Data Article

Dataset describing the genome wide effects on transcription resulting from alterations in the relative levels of the bZIP transcription factors Atf1 and Pcr1 in *Schizosaccharomyces pombe*

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**Abstract**

*Schizosaccharomyces pombe* has been used as an excellent model for studying eukaryotic cell cycle regulation and stress responses. The bZIP transcription factors Atf1 (ATF2 homolog) and Pcr1 (CREB homolog) have been shown to be important for regulating the expression of genes related to both stress response and cell cycle. Pcr1 has in fact been implicated as a determining factor in the segregation of the cell cycle and stress response related functions of Atf1. Interestingly Atf1 and Pcr1 levels are known to vary during the cell cycle thus giving rise to the possibility that their relative levels can influence the periodic transcriptional program of the cell. Here we report our observations on the changes in transcriptome of *S. pombe* cells which have been genetically manipulated to create relative differences in the levels of Atf1 and Pcr1. These results highlight new information regarding the potential role of Atf1 and Pcr1 in orchestrating the integration of the transcriptional programs of cell cycle and stress response.

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Specifications Table

| Subject                  | Type of data       | How the data were acquired                                                                 |
|--------------------------|--------------------|---------------------------------------------------------------------------------------------|
| Specific subject area    |                    | Data was acquired using Next Generation Sequencing                                           |
| Biology                  | Table              | TruSeq stranded mRNA preparation protocol was used to capture RNA, then the mRNA was purified and the cDNA library was prepared. The RNA sequence data were generated as a Fastq file. The quality of the data was checked. Read mapping to the reference genome was done using Cuffdiff. Gene ontology annotations were assigned using Uniprot. The data analysis report was created. |
| Molecular biology        | Venn diagram       |                                               |
| Table                    | Graph              |                                               |

Value of the Data

- The data reflects the gene expression landscape of S. pombe strains with altered levels of Atf1 and Pcr1, which are homologs of mammalian ATF2 and CREB, thus expanding our knowledge about individual functional roles of these two transcription factors in a living cell. Deregulation of both ATF2 and CREB is associated with multiple developmental disorders and tumorigenesis. Clear understanding of the interplay between these two transcription factors and its effect on the cell's transcription program is therefore very important.
- The analysis of the data presented in this report identifies genes whose expression can be regulated by Pcr1 independently of Atf1. This is an important information as in earlier reports Pcr1 functions have been mostly characterized in the context of promoter specificity of Atf1.
- Analysis of this dataset clearly shows the control exerted by Pcr1 on the expression of genes important for many important fundamental biological processes like stress response and cell cycle.
- These data provide an entry point into investigations aimed at understanding how balance of the two transcription factors Atf1 and Pcr1 can regulate cell fate and proliferation. Extrapolation of these data can also facilitate studies aimed at understanding the contribution of ATF2 and CREB in disease progression.
1. Data Description

Studies done in our lab have established Pcr1 to be important in combating stress responses and to have contrasting outcomes on cell cycle progression [1]. In this study, we used genetic manipulations to vary the relative levels of Atf1 and Pcr1 in S. pombe cells. To study the effects of increase in Pcr1 levels, it was overexpressed in wt and \( \Delta atf1 \) cells and the transcriptional profiles of these cells were characterised. The effect of decrease in Atf1 levels was studied by comparing the gene expression profile of wt and \( \Delta atf1 \) cells. The effect of complete absence of both these transcription factors was studied by comparing the transcriptomes of wt and \( \Delta atf1 \Delta pcr1 \) cells. The group of genes identified to be induced and repressed in each set of experiments are reported in (Tables 1–8). We performed a comparative analysis between the datasets obtained between different backgrounds, looking for unique genes. We found only 4 genes to be commonly upregulated by Pcr1 overexpression in both wt and \( \Delta atf1 \) cells (Fig. 1A). 8 genes were found to be downregulated only in the double mutant (Fig. 1B). Comparison of

| Table 1 | List of genes upregulated during Pcr1 overexpression in wt S. pombe cells.
|---------|----------------------------------|
| Gene ID | Gene Symbol | Gene Function |
| SPAC21E11.03c | pcr1 | DNA-binding transcription factor Pcr1 |
| SPAC19G12.16c | adg2 | conserved fungal cell surface protein, Kre9/Knh1 family |
| SPAC212.11 | thl1 | RecQ type DNA helicase |
| SPBC1105.05 | exg1 | cell wall glucan 1,6-beta-glucosidase Exg1 |
| SPAPB1E7.04c | SPAPB1E7.04c | chitinase |
| SPORNNA.32 | sno12 | box H/ACA small nuclear RNA 12/snR99 |
| SPBC1348.14c | ght7 | plasma membrane hexose transmembrane transporter Ght7 |
| SPNCRNA.942 | SPNCRNA.942 | intergenic RNA (predicted) |
| SPRAA.02 | rns | small subunit (15S) rRNA, rns |
| SPAC186.09 | pdc102 | pyruvate decarboxylase |
| SPBPB2B2.08 | SPBPB2B2.08 | conserved fungal protein |
| SPNCRNA.532 | SPNCRNA.532 | non-coding RNA (predicted) |
| SPAC108.05 | isp3 | spore wall structural constituent Isp3 |
| SPAC1039.11c | gto1 | alpha-glucosidase |
| SPAC23A11.02c | ted1 | GPI-remodeling mannose-ethanolamine phosphate phosphodiesterase Ted1 |
| SPBPB1C11.01 | ant1 | plasma membrane ammonium transmembrane transporter |
| SPAC20G8.05c | cdc15 | F-BAR domain protein Cdc15 |
| SPCC306.11 | SPCC306.11 | Schizosaccharomyces specific protein, uncharacterized |
| SPAC13G7.04c | mac1 | plasma membrane anchored protein, claudin family, predicted membrane sensor Mac1 |
| SPRAA.46 | SPRAA.46 | 18S ribosomal RNA |
| SPNCRNA.1374 | cta3-antisense-1 | antisense RNA (predicted) |
| SPAPB1E7.05 | gde1 | glycophosphoryl diester phosphodiesterase Gde |
| SPBC11C11.05 | SPBC11C11.05 | conserved fungal cell wall protein, Kre9/Knh1 family |
| SPBC1235.13 | ght6 | plasma membrane glucose/fructose:proton symporter Ght6 |
| SPBC148.01c | cut2 | sister chromatid separation inhibitor, securin |
| SPAC821.09 | eng1 | cell septum surface endo-1,3-beta-glucanase Eng1 |
| SPAC106.08 | etd1 | Spg1-binding protein Etd1 |
| SPBP26C9.03c | fet4 | plasma membrane iron/zinc ion transmembrane transporter |
| SPBC1685.14c | vid27 | WD repeat protein, Vid27 family, conserved in fungi and plants |

| Table 2 | List of genes downregulated during Pcr1 over expression in wt S. pombe cells. |
|---------|----------------------------------|
| Gene ID | Gene Symbol | Gene Function |
| SPAPB8A.10 | ups1 | mitochondrial phosphatidic acid transfer protein Ups1 |
| SPAP27G11.13c | nop10 | box H/ACA snoRNP complex protein |
these data revealed the identity of genes that can be positively regulated by Pcr1 independently of Atf1 (Table 9). The genes found to be regulated independently by Pcr1 were then analyzed to identify the cellular processes associated with the gene expression changes using DAVID [2,3]. DAVID analysis classified the genes to be important in several biological processes (Fig. 1C). The known expression changes of these genes during stress response [4] and cell division [5] was then looked up and the genes were then classified into Stress response and Cell cycle categories. We found that groups of genes are important during the stress response, the cell cycle, or both (Fig. 1D). 28 genes were found to be upregulated only in the Δatf1Δpcr1 when compared to genes upregulated in Δatf1 cells (Fig. 2A). DAVID analysis identified several pathways that are downregulated by Pcr1 (Fig. 2B). These genes were also classified according to their previously known association with cell cycle and stress response (Fig. 2C). Genes that are downregulated by Pcr1 independently of Atf1 are listed in Table 10. We compared the genes regulated by Pcr1 (Tables 9, 10) with those of the existing datasets of Atf1 dependent gene expression from studies previously conducted by us and other groups [4,6]. This comparison reveals that there are a few
| Gene ID     | Gene Symbol | Gene Function                                                                 |
|------------|-------------|-------------------------------------------------------------------------------|
| SPAP8A3.04c | hsp9        | heat shock protein Hsp9                                                       |
| SPAC19A8.16 | prl65       | tudor domain superfamily protein                                              |
| SPBC32F12.03c | gpx1     | H2O2 scavenger glutathione peroxidase Gpx1                                   |
| SPCC1393.12 | zym1        | metallothionein Zym1                                                          |
| SPCC1393.12 | dak2        | dihydroxyacetone kinase Dak2                                                  |
| SPNCRNA.103 | sme2        | meiosis-specific Smp foci IncRNA SME2, long isoform                          |
| SPAC34.12  | rds1        | ferritin related conserved fungal protein                                      |
| SPCC757.03c | hsp3101     | glyoxylase III Hsp3101                                                        |
| SPAC4H3.08  | SPAC4H3.08  | 3-hydroxyacyl-CoA dehydrogenase                                               |
| SPBC359.06 | msh14       | adducin, involved in actin cytoskeleton organization                           |
| SPNCRNA.570 |            | non-coding RNA (predicted)                                                    |
| SPAPB1A11.02 |            | esterase/lipase                                                               |
| SPBPPB2B.06c |            | extracellular 5'-nucleotidase, human NT5E family                             |
| SPAC15E1.02c |            | NADH/NADPH-dependent indole-3-acetaldehyde reductase, implicated in cellular  |
| SPAC19G12.09 |            | detoxification                                                                |
| SPAC4F10.20 | grx1        | glutaredoxin Grx1                                                             |
| SPBC56F2.15 | tam13       | Schizosaccharomyces specific protein, uncharacterized                          |
| SPAC16E5.16c | bsd90      | Lsd90 protein                                                                 |
| SPBC21C3.19 | rct3        | SBDS family protein Rct3                                                       |
| SPBC215.05 | gpd1        | glycerol-3-phosphate dehydrogenase Gpd1                                       |
| SPBC289.14 |            | adducin                                                                       |
| SPAC3G6.07  | SPAC3G6.07  | Schizosaccharomyces specific protein, uncharacterized                          |
| SPABP24D3.10c |            | malteose alpha-glucosidase Agl1                                               |
| SPBPPB21E.07 |            | pseudogene                                                                    |
| SPNCRNA.1255 |           | intergenic RNA (predicted)                                                    |
| SPAC26F1.07 | SPAC26F1.07 | NADPH-dependent aldo-keto reductase                                            |
| SPBC725.10 | tsp0        | mitochondrial outer membrane protein, TspO/MBR-related, implicated in lipid/     |
|             |             | sterol transport, tspO                                                        |
| SPCCPB16A4.07 |            | Stm1/Oga1 family protein Smp4                                                  |
| SPNCrna.1223 |            | antisen RNA (predicted)                                                       |
| SPBCPB16A4.06c |            | Schizosaccharomyces specific protein, uncharacterized                          |
| SPCC33.12   |            | vacuolar proteinase B inhibitor Pbi2                                           |
| SPBC1198.14 | fbp1        | fructose-1,6-bisphosphatase Fbp1                                               |
| SPBC11C11.06c |            | Schizosaccharomyces specific protein, uncharacterized                          |
| SPAC23C4.11 | atp18       | F1-FO ATP synthase subunit J                                                  |
| SPBC713.11c |            | plasma membrane proteolipid Pmp3                                              |
| SPAC29B12.13 | SPAC29B12.13 | CENP-V, S-(hydroxymethyl)glutathione synthase                                 |
| SPCC33.06c  |            | thioredoxin-related chaperone Pmp20                                            |
| SPAC11D3.01c |            | Con-6 family conserved fungal protein                                          |
| SPBC16A3.02c |            | mitochondrial CH-OH/GP oxidoreductase, human RTN4IP1 ortholog, implicated in    |
|             |             | mitochondrial organization or tethering                                        |
| SPAC977.15  | SPAC977.15  | dienelactone hydrolase family, implicated in cellular detoxification           |
| SPCC757.07c | ctt1        | catalase                                                                       |
| SPNCRNA.445 |            | non-coding RNA                                                                 |
| SPAC10F6.06 | vip1        | RNA-binding protein Vip1                                                       |
| SPAC25G10.06 | rps2801     | 40S ribosomal protein S28                                                      |
| SPCC794.01c |            | glucose dehydrogenase Gcd1                                                    |
| SPBC26H8.14c |            | mitochondrial copper chaperone for cytochrome c oxidase Cox17                 |
| SPBC3E7.02c |            | heat shock protein Hsp16                                                       |
| SPBC215.11c |            | aldo/keto reductase, unknown biological role                                   |
| SPAC17D1.17 | tam11       | Schizosaccharomyces specific protein, uncharacterized                          |
| SPNCrna.1436 |            | non-coding RNA                                                                 |
| SPBC272.03  | SPBC272.03  | pyridoxamine 5’-phosphate oxidase                                              |
| SPAC3G9.11c |            | pyruvate decarboxylase                                                         |

(continued on next page)
| Gene ID     | Gene Symbol | Gene Function                                                                 |
|------------|-------------|-------------------------------------------------------------------------------|
| SPBC32H8.07 | git5        | heterotrimeric G protein beta (WD repeat) subunit Git5                        |
| SPAC9E9.04 | SPAC9E9.04  | bcap family homolog, implicated in vesicle-mediated transport                 |
| SPAC15A10.05c | mug182    | NADHX epimerase                                                             |
| SPAC4G9.12 | idn1        | gluconokinase                                                                |
| SPBC23G7.16 | ctr6        | vacuolar copper exporter Ctr6                                                |
| SPBC21B10.04c | nrf1     | vacuolar transporter chaperone (VTC) complex, GTPase regulator subunit Nrf1 |
| SPCC965.06 | osr2        | potassium channel, beta subunit, aldo-keto reductase                         |
| SPNCRNA.906 | snR30      | non-coding RNA                                                               |
| SPAC823.17 | tom6        | mitochondrial TOM complex subunit Tom6                                      |
| SPAC688.16 | SPAC688.16  | human TMEM254 ortholog                                                      |
| SPAC186.05c | gdt1        | Golgi calcium and manganese antipporter Gdt1                                  |
| SPBC660.05 | wwm3        | WW domain containing conserved fungal protein Wwm3                           |
| SPBC2A9.02 | SPBC2A9.02  | NADH-dependent glycolaldehyde-/furfural/butyraldehyde/propylaldehyde reductase |
| SPAC22F8.05 | SPAC22F8.05 | alpha, alpha-trehalose-phosphate synthase                                    |
| SPAC4G8.02c | sss1        | translocon gamma subunit Sss1                                                |
| SPCC794.04c | SPCC794.04c | amino acid transmembrane transporter                                         |
| SPAC26F1.14c | aif1       | mitochondrial inner membrane anchored oxidoreductase                         |
| SPAC4F8.10c | stg1        | SM22/transgelin-like actin modulating protein Stg1                            |
| SPBC30D10.14 | SPBC30D10.14 | dienelactone hydrolase family                                               |
| SPAC27D7.09c | SPAC27D7.09c | But2 family protein, similar to cell surface molecules                      |
| SPBC337.08c | ubi4        | protein modifier, ubiquitin                                                  |
| SPAC1705.02 | SPAC1705.02 | SRF family protein, DUF, human 4F5S homolog, implicated in mRNA splicing     |
| SPBP4H10.12 | SPBP4H10.12 | protein with a role in ER insertion of tail-anchored membrane proteins      |
| SPAC172.07 | qcr8        | ubiquinol-cytochrome-c reductase complex subunit 7                           |
| SPBC23G7.10c | SPBC23G7.10c | NADH-dependent flavin oxidoreductase, implicated in cellular detoxification |
| SPAC3G6.13c | rp1401      | 60S ribosomal protein L41                                                    |
| SPBC800.14c | SPBC800.14c | mitochondrial DUF1772 family protein, multimembrane spanning anthrone oxygenase-like |
| SPCC191.01 | SPCC191.01  | Schizosaccharomyces specific protein, uncharacterized                        |
| SPBC4B4.05 | smg1        | Sm snRNP core protein Smg1                                                   |
| SPAC922.04 | SPAC922.04  | Schizosaccharomyces specific protein, uncharacterized                        |
| SPAPJ891.03 | mic10       | MICOS complex subunit Mic10                                                 |
| SPBC3B9.13c | rpp102      | ribosomal protein P1 Rpp102                                                  |
| SPBC405.04c | ypt7        | GTPase Ypt7                                                                  |
| SPAC2F3.05c | SPAC2F3.05c | xylose and arabinose reductase                                               |
| SPNCRNA.844 | SPNCRNA.844 | intergenic RNA (predicted)                                                   |
| SPAC4D7.02c | ppc1        | phosphatidylglycerol phospholipase C Ppc1                                    |
| SPAC11D3.19 | SPAC11D3.19 | Schizosaccharomyces pombe specific protein                                   |
| SPCC16A11.15c | SPCC16A11.15c | Schizosaccharomyces specific protein, uncharacterized                       |
| SPAC1F8.08 | SPAC1F8.08  | Schizosaccharomyces specific protein, uncharacterized                        |
| SPAP27G11.13c | nop10   | box H/ACA snRNP complex protein                                               |
| SPAC4H3.03c | SPAC4H3.03c | glucan 1,4-alpha-glucosidase                                                  |
| SPAC8G9.07c | arc4        | ARP2/3 actin-organizing complex subunit Arc4                                   |
| SPAC23H3.02c | ini1        | RING finger-like protein Ini1                                                |
| SPAC19B12.06c | rbd4     | rhomboid family protease, unknown biological role, associated with COP1 coated vesicle |
| SPAC26F1.10c | pyp1        | protein tyrosine phosphatase Pyp1                                             |
| SPAC6S0.11 | vps55       | vacuolar sorting protein Vps55                                                |
| SPCC24B10.05 | tim9       | Tim9-Tim10 complex subunit Tim9                                              |
| SPAPB24D3.08c | SPAPB24D3.08c | NADP-dependent oxidoreductase, implicated in cellular detoxification          |
| SPAC6F12.04 | tvp15       | COPI-coated vesicle associated protein                                        |
| SPCC663.02 | wtf14       | wtf element Wtf14                                                             |
Table 5
List of genes upregulated during Pcr1 overexpression in Δatf1 background.

| Gene ID       | Gene Symbol | Gene Function                                                                 |
|---------------|-------------|--------------------------------------------------------------------------------|
| SPBC32F12.03c | gpx1        | H2O2 scavenger glutathione peroxidase Gpx1                                     |
| SPBP2B2.06c  | prl65       | extracellular 5'-nucleotidase, human NT5E family                                |
| SPAC19A8.16  | rpp202     | tudor domain superfamily protein                                                |
| SPBC23G7.15c | sme2        | 60S acidic ribosomal protein P2                                                 |
| SPNCRNA.103  | grx1        | meiosis-specific Smf foci IncRNA Smf2, long isoform                              |
| SPAC4F10.20  | zym1        | glutaredoxin Grx1                                                               |
| SPAC22H10.13 | ght3        | plasma membrane glutonate:proton symporter Ght3                                 |
| SPAC21E1.03c | pcr1        | DNA-binding transcription factor Pcr1                                           |
| SPBC56F2.15  | tam13       | Schizosaccharomyces specific protein Tam13                                       |
| SPCC1393.12  | SPCC1393.12 | Schizosaccharomyces specific protein, uncharacterized                            |
| SPBC11C1.06c | SPBC11C1.06c| Schizosaccharomyces specific protein, uncharacterized                            |
| SPNCRNA.570  | SPNCRNA.570 | non-coding RNA (predicted)                                                      |
| SPCPB16A4.07 | SPCPB16A4.07| Stm1/Oga1 family protein Smp4                                                    |
| SPBPB21E7.08 | pseudogene  | Stm1/Oga1 family protein Smp4                                                    |
| SPAC15E1.02c | SPAC15E1.02c| DUF1761 family protein                                                          |
| SPNCRNA.1436 | SPNCRNA.1436| non-coding RNA                                                                  |
| SPCC338.12   | pbi2        | vacuolar proteasome B inhibitor Pbi2                                             |
| SPAC19G12.09 | SPAC19G12.09| NADH/NADPH-dependent indole-3-acetaldehyde reductase, implicated in cellular detoxification |
| SPNCRNA.942  | SPNCRNA.942 | intergenic RNA (predicted)                                                      |
| SPBC359.06   | mug14       | adducin, involved in actin cytoskeleton organization                            |
| SPNCRNA.98   | srg7        | 7SL signal recognition particle component                                        |
| SPNSNORNA.32 | sno12       | box H/ACA small nuclear RNA 12/snR99                                             |
| SPBC725.10   | tps0        | mitochondrial outer membrane protein, TspO/MBR-related, implicated in lipid/sterol transport, tspO |
| SPAC26F1.07  | SPAC26F1.07 | NADPH-dependent aldo-keto reductase                                              |
| SPAC23C4.11  | atp18       | F1-FO ATP synthase subunit J                                                    |
| SPAC9E9.04   | SPAC9E9.04  | bcap family homolog, implicated in vesicle-mediated transport                    |
| SPNCRNA.808  | SPNCRNA.808 | intergenic RNA (predicted)                                                      |
| SPBC26H8.14c | cox17       | mitochondrial copper chaperone for cytochrome c oxidase Cox17                   |
| SPAC1F8.03c  | str3        | plasma membrane heme transmembrane transporter Str3                              |
| SPAC1F8.05   | isp3        | spore wall structural constituent Isp3                                           |
| SPBC21B1.04c | nrf1        | vacuolar transporter chaperone (VTC) complex, GTPase regulator subunit Nrf1     |
| SPBC215.11c  | SPBC215.11c | aldo/keto reductase, unknown biological role                                    |
| SPBC11B10.10c| pht1        | histone H2A variant H2AZ Pht1                                                   |
| SPAC4F8.10c  | stg1        | SM22/transgelin-like actin modulating protein Stg1                               |
| SPAC22F8.05  | SPAC22F8.05 | alpha,alpha-trehalose-phosphate synthase                                         |

genes whose expression is regulated in a contrasting manner by Atf1 and Pcr1 (Tables 11, 12). We compared our gene list obtained from this study with existing data for Atf1-dependent gene expression [4] and found 75 new genes that are upregulated by Atf1 and 34 new genes that are downregulated by it (Fig. 3A, B) in absence of stress.

Genes upregulated and downregulated in each of the experimental backgrounds are mentioned in the tables below.
Table 6
List of genes downregulated during Pcr1 overexpression in Δatf1 background.

| Gene ID        | Gene Symbol | Gene Function                                                                 |
|---------------|-------------|-------------------------------------------------------------------------------|
| SPCC576.01c   | xan1        | alpha-ketoglutarate-dependent xanthine dioxygenase Xan1                       |
| SPCC1223.09   | uro1        | uricase Uro1                                                                  |
| SPAC1002.19   | urg1        | GTP cyclohydrolase II Urg1                                                    |
| SPAC1039.02   | SPAC1039.02 | extracellular 5′-nucleotidase, human NT5E family                              |
| SPCC4B3.10c   | ipk1        | inositol 1,3,4,5,6-pentakisphosphate (IP5) kinase                             |
| SPAC56F8.03   | SPAC56F8.03 | translation initiation factor eIF5B Tif52                                     |
| SPAC29B12.14c | SPAC29B12.14c | plasma membrane purine transmembrane transporter                          |

Fig. 1. Transcriptome analysis reveals targets which are differentially upregulated by Pcr1. Analysis was done using BioVenn [7] to find out the overlaps between different datasets. (A) Overlap between Pcr1-OP in wt and Pcr1-OP in Δatf1 cells showed 36 genes to be upregulated by Pcr1, independent of regulation by Atf1. (B) Upon comparing Δatf1 and Δatf1Δpcr1, we found 8 genes to be uniquely downregulated in the latter, which could be considered as targets induced solely by Pcr1. (C) Genes found to be positively upregulated by Pcr1 independently of Atf1 were sorted into significant functional clusters obtained from DAVID based analysis of genes represented in Table 9. (D) Graph represents the association of the genes positively upregulated by Pcr1 independently of Atf1 with cell cycle and/or stress response or both.
**Table 7**

List of genes upregulated in Δatf1Δpcr1 cells.

| Gene ID       | Gene Symbol | Gene Function                                                                 |
|---------------|-------------|-------------------------------------------------------------------------------|
| SPAC212.11    | tlh1        | RecQ type DNA helicase                                                        |
| SPAC19G12.16c | adg2        | conserved fungal cell surface protein, Kre9/Knh1 family, Adg2                  |
| SPBC1348.14c  | ght7        | plasma membrane hexose transmembrane transporter Ght7                         |
| SPAPB1E7.04c  | SPAPB1E7.04c| chitinase                                                                      |
| SPBC1105.05   | exg1        | cell wall 1,6-beta-glucosidase Exg1                                           |
| SPRRNA.02     | 15S_rRNA    | small subunit (15S) rRNA, rns                                                  |
| SPAC1039.11c  | gto1        | alpha-glucosidase                                                             |
| SPSNORNA.32   | sno12       | box H/ACA small nucleolar RNA 12/snR99                                         |
| SPAC186.09    | pdc102      | pyruvate decarboxylase                                                         |
| SPAC19B12.02c | gas1        | cell wall 1,3-beta-glucanosyltransferase Gas1                                  |
| SPBC4F6.12    | pxl1        | paxillin-like protein Px1                                                      |
| SPRRNA.45     | SPRRNA.45   | 18S ribosomal RNA                                                             |
| SPAC1F8.05    | isp3        | spore wall structural constituent Isp3                                         |
| SPAC750.01    | SPAC750.01  | NADP-dependent aldo/keto reductase, unknown biological role, implicated in cellular detoxification |
| SPRRNA.46     | SPRRNA.46   | 18S ribosomal RNA                                                             |
| SPMIT.06      | SPMIT.06    | mitochondrial DNA binding endonuclease (intron encoded)                       |
| SPNCRNA.532   | SPNCRNA.532 | non-coding RNA (predicted)                                                     |
| SPRRNA.44     | SPRRNA.44   | 18S ribosomal RNA                                                             |
| SPAC2G8.05c   | cdc15       | F-BAR domain protein Cdc15                                                    |
| SPRRNA.01     | rnl         | large subunit (21S) rRNA, rnl                                                  |
| SPBPB2B2.13   | gal1        | galactokinase Gal1                                                            |
| SPNCRNA.942   | SPNCRNA.942 | intergenic RNA (predicted)                                                     |
| SPAPB1E7.05   | gde1        | glycerophosphoryl diester phosphodiesterase Gde1                              |
| SPAC13G7.04c  | mac1        | plasma membrane anchored protein, claudin family, predicted membrane sensor Mac1 |
| SPCC306.11    | SPCC306.11  | Schizosaccharomyces specific protein, uncharacterized                          |
| SPNCRNA.1374  | cta3-antisense-1 | antisense RNA (predicted)                                       |
| SPBC11C11.05  | SPBC11C11.05| conserved fungal cell wall protein, Kre9/Knh1 family                           |
| SPAC23A1.02c  | SPAC23A1.02c| GPI-remodeling mannose-ethanolamine phosphate phosphodiesterase Ted1          |
| SPAC1006.08   | etd1        | Spg1-binding protein Etd1                                                     |
| SPBC1289.01c  | chr4        | SEL1/TPR repeat protein1, 3-beta-glucan synthase regulatory factor Ch3/Chr4    |
| SPAC1F7.05    | cdc22       | ribonucleoside reductase large subunit Cdc22                                  |
| SPBC1685.14c  | vid27       | WD repeat protein, Vid27 family, conserved in fungi and plants                 |
| SPAC821.09    | eng1        | cell septum surface endo-1,3-beta-glucanase Eng1                              |
| SPBC1289.04c  | pob1        | Boi family protein                                                            |
| SPBC31E1.06   | bms1        | GTP binding protein Bms1                                                      |
Table 8
List of genes downregulated in Δatf1Δpcr1 cells.

| Gene ID       | Gene Symbol | Gene Function                                                                 |
|---------------|-------------|-------------------------------------------------------------------------------|
| SPAC688.16    | SPAC688.16  | human TMEM254 ortholog                                                        |
| SPNCRNA.1255  | SPNCRNA.1255| intergenic RNA (predicted)                                                    |
| SPAC29A4.12c  | SPAC29A4.12c| Schizosaccharomyces specific protein, uncharacterized                         |
| SPBC660.05    | wwm3        | WW domain containing conserved fungal protein Wwm3                            |
| SPAPB18E9.05c | SPAPB18E9.05c| dubious                                                                       |
| SPNCRNA.1223  | SPCC191.10-antisense-1 | antisense RNA (predicted)                                                     |
| SPBPB21E7.11  | SPBPB21E7.11| Schizosaccharomyces pombe specific protein, uncharacterized                   |
| SPAP27G11.13c | nop10       | box H/ACA snoRNP complex protein                                              |
| SPAC9A8.16    | prf65       | tudor domain superfamily protein                                              |
| SPAC513.03    | mfm2        | M-factor precursor Mfm2                                                       |
| SPBC56F2.15   | tam13       | Schizosaccharomyces specific protein, uncharacterized                         |
| SPCCP16A4.07  | SPCCP16A4.07| Stm1/Oga1 family protein Smp4                                                 |
| SPAC15E1.02c  | SPAC15E1.02c| DUF1761 family protein                                                        |
| SPBC26H8.14c  | cox17       | mitochondrial copper chaperone for cytochrome c oxidase Cox17                 |
| SPCC684.13c   | rpl1201     | 60S ribosomal protein L12.1/L12A                                               |
| SPAC823.17    | tom6        | mitochondrial TOM complex subunit Tom6                                       |
| SPCC663.02    | wtf14       | wtf element Wtf14                                                            |
| SPBC1604.11   | atp17       | F1-FO ATP synthase subunit F                                                  |
| SPBC484.05    | smg1        | Sm snRNP core protein Smg1                                                   |
| SPCC31H12.04c | rpl1202     | 60S ribosomal protein L12.1/L12A                                               |
| SPCC1259.05c  | cox9        | cytochrome c oxidase subunit VIIa                                              |
Table 9
List of genes upregulated by Pcr1 independent of Atf1.

| Gene ID             | Gene Symbol | Gene Function                                                                 |
|---------------------|-------------|--------------------------------------------------------------------------------|
| SPBC32F12.03c       | gpx1        | H2O2 scavenger glutathione peroxidase Gpx1                                      |
| SPBPB2B2.06c        | prl65       | tudor domain superfamily protein                                                |
| SPAC19A8.16         | rpp202      | 60S acidic ribosomal protein P2                                                  |
| SPBC23G7.15c        | sme2        | meiosis-specific Smp foci IncRNA Smc2, long isoform                              |
| SPAC4F10.20         | grx1        | glutaredoxin Grx1                                                               |
| SPAC22H10.13        | zym1        | metallothionein Zym1                                                            |
| SPAC1F8.01          | ght3        | plasma membrane glucose:proton symporter Ght3                                   |
| SPAC21E11.03c       | pcr1        | DNA-binding transcription factor Pcr1                                            |
| SPBC56F2.15         | tam13       | Schizosaccharomyces specific protein, uncharacterized                           |
| SPCC1393.12         | SPCC1393.12 | Schizosaccharomyces specific protein, uncharacterized                           |
| SPBC11C8.16         | prl65       | tudor domain superfamily protein                                                |
| SPNCRNA.103         | sme2        | meiosis-specific Smp foci IncRNA Smc2, long isoform                              |
| SPAC23C4.11         | stri3       | plasma membrane heme transmembrane transporter Str3                              |
| SPAC1F8.05          | isp3        | spore wall structural constituent Isp3                                           |
| SPAC2B10.04c        | nrf1        | vacuolar transporter chaperone (VTC) complex, GTPase regulator subunit Nrf1      |
| SPBC215.11c         | SPBC215.11c | aldo/keto reductase, unknown biological role                                    |
| SPBC11B10.10c       | pht1        | histone H2A variant H2A.Z Pht1                                                   |
| SPAC4F8.10c         | stk1        | SM22/transgelin-like actin modulating protein Stg1                              |
| SPACF18.05          | SPACF18.05  | alpha,alpha-trehalose-phosphate synthase                                         |
| SPAPB18E9.05c       | SPAPB18E9.05c | dubious                                                                          |
| SPBBP21E7.11        | SPBBP21E7.11 | Schizosaccharomyces pombe specific protein, uncharacterized                     |
| SPAC513.03          | mfm2        | M-factor precursor Mfm2                                                         |
| SPCC16C4.13c        | rpl1201     | 60S ribosomal protein L12.1/L12A                                                |
| SPBC16D4.11         | atp17       | F1-FO ATP synthase subunit F                                                    |
| SPCC31H12.04c       | rpl1202     | 60S ribosomal protein L12.1/L12A                                                |
| SPCC1259.05c        | cox9        | cytochrome c oxidase subunit VIIa                                               |
| SPAC29A4.12         | mug108      | Schizosaccharomyces specific protein, uncharacterized                           |
**Fig. 2.** Transcriptome analysis reveals targets which are differentially downregulated by Pcr1 (A) 28 genes that were found to be upregulated in Δatf1 Δpcr1 cells are possible targets negatively regulated by Pcr1. B) Categories with the highest number of genes in the significant functional clusters obtained from DAVID based analysis are represented for the 35 genes found to be downregulated by Pcr1. (C) Genes were sorted on the basis of their function in the cell cycle and/or stress response. Graph reflects the distribution of genes in each category.
Table 10
List of genes repressed by Pcr1 independent of Atf1.

| Gene ID      | Gene Symbol | Gene Function                                                                 |
|--------------|-------------|-------------------------------------------------------------------------------|
| SPAC212.11   | tlh1        | RecQ type DNA helicase                                                        |
| SPBC1348.14c | ght7        | plasma membrane hexose transmembrane transporter Ght7                         |
| SPBC1105.05  | exg1        | cell wall glucan 1,6-beta-glucosidase Exg1                                    |
| SPAC1309.11c | go1         | alpha-glucosidase                                                             |
| SPNORNA.32   | sno12       | box H/ACA small nucleolar RNA 12/snR99                                        |
| SPAC186.09   | pdc102      | pyruvate decarboxylase                                                         |
| SPAC19B12.02c| gas1        | cell wall 1,3-beta-glucanoylsynterase Gas1                                    |
| SPBC4F6.12   | pxl1        | paxillin-like protein Pxl1                                                     |
| SPRRNA.45    | SPRRNA.45   | 18S ribosomal RNA                                                             |
| SPAC1F8.05   | isp3        | spore wall structural constituent Isp3                                         |
| SPAC750.01   | SPAC750.01  | NADP-dependent aldo/keto reductase, unknown biological role, implicated in cellular detoxification |
| SPRRNA.46    | SPRRNA.46   | 18S ribosomal RNA                                                             |
| SPNCRNA.532  | SPNCRNA.532 | non-coding RNA (predicted)                                                    |
| SPAC2G8.05c  | cdc15       | F-BAR domain protein Cdc15                                                    |
| SPBHP282.13  | gal1        | galactokinase Gal1                                                            |
| SPNCRNA.942  | SPNCRNA.942 | intergenic RNA (predicted)                                                    |
| SPAPB1E7.05  | gde1        | glycrophosphoryl diester phosphodiesterase Gde1                               |
| SPAC13G7.04c | mac1        | plasma membrane anchored protein, claudin family, predicted membrane sensor Mac1 |
| SPCC306.11   | SPCC306.11  | Schizosaccharomyces specific protein, uncharacterized                         |
| SPCC1374     | cta3-antisense-1 | antisense RNA (predicted)                        |
| SPBC11C11.05 | SPBC11C11.05 | conserved fungal cell wall protein, Kre9/Knh1 family                          |
| SPAC23A1.02c | ted1        | GPI-remodeling mannose-ethanolamine phosphate phosphodiesterase Ted1          |
| SPAC1006.08  | etd1        | Spg1-binding protein Etd1                                                     |
| SPBC1289.01c | chr4        | SEL1/TPR repeat protein, 3-beta-glucan synthase regulatory factor Chf3/Chr4    |
| SPAC1F7.05   | cdc22       | ribonucleoside reductase large subunit Cdc22                                  |
| SPBC1685.14c | vid27       | WD repeat protein, Vid27 family, conserved in fungi and plants                 |
| SPBC1289.04c | poh1        | Boi family protein                                                            |
| SPCC576.01c  | xan1        | alpha-ketoglutarate-dependent xanthine dioxygenase Xan1                       |
| SPCC1223.09  | uro1        | uricase Uro1                                                                  |
| SPAC1002.19  | urg1        | GTP cyclohydrolase II Urg1                                                     |
| SPAC1039.02  | SPAC1039.02 | extracellular 5'-nucleotidase, human NT5E family                              |
| SPCC4B3.10c  | ipk1        | inositol 1,3,4,5,6-pentakisphosphate (IP5) kinase                              |
| SPAC568.03   | tif52       | translation initiation factor eIF5B Tif52                                      |
| SPAC29B12.14c| SPAC29B12.14c| plasma membrane purine transmembrane transporter                              |

Table 11
Genes upregulated by Atf1 and downregulated by Pcr1.

| Gene name     | Gene Symbol | Gene Function                                                                 |
|---------------|-------------|-------------------------------------------------------------------------------|
| SPCC1906.04   | wtf20       | wtf antidote-like meiotic drive suppressor Wtf20                               |
| SPAC1834.04   | hlt1        | histone H3 h3.1                                                               |
| SPCC1739.15   | wtf21       | wtf meiotic drive antidote-like Wtf21                                          |
| SPBC1105.12   | hlf3        | histone H4 h4.3                                                               |
| SPAC1834.03c  | hlf1        | histone H4 h4.1                                                               |
| SPAC750.01    | SPAC750.01  | NADP-dependent aldo/keto reductase, unknown biological role, implicated in cellular detoxification |
| SPBPB282.13   | gal1        | galactokinase Gal1                                                            |
| SPAC1002.19   | urg1        | GTP cyclohydrolase II Urg1                                                     |
Table 12
Genes downregulated by Atf1 and upregulated by Pcr1.

| Gene Name         | Gene Symbol | Gene Function                                                                 |
|-------------------|-------------|--------------------------------------------------------------------------------|
| SPAPB1E7.04c      | SPAPB1E7.04c| chitinase                                                                      |
| SPAC19G12.16c     | adg2        | conserved fungal cell surface protein, Kre9/Knh1 family, Adg2                  |
| SPCPB1C11.01      | amt1        | plasma membrane ammonium transmembrane transporter Amt1                       |
| SPRRNA.02         | rns         | small subunit (15S) rRNA, rns                                                 |
| SPAC821.09        | eng1        | cell septum surface endo-1,3-beta-glucanase Eng1                              |
| SPBPB2B2.06c      | SPBPB2B2.06c| extracellular 5′-nucleotidase, human NT5E family                              |
| SPAC1F8.01        | ght3        | plasma membrane gluconate:proton symporter Ght3                               |
| SPBC359.06        | mug14       | adducin, involved in actin cytoskeleton organization                           |
| SPAC1F8.03c       | str3        | plasma membrane heme transmembrane transporter Str3                            |
| SPAC513.03        | mfm2        | M-factor precursor Mfm2                                                       |

Fig. 3. Comparative analysis of transcriptome reveals new targets of Atf1. The genes found to be differentially regulated by Atf1 in our analysis have been compared with published data [3]. (A) Comparing genes downregulated in Δatf1 *S. pombe* cells of our analysis with genes already reported as targets of Atf1, we found 75 unique genes to be upregulated by Atf1, which can be identified as new targets of the transcription factor. (B) Comparison between genes upregulated in Δatf1 cells and genes reported to be de-repressed in Δatf1 cells have revealed 34 new targets of Atf1, which are uniquely downregulated in our analysis.
2. Materials and Methods

2.1. Experimental design

Differential gene expression studies based on RNA sequencing were carried out following overexpression experiments in a series of *S. pombe* transformants and mutants. All samples were processed in duplicates.

2.2. Strains, media and growth conditions

*S. pombe* strains used in this study are listed in (Table 13). Cells were grown as described in [8]. For overexpression experiments, cells were grown overnight in Edinburgh Minimal Medium, EMM (Leu-) supplemented with 20 μM thiamine, harvested, washed, resuspended in EMM (Leu-) and incubated for 24 h at 30 °C. Cells were thereafter harvested, washed and resuspended in RNAlater Stabilization Solution (Thermo Scientific).

2.3. *S. pombe* transformation

1 ml of overnight *S. Pombe* cultured in YES was harvested and then resuspended in 0.5 ml PEGLET (10 mM Tris [pH 8], 1mM EDTA, 0.1 M lithium acetate, 40% polyethylene glycol [PEG]). 5μl of denatured salmon sperm DNA (10 mg/ml) was added to it. 1 μg of the purified plasmid DNA was then added to this mixture and allowed to stand overnight at room temperature, after which the cells were resuspended in 150 μl YES and spread onto appropriate selection plates.

2.4. RNA isolation

TRIzol™ Reagent (Invitrogen) was used for RNA isolation. After homogenizing the sample with TRIzol™ reagent, chloroform was added, and the homogenate was allowed to separate into a clear upper aqueous layer (containing RNA), an interphase, and a red lower organic layer (containing the DNA and proteins). RNA was thereafter precipitated from the aqueous layer with isopropanol. Furthermore, the steps of cDNA library preparation and Next Generation Sequencing and Analysis were done by Agrigenome.

2.5. Library preparation

TruSeqstranded mRNA sample preparation protocol was used to capture coding RNA and multiple forms of noncoding polyadenylated RNA using poly-T oligo attached magnetic beads. After fragmentation of mRNA, first-strand cDNA was done using reverse transcriptase (strand specificity was obtained by replacing dTTP with dUTP, followed by second-strand cDNA synthesis.

| Strain/Plasmid Number | Genotype/Description | Source |
|-----------------------|----------------------|--------|
| GSY001                | h- leu1-32 ura4-D18   | Paul Russel (PR109) |
| GSY027                | h- atf1::ura4+        | Kazuhiro Shiozaki (KS1497) |
| GSY499                | h- leu1 ura4pcr1::ura4+ atf1::kanMX6 | Elena Hidalgo (MS48) |
| pGS017                | pREP41               | Yeast Genetic Resource centre |
| pGS044                | pREP41+Pcr1          | Lab Stock [1] |

Table 13
List of strains used in the study.
using DNA Polymerase I and RNase H. Then adenylation of the 3’ ends are done following ligation of adapters. The products are then purified and enriched with PCR to create the final cDNA library. Finally, quality control analysis and quantification of the DNA library templates were performed to create optimum cluster densities across every lane of flow cell.

2.6. Data analysis

Raw sequence data generation was done using Fastq [9] file followed by data quality check. Mapping is done to the reference genome using Kim et al [10]. to evaluate sample quality, followed by differential expression analysis using cuffdiff [11,12] Gene Ontology Annotations were assigned using Uniprot [13] and the report of the analysis was produced. Correlation analyses were performed to check the variability between replicates and across samples The box plot was used to show the distribution of data based on the five number summary. Log transformation is performed to make the variation similar across orders of magnitude (See Supplementary Figure S1). The correlation between the samples being compared was revealed by the scatter plot. The samples being compared are said to be highly correlated if the data falls in a straight line (See Supplementary Figure S2). The distance matrix plot showed the correlation between the samples being compared. (See Supplementary Figure S3). The matrix plot describes the number of significant genes at 5% FDR for each pairwise interaction tested. It gives a quick view of the number of significant features at a given q value cutoff <= 0.05 (See Supplementary Figure S4). The Volcano plot helps visualize the statistically significant differentially expressed genes. The plot is constructed by plotting -log10 (p-value) on the y-axis, and the log2 fold change between the two samples on the X-axis. Genes that pass the filtering of q-value <0.05 are indicated on the plot in red (See Supplementary Figure S5). Further analysis was performed in lab. Genes with significant fold changes were taken for analysis and a cut off of ≥1.5 fold for up-regulated genes and ≤0.75 fold for down-regulated genes was set for further analysis of the differential expression in the gene sets. Gene clusters and functions were generated using DAVID Functional Annotation Bioinformatics tool (David v6.8) [2,3]. Lock et al [14], was used to assign and verify specific functions of the respective genes. Gene expression profiles during cell cycle and stress were explored using Chen Lab Resources [4,5]. Hulsen et al [7], application was used for the comparison and visualization of gene lists using area proportional Venn diagrams.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

CRediT Author Statement

Sohini Basu: Methodology, Formal analysis, Data curation, Writing – review & editing; Priyanka Sarkar: Formal analysis, Data curation, Writing – review & editing; Suchismita Datta: Methodology, Formal analysis, Data curation, Writing – review & editing; Geetanjali Sundaram: Methodology, Formal analysis, Data curation, Writing – review & editing.
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Supplementary Materials

Supplementary material associated with this article can be found in the online version at doi:10.1016/j.dib.2022.108034.

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