Curr Genet (2017) 63:65–68
DOI 10.1007/s00294-016-0621-3

REVIEW

Multiple roles of the general regulatory factor Abf1 in yeast ribosome biogenesis

Beatrice Fermi[1] · **Maria Cristina Bosio**[1,2] · **Giorgio Dieci**[1]

Received: 24 May 2016 / Revised: 27 May 2016 / Accepted: 28 May 2016 / Published online: 4 June 2016

Abstract In *Saccharomyces cerevisiae*, the large majority of the genes coding for cytoplasmic ribosomal proteins (RPs) depend on the general regulatory factor Rap1 for their transcription, but a small cohort of them relies on Abf1 regulatory activity. A recent study showed that unlike Rap1, whose association with RP gene promoters is not affected by environmental changes causing RP gene repression/reactivation, Abf1 association with both RP gene and ribosome biogenesis (Ribi) gene promoters dynamically responds to changes in growth conditions. This observation changes the paradigm of general regulatory factors as relatively static DNA-binding proteins constitutively bound to highly active promoters, and point to Abf1, which binds hundreds of non-RPG promoters within the yeast genome, as a possible key regulatory switch in nutrient- and stress-dependent transcriptional modulation. Moreover, the frequent presence of Abf1 binding sites in the promoters of mitochondrial RP genes evokes the possibility that Abf1 might orchestrate still unexplored levels of co-regulation involving growth-related gene networks in yeast cells.

Keywords Abf1 · Ribosomal protein · *Saccharomyces cerevisiae* · Ribosome biogenesis

Communicated by M. Kupiec.

✉ Giorgio Dieci
giorgio.dieci@unipr.it

[1] Dipartimento di Bioscienze, Università degli Studi di Parma, Parco Area delle Scienze 23/A, 43124 Parma, Italy

[2] Dipartimento di Bioscienze, Università degli Studi di Milano, Via Celoria 26, 20133 Milan, Italy

The *Saccharomyces cerevisiae* genome is interspersed with binding sites for relatively abundant DNA-binding proteins collectively known as general regulatory factors (GRF) because of their involvement as global regulators in diverse chromosomal functions. GRFs include Rap1, Reb1, Tbf1 and Abf1 proteins, all containing Myb-related motifs capable of sequence-specific DNA binding (Azad and Tomar 2016; Brigati et al. 1993; Chasman et al. 1990; Ju et al. 1990; Shore 1994). GRF-binding sites are scattered throughout the genome, with a higher frequency within promoter regions, where the presence of bound GRFs influences promoter activity, but also within telomeric and subtelomeric regions, where GRFs contribute to telomere structure and regulation (Fourel et al. 1999; Grunstein 1997), and at replication origins whose function can be influenced by bound GRFs (Raychaudhuri et al. 1997). It is thought that a common role shared by these DNA-binding proteins at different chromosome locations might be the establishment and maintenance of well-defined chromatin structures (Ganapathi et al. 2011; Hartley and Madhani 2009).

At promoter regions, GRFs may additionally act by recruiting the basal transcription machinery (Papai et al. 2010). Interestingly, promoter regions marked by GRFs generally belong to heavily transcribed genes, many of which are involved in growth-related functions such as ribosome biogenesis (Bosio et al. 2011). Indeed, it has long been known that in *S. cerevisiae* most of the 138 ribosomal protein (RP) genes contain Rap1-binding sites in their promoter regions, with the exception of a small subset of RP genes for which Rap1 is replaced by Abf1 (Lascaris et al. 1999). These two GRFs appear to be interchangeable in this case, as they both act as constitutive transcriptional activators (Mager and Planta 1990).

Given the high number of binding sites located across the genome and the ability of GRFs to influence nucleosome occupancy/chromatin structure, GRFs have the potential to act as master regulators of genome expression in response to nutritional cues or other environmental perturbations. Suggestively with respect to this possibility, the four GRFs mentioned above have all been reported to be phosphoproteins (Albuquerque et al. 2008; Francesconi and Eisenberg 1991; Morrow et al. 1990). In particular, the balance between Abf1 hyper- and hypo-phosphorylated forms was reported to vary in response to nutrient availability (Silve et al. 1992), and Abf1 was included among candidate proximal targets of TORC1 by a recent phosphoproteomic study (Oliveira et al. 2015). However, until recently no evidence was reported for regulated changes in GRF association with cognate promoter elements, nor for any other functional alteration in their activity at promoters, in response to environmental stimuli. This consolidated the notion of GRFs as permanent "placeholders" constitutively acting at promoter regions by counteracting nucleosome deposition and, thus, favoring regulatory transactions due to other, more tunable transcription factors (Bhattacharya and Warner 2008).

Recently, a study by our laboratory challenged this notion by showing that the association of Abf1 with at least some of its target promoters is responsive to nutritional status (Fermi et al. 2016). In particular, we found that inhibition of the nitrogen-sensitive TORC1 pathway by rapamycin, a perturbation known to cause quick repression of ribosomal protein (RP) gene transcription, unexpectedly entails a large increase in Abf1 association with the small cohort of RP genes whose promoter is demarcated by Abf1 instead of Rap1 binding sites. Even though the dependence of this phenomenon on changes of Abf1 phosphorylation state is unknown at present, this observation points to Abf1 as the first GRF whose association to DNA is modulated by nutritional status, and, at the same time, raises at least three orders of questions: (a) why should a repressive stimulus (TORC1 inactivation) entail an increased recruitment of an activator to transcriptionally repressed promoters? (b) what is the physiological reason (if any) for the existence in *S. cerevisiae* of two distinct subsets of RP gene promoters, one characterized by nutrient-unaffected Rap1 binding, the other by nutrient-dependent fluctuations in Abf1 binding? (c) considering other genes bound by Abf1 in their promoter regions, how does Abf1 behave at these promoters in response to nutritional stress, and which are the possible regulatory interconnections between the different categories of Abf1-demarcated genes?

As to the first question, we provided evidence that increased Abf1 association with RP gene promoters observed in response to TOR pathway inactivation might help the transcriptional rescue of these repressed promoters, once more favorable nutritional conditions are established (Fermi et al. 2016). But quick changes in promoter occupancy by Abf1, which were also observed, albeit to lower extents, at ribosome-unrelated promoters in response to TORC1 inhibition (Fermi et al. 2016), might also be related to extra-transcriptional roles of Abf1 (Reed et al. 1999; Yu et al. 2009).

The second and third questions are related to each other, because the different behaviors of Abf1 at its target promoters [which have already been noted in association with different chromatin organization propensities (Paul et al. 2015)] might deal with still unexplored levels of co-regulation between different groups of Abf1-demarcated genes. An intriguing possibility is that the Abf1-dependent subset of RP genes shares some specific regulatory features with the genes required for ribosome biogenesis belonging to the so-called Ribi regulon. According to such hypothesis, the promoters of these genes are enriched in Abf1-binding sites (Yarragudi et al. 2007; Bosio et al. 2016), and their association with Abf1 increases in response to TORC1 inactivation exactly as it does in the case of Abf1-dependent RP genes (Fermi et al. 2016).

In a systematic search for other *S. cerevisiae* gene regulons whose promoter regions tend to be enriched in Abf1 binding sites, we noticed that such *cis*-acting elements appear to be overrepresented also in the promoter regions of the nuclear genes coding for mitochondrial ribosomal proteins (MRPs). As shown in Table 1, among the 74 *S. cerevisiae* genes coding for MRPs (according to the Saccharomyces Genome Database), those displaying in the promoter region one or more Abf1-binding sites are 24, 13 of which have been shown to be bound by Abf1 in vivo and/or to be transcriptionally dependent on Abf1 (Chang et al. 2011; Schlecht et al. 2008; Yarragudi et al. 2007). Interestingly, 15 more mitochondrial ribosomal protein (MRP) gene promoters contained one or more Reb1-binding sites, and 3 more MRP promoters displayed one or more Rap1-binding sites (Chang et al. 2011). Therefore, the majority of MRP promoters are demarcated by GRFs, in particular by Abf1. In light of these observations, it is tempting to speculate that Abf1 might orchestrate a network of subtle regulatory interconnections between genes involved in the biogenesis and structure of both cytoplasmic and mitochondrial ribosomes. With this respect, it is worth noting that Abf1 has been suggested by a previous study to be involved in the intergenomic signaling pathway of mitochondrial–nuclear communication (Woo et al. 2009).

Given that there are several hundreds of Abf1 target promoters in the budding yeast genome, with many of them regulating growth-related genes, this transcription factor might represent one of the key regulators of genome expression reprogramming which allows for rapidly alternating growth arrest and growth burst phases in response to

Table 1 Mitochondrial ribosomal protein (MRP) genes displaying one or more Abf1-binding sites in their promoter region according to YPA

Systematic name	Common name	Mitochondrial ribosomal subunit	Abf1 binding site (YPA)[a]	Abf1 association (ChIP)[b]	Abf1 requirement for transcription[c]
YCR071C	IMG2	Large	−105		+
YDR296 W	MHR1	Large	−132	+	+
YDR347 W	MRP1	Small	−96		
YDR405 W	MRP20	Large	−233; −132; −117	+	+
YKL167C	MRP49	Large	−89	+	+
YPL118 W	MRP51	Small	−104		
YNL005C	MRP7	Large	−300		
YBL038 W	MRPL16	Large	−187	+	+
YBR282 W	MRPL27	Large	−90		
YDR462 W	MRPL28	Large	−117; −101	+	
YCR003 W	MRPL32	Large	−81	+	+
YBR122C	MRPL36	Large	−97	+	
YML009C	MRPL39	Large	−95		
YLR439 W	MRPL4	Large	−94		
YMR225C	MRPL44	Large	−94		
YPR100 W	MRPL51	Large	−119		
YJL063C	MRPL8	Large	−15	+	
YPL013C	MRPS16	Small	−198	+	+
YNL306 W	MRPS18	Small	−304; −103	+	+
YBR146 W	MRPS9	Small	−202; −103	+	+
YOR158 W	PET123	Small	−129		
YNR037C	RSM19	Small	−108	+	+
YDR175C	RSM24	Small	−104		
YGR215 W	RSM27	Small	−88		

[a] For each MRP gene, the column reports the position of Abf1-binding sites within the promoter region according to YPA (Chang et al. 2011)

[b] The + symbol indicates that experimental evidence has been obtained for in vivo physical interaction of Abf1 with the corresponding binding site(s) in previous genome-wide location analyses (Harbison et al. 2004; Schlecht et al. 2008)

[c] The + symbol indicates that transcriptional dependence on Abf1 has been experimentally observed by a previous study using an *abf1 ts* mutant (Yarragudi et al. 2007)

changing nutrient availability (Ho and Gasch 2015; Soontorngun 2016).

Acknowledgments We thank Paola Frigeri (University of Parma) for help with MRP promoter analysis. This work was supported by a Grant from the Italian Ministry of Education, University and Research (MIUR, PRIN 2009 to G.D.) and Italian Association for Cancer Research (AIRC).

References

Albuquerque CP, Smolka MB, Payne SH, Bafna V, Eng J, Zhou H (2008) A multidimensional chromatography technology for in-depth phosphoproteome analysis. Mol Cell Proteom: MCP 7:1389–1396

Azad GK, Tomar RS (2016) The multifunctional transcription factor Rap1: a regulator of yeast physiology. Front Biosci 21:918–930

Bhattacharya A, Warner JR (2008) Tbf1 or not Tbf1? Mol Cell 29:537–538

Bosio MC, Fermi B, Spagnoli G, Levati E, Rubbi L, Ferrari R, Pellegrini M, Dieci G et al (2016) Abf1 and other general regulatory factors control ribosome biogenesis gene expression in budding yeast (**Submitted**)

Brigati C, Kurtz S, Balderes D, Vidali G, Shore D (1993) An essential yeast gene encoding a TTAGGG repeat-binding protein. Mol Cell Biol 13:1306–1314

Chang DT, Huang CY, Wu CY, Wu WS (2011) YPA: an integrated repository of promoter features in *Saccharomyces cerevisiae*. Nucl Acids Res 39:D647–D652

Chasman DI, Lue NF, Buchman AR, LaPointe JW, Lorch Y, Kornberg RD (1990) A yeast protein that influences the chromatin structure of UASG and functions as a powerful auxiliary gene activator. Genes Dev 4:503–514

Fermi B, Bosio MC, Dieci G (2016) Promoter architecture and transcriptional regulation of Abf1-dependent ribosomal protein genes in *Saccharomyces cerevisiae*. Nucl Acids Res. doi:10.1093/nar/gkw194

Fourel G, Revardel E, Koering CE, Gilson E (1999) Cohabitation of insulators and silencing elements in yeast subtelomeric regions. EMBO J 18:2522–2537

Francesconi SC, Eisenberg S (1991) The multifunctional protein OBF1 is phosphorylated at serine and threonine residues in *Saccharomyces cerevisiae*. Proc Natl Acad Sci USA 88:4089–4093

Ganapathi M, Palumbo MJ, Ansari SA, He Q, Tsui K, Nislow C, Morse RH (2011) Extensive role of the general regulatory factors, Abf1 and Rap1, in determining genome-wide chromatin structure in budding yeast. Nucl Acids Res 39:2032–2044

Grunstein M (1997) Molecular model for telomeric heterochromatin in yeast. Curr Opin Cell Biol 9:383–387

Harbison CT, Gordon DB, Lee TI, Rinaldi NJ, Macisaac KD, Danford TW, Hannett NM, Tagne JB, Reynolds DB, Yoo J, Jennings EG, Zeitlinger J, Pokholok DK, Kellis M, Rolfe PA, Takusagawa KT, Lander ES, Gifford DK, Fraenkel E, Young RA (2004) Transcriptional regulatory code of a eukaryotic genome. Nature 431:99–104

Hartley PD, Madhani HD (2009) Mechanisms that specify promoter nucleosome location and identity. Cell 137:445–458

Ho YH, Gasch AP (2015) Exploiting the yeast stress activated signaling network to inform on stress biology and disease signaling. Curr Genet 61:503–511

Ju QD, Morrow BE, Warner JR (1990) REB1, a yeast DNA-binding protein with many targets, is essential for growth and bears some resemblance to the oncogene myb. Mol Cell Biol 10:5226–5234

Lascaris RF, Mager WH, Planta RJ (1999) DNA-binding requirements of the yeast protein Rap1p as selected in silico from ribosomal protein gene promoter sequences. Bioinformatics 15:267–277

Mager WH, Planta RJ (1990) Multifunctional DNA-binding proteins mediate concerted transcription activation of yeast ribosomal protein genes. Biochim Biophys Acta 1050:351–355

Morrow BE, Ju Q, Warner JR (1990) Purification and characterization of the yeast rDNA binding protein REB1. J Biol Chem 265:20778–20783

Oliveira AP, Ludwig C, Zampieri M, Weisser H, Aebersold R, Sauer U (2015) Dynamic phosphoproteomics reveals TORC1-dependent regulation of yeast nucleotide and amino acid biosynthesis. Sci Signal 8:rs4

Papai G, Tripathi MK, Ruhlmann C, Layer JH, Weil PA, Schultz P (2010) TFIIA and the transactivator Rap1 cooperate to commit TFIID for transcription initiation. Nature 465:956–960

Paul E, Tirosh I, Lai W, Buck MJ, Palumbo MJ, Morse RH (2015) Chromatin mediation of a transcriptional memory effect in yeast. G3 5:829–838

Raychaudhuri S, Byers R, Upton T, Eisenberg S (1997) Functional analysis of a replication origin from *Saccharomyces cerevisiae*: identification of a new replication enhancer. Nucl Acids Res 25:5057–5064

Reed SH, Akiyama M, Stillman B, Friedberg EC (1999) Yeast autonomously replicating sequence binding factor is involved in nucleotide excision repair. Genes Dev 13:3052–3058

Schlecht U, Erb I, Demougin P, Robine N, Borde V, van Nimwegen E, Nicolas A, Primig M (2008) Genome-wide expression profiling, in vivo DNA binding analysis, and probabilistic motif prediction reveal novel Abf1 target genes during fermentation, respiration, and sporulation in yeast. Mol Biol Cell 19:2193–2207

Shore D (1994) RAP1: a protean regulator in yeast. Trends Genet 10:408–412

Silve S, Rhode PR, Coll B, Campbell J, Poyton RO (1992) ABF1 is a phosphoprotein and plays a role in carbon source control of COX6 transcription in *Saccharomyces cerevisiae*. Mol Cell Biol 12:4197–4208

Soontorngun N (2016) Reprogramming of nonfermentative metabolism by stress responsive transcription factors in the yeast *Saccharomyces cerevisiae*. Curr Genet. doi:10.1007/s00294-016-0609-z

Woo DK, Phang TL, Trawick JD, Poyton RO (2009) Multiple pathways of mitochondrial-nuclear communication in yeast: intergenomic signaling involves ABF1 and affects a different set of genes than retrograde regulation. Biochim Biophys Acta 1789:135–145

Yarragudi A, Parfrey LW, Morse RH (2007) Genome-wide analysis of transcriptional dependence and probable target sites for Abf1 and Rap1 in *Saccharomyces cerevisiae*. Nucl Acids Res 35:193–202

Yu S, Smirnova JB, Friedberg EC, Stillman B, Akiyama M, Owen-Hughes T, Waters R, Reed SH (2009) ABF1-binding sites promote efficient global genome nucleotide excision repair. J Biol Chem 284:966–973