acid in humans (Curi et al., 2007 [1]), has many functions. In addition to protein, amino acid, and nucleic acid biosynthesis, glutamine also regulates the expression of genes related to metabolism, cell defense, and signal transduction in humans (Curi et al., 2007 [1]; Brasse-Lagnel et al., 2009 [2]). Glutamine is also one of the major forms of nitrogen in rice (Fukumorita and Chino, 1982 [3]). In addition to metabolic and nutritional effects, glutamine may function as a signaling molecule to regulate gene expression in plants. To this end, we used microarray analysis to identify genes that are rapidly induced by 2.5 mM glutamine in rice roots. The results revealed that glutamine induced the expression of at least 35 genes involved in metabolism, transport, signal transduction, and stress responses within 30 min (Kan et al., 2015 [4]). Here, we provide the details of the experimental procedure associated with our microarray data deposited in NCBI's Gene Expression Omnibus (GEO ID: GSE56770).

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Specifications	
Organism/cell line/tissue	<i>Oryza sativa</i> L. ssp. japonica cv. TNG67/root tissue
Sex	N/A
Sequencer or array type	Affymetrix GeneChip Rice Genome Array
Data format	Raw data (CEL files)
Experimental factors	Glutamine (2.5 mM, 30 min) vs. no nitrogen control
Experimental features	Gene expression profiling to identify transcripts that are rapidly regulated by glutamine
Consent	N/A
Sample source location	Taipei, Taiwan

## 1. Direct link to deposited data

The deposited data can be found at: <http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE56770>.

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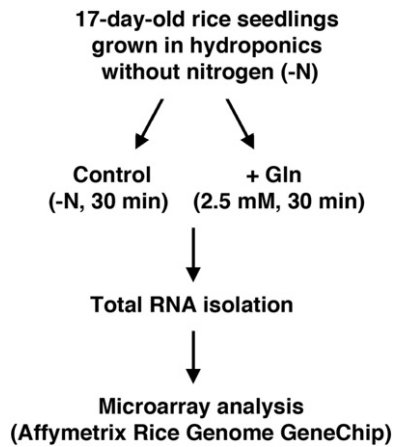
## 2. Experimental design, materials and methods

## 2.1. Experimental design

To identify genes that are rapidly regulated by glutamine, 17-day-old rice seedlings grown in hydroponic solution [5] without nitrogen (–N) were treated with 2.5 mM glutamine for 30 min (+Gln). Total RNAs isolated from –N and +Gln rice roots were used to perform gene expression profiling with the Affymetrix GeneChip Rice Genome Array (Fig. 1).

## 2.2. Plant material and growth conditions

Rice (*Oryza sativa* L. ssp. japonica cv. TNG67) seeds were germinated on wet filter paper in darkness at 30 °C for 3 days and then transferred to –N hydroponic solution, and grown under controlled conditions (30 °C, 12-h photoperiod, 200 μmol photons m<sup>-2</sup> s<sup>-1</sup>, 70% relative humidity) for 2 weeks. The hydroponic solution was renewed every 3 days. 17-Day-old rice seedlings grown in –N hydroponic solution were transferred to fresh –N solution or –N solution supplemented with 2.5 mM glutamine (+Gln) for 30 min. Roots and shoots were harvested separately for total RNA isolation using phenol extraction as previously described [6]. We used 2.5 mM glutamine, a concentration comparable to the concentration of inorganic nitrogen (1.43 mM NO<sub>3</sub><sup>-</sup> and 1.43 mM NH<sub>4</sub><sup>+</sup>) commonly used in hydroponic solution [5].



**Fig. 1.** Schematic diagram of the steps involved in this study. Roots and shoots were harvested separately for total RNA isolation. Two independent sets of biological samples were used for the microarray analysis using Affymetrix Rice Genome GeneChip.

### 2.3. Microarray analysis

For microarray analysis, RNA samples from two biological repeats were sent to the Affymetrix Gene Expression Service Lab at Academia Sinica, Taipei, Taiwan (<http://ipmb.sinica.edu.tw/affy/>) for target preparation, and hybridization to the GeneChip Rice Genome Array (Affymetrix, Santa Clara, CA, USA). The standard washes and double-staining were performed on the Affymetrix GeneChip Fluidics Station 450, and the arrays were scanned on the Affymetrix GeneChip Scanner 3000. The scanned arrays were analyzed by Affymetrix GCOS version 1.4 and the raw data were saved as CEL files. The GeneSpring GX 11.5 software was used to analyze the microarray data. Unpaired *t*-tests were performed to examine the reproducibility of the data. A two-fold cutoff and a *P*-value less than 0.05 were applied to select for up- and down-regulated genes after 2.5 mM Gln treatment for 30 min. In roots, 41 genes, including 39 up- and 2 down-regulated by glutamine, were identified in the microarray analysis. However, we were unable to verify the up-regulation of 4 genes and the down-regulation of 2 genes by reverse

transcription (RT)-PCR. Thus, we concluded that glutamine rapidly up-regulated the expression of at least 35 genes in rice roots [4]. We were unable to identify any genes that were up- or down-regulated (2-fold cutoff) by 2.5 mM glutamine treatment for 30 min in rice shoots under hydroponic conditions by microarray and RT-PCR analyses.

### 3. Discussion

This study provides an evidence to show that glutamine can rapidly induce the expression of at least 35 genes in rice roots. The functions of these genes are related to metabolism, transport, signal transduction, and stress responses [4]. Interestingly, 10 of the 35 early glutamine responsive genes encode putative transcription factors involved in the regulation of nitrogen metabolism and stress responses. These results suggest that glutamine may efficiently amplify its signal and interact with the other signal transduction pathways to regulate plant growth and stress responses.

### Acknowledgments

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