



Data in Brief

Microarray data analyses of yeast RNA Pol I subunit RPA12 deletion strain



Kamlesh Kumar Yadav, Ram Rajasekharan *

Lipidomic Center, Department of Lipid Science, CSIR-Central Food Technological Research Institute (CFTRI), Mysore 570020, Karnataka, India

ARTICLE INFO

Article history:

Received 16 April 2016

Accepted 16 April 2016

Available online 19 April 2016

Keywords:

Ribosomes
rRNA biosynthesis
RNA Pol I
Storage lipids
Triacylglycerol

ABSTRACT

The ribosomal RNA (rRNA) biosynthesis is the most energy consuming process in all living cells and the majority of total transcription activity is dedicated for synthesizing rRNA. The cells may adjust the synthesis of rRNA with the availability of resources. rRNA is mainly synthesized by RNA polymerase I that is composed of 14 subunits. Deletion of RPA12, 14, 39 and 49 are viable. RPA12 is a very small protein (13.6 kDa), and the amount of protein in the cells is very high (12,000 molecules per cell), but the role of this protein is unknown in other cellular metabolic processes (Kulak et al., 2014 [1]). RPA12 consists of two zinc-binding domains and it is required for the termination of rRNA synthesis (Mullem et al., 2002 [2]). Deletions of RPA12 in *Saccharomyces cerevisiae* and *Schizosaccharomyces pombe* cause a conditional growth defect (Nogi et al., 1993 [3]). In *S. pombe*, C-terminal deletion behaves like wild-type (Imazawa et al., 2001 [4]). This prompted us to investigate in detail the physiological role of RPA12 in *S. cerevisiae*, we performed the microarray of *rpa12Δ* strain and deposited into Gene Expression Omnibus under GSE68731. The analysis of microarray data revealed that the expression of major cellular metabolism genes is high. The amino acid biosynthesis, nonpolar lipid biosynthesis and glucose metabolic genes are highly expressed. The analyses also revealed that the *rpa12Δ* cells have an uncontrolled synthesis of cell metabolites, so RPA12 could be a master regulator for whole cellular metabolism.

© 2016 The Authors. Published by Elsevier Inc. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Specifications

Organism/cell line/tissue	<i>Saccharomyces cerevisiae</i> Wild-type (BY4741) and <i>rpa12Δ</i> strain
Sequencer or array type	Affymetrix Yeast Genome 2.0 Array
Data format	Raw data (CEL file)
Experimental factors	Wild-type Vs <i>rpa12Δ</i> cells
Experimental features	Identification of genes that are regulated through RPA12
Consent	Publicly available from NCBI GEO
Sample source location	Yeast deletion strains are maintained in CSIR-CFTRI, Mysore-570020, Karnataka

1. Direct link to deposited data

The data are available at the GEO database under: <http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE68731>.

* Corresponding author.

E-mail address: ram@cftri.com (R. Rajasekharan).

2. Experimental design, materials and methods

2.1. Experimental design

Wild-type and *rpa12Δ* cells were grown to stationary phase in a synthetic complete medium. Stationary phase cells were used for RNA extraction and hybridization on Affymetric microarrays. We used microarrays to study the effect of RPA12 deletion on the cellular metabolism of yeast and identified that distinct class of genes were up-regulated in *rpa12Δ* strain.

2.2. Materials

- Yeast wild-type and deletion strains: Euroscarf
- TRIzol: Invitrogen, Cat. No. 10-296-028
- Synthetic complete media
- Yeast nitrogenous base: Difco
- Yeast drop-out: Sigma-Aldrich

2.3. Sample preparation

Wild-type and *rpa12Δ* cells were first inoculated in yeast extract, peptone and 2% dextrose (YPD) medium. Stationary phase cells were subcultured in a synthetic complete (SC) medium containing 2% dextrose as a carbon source along with kanamycin (50 µg/ml) at 30 °C [5].

After 24 h, the cells were pelleted and washed with phosphate-buffered saline to remove the remaining medium. Total RNA was isolated from both the samples using TRIzol. These samples were hybridized to the Affymetrix Yeast Genome 2.0 Array according to the manufacturer's instructions.

2.4. Statistical analysis of the microarray data

All the original microarray data or raw data (CEL file) were first normalized using the Robust Multiarray Average (RMA) method [6] that consisted of three steps: a background adjustment, quantile normalization and finally summarization. All above procedures were done by RMA algorithm in Gene SpringGx11.5 software from Agilent technologies. The genes of low intensity information content in each data set were filtered as follows: first, the probes of intensities <20.0 percentile in the raw data were excluded and then the probes whose intensities' coefficient of variation (CV) <50.0% at least 1 out of 4 types remained. In differential gene expression (DGE) analyses, we identified many genes involved in cellular metabolism are differentially expressed in *rpa12Δ* strain. Normalized data were filtered for probe sets between 20 and 100 percentile. Fold change (FC) analysis was performed in Gene Spring 11.0 using the threshold $FC \geq 1.0$ and $FC \geq 2.0$. Fold change ≥ 2.0 was selected because the number of gene lists were large in $FC \geq 1.0$.

3. Results

To identify the RPA12-regulated genes, the expression profiles of *rpa12Δ* strain with wild-type (BY4741) were compared. On the basis

of microarray data analyses, we observed that there are significant changes in the gene expression profile in *rpa12Δ* as compared to wild-type. Expression of amino acid, lipid and carbohydrate metabolism genes is also high. The upregulation of all metabolic genes in *rpa12Δ* strain suggested that RPA12 could be a master regulator of whole cellular metabolism.

Acknowledgments

This work was supported by Council of Scientific and Industrial Research (CSIR), New Delhi under the 12th five year plan project LIPIC.

References

- [1] N.A. Kulak, G. Pichler, I. Paron, N. Nagaraj, M. Mann, Minimal, encapsulated proteomic-sample processing applied to copy-number estimation in eukaryotic cells. *Nat. Methods* 11 (2014) 319–324.
- [2] V.V. Mullem, E. Landrieux, J. Vandenhoute, P. Thuriaux, Rpa12p, a conserved RNA polymerase I subunit with two functional domains. *Mol. Microbiol.* 43 (2002) 1105–1113.
- [3] Y. Nogi, R. Yano, J. Dodo, C. Carles, M. Nomura, Gene RRN4 in *Saccharomyces cerevisiae* encodes the A12.2 subunit of RNA polymerase I and is essential only at high temperatures. *Mol. Cell. Biochem.* 13 (1993) 114–122.
- [4] Y. Imazawa, K. Imai, Y. Yao, K. Yamamoto, K. Hisatake, M. Muramatsue, Y. Nogi, Isolation and characterization of the fission yeast gene *Spipa12+* reveals that the conserved C-terminal zinc-finger region is dispensable for the function of its product. *Mol. Gen. Genet.* 264 (2001) 852–859.
- [5] M.D. Rose, F. Winston, P. Heiter, *Methods in Yeast Genetics: A Laboratory Course Manual*. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 1990.
- [6] Y. Benjamini, Y. Hochberg, Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J. R. Stat. Soc.* 57 (1995) 289–300.