





Tra1 controls the transcriptional landscape of the aging cell

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Abstract

Gene expression undergoes considerable changes during the aging process. The mechanisms regulating the transcriptional response to cellular aging remain poorly understood. Here, we employ the budding yeast *Saccharomyces cerevisiae* to better understand how organisms adapt their transcriptome to promote longevity. Chronological lifespan assays in yeast measure the survival of nondividing cells at stationary phase over time, providing insights into the aging process of postmitotic cells. Tra1 is an essential component of both the yeast Spt-Ada-Gcn5 acetyltransferase/Spt-Ada-Gcn5 acetyltransferase-like and nucleosome acetyltransferase of H4 complexes, where it recruits these complexes to acetylate histones at targeted promoters. Importantly, Tra1 regulates the transcriptional response to multiple stresses. To evaluate the role of Tra1 in chronological aging, we took advantage of a previously characterized mutant allele that carries mutations in the *TRA1* PI3K domain (*tra1_{Q3}*). We found that loss of functions associated with *tra1_{Q3}* sensitizes cells to growth media acidification and shortens lifespan. Transcriptional profiling reveals that genes differentially regulated by Tra1 during the aging process are enriched for components of the response to stress. Notably, expression of catalases (*CTA1*, *CTT1*) involved in hydrogen peroxide detoxification decreases in chronologically aged *tra1_{Q3}* cells. Consequently, they display increased sensitivity to oxidative stress. *tra1_{Q3}* cells are unable to grow on glycerol indicating a defect in mitochondria function. Aged *tra1_{Q3}* cells also display reduced expression of peroxisomal genes, exhibit decreased numbers of peroxisomes, and cannot grow on media containing oleate. Thus, Tra1 emerges as an important regulator of longevity in yeast via multiple mechanisms.

Keywords: chronological aging; yeast; SAGA complex; Tra1; peroxisomes

Introduction

Saccharomyces cerevisiae has been extensively studied as a eukaryotic model for lifespan regulation since many of the underlying molecular mechanisms are conserved from yeast to mammals (Mortimer and Johnston 1959; Fontana et al. 2010; Kaeberlein 2010; Mirisola and Longo 2022). Chronological lifespan (CLS, defined as the extent of time nondividing cells survive in a nutrient-deprived environment; MacLean et al. 2001; Laun et al. 2006; Murakami et al. 2008) and replicative lifespan (the number of cell divisions before replicative senescence; Steinkraus et al. 2008) are 2 distinct experimental aging models (Laun et al. 2006). CLS experiments are carried out by culturing cells for a substantial period at the stationary phase. During chronological aging, yeast cells go through distinct growth phases: the mid-log phase, diauxic shift, and stationary phase. Each growth phase is illustrated by divergent metabolic activities and gene expression profiles, which are analogous to crucial characteristics of mammalian aging cells including cell cycle arrest, increased respiratory activity, and lipid and protein homeostasis (Tissenbaum and Guarente 2002; Beach et al. 2013, 2015; Medkour and Titorenko 2016; Chadwick and Lajoie 2019; Chadwick et al. 2020; Mohammad et al. 2020). Studies of yeast chronological aging have enabled researchers to identify several key pathways that regulate longevity (Longo and Fabrizio 2012) such as Ras/Pka (Longo 1999)

and Sch9/Tor (Fabrizio et al. 2001) and the role of sirtuins (Kaeberlein et al. 1999), not only in yeast but also in other organisms.

How cells modulate their gene expression in response to stresses including aging involves all components of the transcriptional machinery. The recruitment of RNA polymerase II to promoters is tightly regulated by general transcription factors, gene-specific activators, and coactivators. Coactivators are multisubunit protein complexes that interface between general transcription factors and gene-specific activators (Kingston et al. 1996; Eberharter and Becker 2002; Helmlinger et al. 2011) and/or regulate modification of histone proteins and nucleosome remodeling (Roberts and Winston 1997; Rando and Winston 2012). Thus, coactivators play a vital role in global gene regulation. Spt-Ada-Gcn5 acetyltransferase (SAGA) and nucleosome acetyltransferase of H4 (NuA4) are prototypical multisubunit coactivator protein complexes that are conserved among eukaryotic organisms. SAGA and NuA4 contain lysine acetyltransferases Gcn5 and Esa1, respectively, which acetylate both histone and nonhistone proteins (Grant et al. 1997; Clarke et al. 1999; Steunou et al. 2014; Downey 2021). Transcriptome analyses for yeast strains lacking the SAGA components Gcn5 or Spt3 revealed that 10% of the stress-related genome is controlled by the SAGA complex (Huisinga and Pugh 2004). SAGA and NuA4 also participate in

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other non-transcriptional activities, such as telomere maintenance and DNA repair (Bird *et al.* 2002; Downs *et al.* 2004; Lin *et al.* 2008; Atanassov *et al.* 2009; Cheng *et al.* 2018, 2021). In addition, SAGA contains a deubiquitinating module (DUB) that includes the ubiquitin protease Ubp8 that cleaves ubiquitin from histone H2B (Henry *et al.* 2003; Morgan *et al.* 2016) and other targets. The additional targets for deubiquitination by the mammalian homolog of Ubp8, USP22, are particularly noteworthy for their role in cancer progression (Prokakis *et al.* 2021; Ning *et al.* 2022; De Luca *et al.* 2022).

Tra1 is uniquely found in both the SAGA and NuA4 complexes (Saleh *et al.* 1998; Grant *et al.* 1998; Allard *et al.* 1999). Tra1 belongs to the phosphoinositide-3-kinase-related kinase (PIKK) family, which includes ataxia telangiectasia mutated (ATM; Tel1 in *S. cerevisiae*), ataxia telangiectasia, and Rad3 related (ATR; Mec1 in *S. cerevisiae*), the DNA-dependent protein kinase catalytic subunit (DNA-PKc), mammalian target of rapamycin (Tor1 and Tor2 in *S. cerevisiae*), and SMG-1 (suppressor with morphological effect on genitalia family member) (Smith and Jackson 2003; Hill and Lee 2010; Shiloh and Ziv 2013). PIKK proteins have 4 common principal domains: an N-terminal Huntingtin, elongation factor 3 (EF3), protein phosphatase 2A (PP2A), and TOR1 (HEAT), followed by FRAP-ATM-TRRAP (FAT), phosphatidylinositol 3-kinase (PI3K), and FRAP-ATM-TRRAP C-terminus (FATC) domains (Keith and Schreiber 1995; Bosotti *et al.* 2000; Mordes *et al.* 2008). Approximately, half of the Tra1 molecule consists of helical HEAT repeats and this section interacts with activator proteins to initiate transcription (Brown *et al.* 2001; Bhaumik *et al.* 2004; Knutson and Hahn 2011; Lin *et al.* 2012). A helical FAT domain located C-terminal to the HEAT repeats wraps the N-terminal section of the PI3K domain (Díaz-Santín *et al.* 2017; Sharov *et al.* 2017). At the C-terminus, PIKK family members share a highly conserved phosphoinositide-3-kinase (PI3K) domain. The PI3K domain consists of N- and C-terminal subdomains with a cleft between these 2 lobes, where ATP binds in the catalytically active PIKK family members (Huse and Kuriyan 2002; Taylor and Kornev 2011; Pavletich and Yang 2013). In all PIKK family members except Tra1/TRRAP, the PI3K domain regulates cell signaling by phosphorylating downstream target proteins. The PI3K domain of Tra1 lacks residues essential for ATP binding and phosphate transfer and therefore has no demonstrable kinase activity (Saleh *et al.* 1998; Nelson *et al.* 2006; Mutiu *et al.* 2007). Interestingly, however, mutation of conserved arginine residues to glutamine (termed *tra1_{Q3}*) in the kinase cleft domain results in slow growth, increased stress sensitivity, and decreased transcription of SAGA-regulated genes supporting a role for this pseudokinase domain (Berg *et al.* 2018).

In light of the importance of epigenetic changes in aging (Booth and Brunet 2016; Kane and Sinclair 2019), the involvement of SAGA and NuA4 in stress response processes, and the unique role of Tra1 in both SAGA and NuA4 (Helmlinger *et al.* 2011; Cheung and Díaz-Santín 2019), we hypothesized that Tra1 contributes to longevity by regulating the transcriptional responses to stress associated with aging. Indeed, using a loss-of-function mutant, we find that Tra1 significantly alters the transcriptional landscape of the aging cell. Tra1 therefore emerges as a new regulator of the chronological aging process in yeast.

Materials and methods

Drugs

Oleic and myristic acid and diamide were purchased from Milipore-Sigma. Propidium iodide was from Thermo Fisher

Scientific. 2-(N-morpholino)ethanesulfonic acid (MES) buffer solution was from Alfa Aesar.

Strains and plasmids

Yeast strains were constructed in the W303A background or BY4742 and are described in Supplementary Table 1. Strains expressing either wild-type FLAG-tagged TRA1 or *tra1_{Q3}* were created by integrating an *SphI-SacI* fragment from pCB2527 or pCB2537, respectively, into W303A and selecting HIS⁺ cells expressing either wild-type URA3-FLAG-tagged TRA1 or *tra1_{Q3}* PI3K domain and YCplac111-DED1pr-YHR100 as previously described (Berg *et al.* 2018). yemRFP-SKL was generated by cloning yemRFP-SKL into the *SpeI/SalI* sites of pRS416-GDP (Mumberg *et al.* 1995).

Culture conditions and cell viability assay

Both the TRA1 and *tra1_{Q3}* yeast cells (W303A derivatives) were grown to saturation overnight in synthetic complete medium (2% glucose with appropriate selection). An aliquot of the overnight cultures was diluted in fresh media and grown at 30°C in a rotating drum. Cell viability assays were conducted using propidium iodide as described previously (Chadwick *et al.* 2016). Briefly, cells were washed and resuspended in phosphate-buffered saline (PBS) containing 1 mM of propidium iodide. The positive control was prepared by boiling cells for 10 min before resuspending cells in propidium iodide staining solution. Unstained cells were used as a negative control. All samples were incubated for 10 min at room temperature in 96-well plates before imaging on a Gel doc system (Bio-Rad). The optical density (OD₆₀₀) was measured using a BioTek Epoch 2 microplate reader. Both the TRA1 and *tra1_{Q3}* cells were cultivated for several days and the cell viability of the chronologically aged cells was quantified at various time points throughout the aging process. Alternatively, the CLS assay was performed using caloric restricted medium (0.1% glucose), medium buffered with 0.1 M MES buffer, or media containing 0.1% oleic acid, 0.1% myristic acid, or 2% glycerol. To assess sensitivity to acetic acid, cells were grown in synthetic complete medium for 4 days and treated with acetic acid for 200 min with concentrations up to 0.08 mM and the cell viability assessed using propidium iodide. Survival rates were computed using the mean gray value from images along with OD₆₀₀ using the ANALYSR program (Chadwick *et al.* 2016).

Growth assays

A single colony of yeast strain was inoculated and grown overnight in 2% synthetic medium without leucine at 30°C, 220 rpm. Overnight cultures were serially diluted and OD₆₀₀ measured using a spectrophotometer. Cell cultures were normalized to OD₆₀₀ 0.1- and 5-fold serial dilutions were spotted on solid media. Plates were incubated for 2 days at 30°C before taking images using a colony imager (S&P Robotics). Growth was quantified as previously described (Petropavlovskiy *et al.* 2020).

RNA isolation and sequencing

Both TRA1 and *tra1_{Q3}* cells were grown to saturation overnight, diluted 10-fold in 2% synthetic minus leucine medium and grown for 4 h at 30°C in a rotating drum. An aliquot of the cells was spun down at day 0 and day 3 and stored at -80°C. Total RNA was isolated from cells using the RiboPure yeast kit (Thermo Fisher Scientific) according to the manufacturer's instructions. RNA samples were prepared with 3 replicates for each genotype and time point. Quality of the RNA samples were analyzed using a Bioanalyzer to ensure a RIN value of 8 or higher.

Total RNA sequencing analysis was conducted by Azenta Life Sciences. RNA from each sample was converted into single stranded Illumina TruSeq cDNA libraries with poly dT enrichment. Libraries were sequenced on an Illumina HiSeq and each sample yielded between 27.5 and 39.7 million 150-bp paired-end sequencing reads. The raw reads and RNA count data were deposited in NCBI's Gene Expression Omnibus (Edgar et al. 2002).

Quality control, trimming, read alignment, and differential gene expression analysis

Read quality was analyzed using FastQC (www.bioinformatics.babraham.ac.uk/projects/fastqc/). Trimmomatic default settings were used to trim low quality bases and adapter sequences (Bolger et al. 2014). Next, reads were aligned with the reference genome of *S. cerevisiae* S288C sequence (R64-2-1; www.yeastgenome.org/) using STAR (Dobin et al. 2013). Only uniquely mapping reads were retained. featureCount was used to count the reads mapping to each gene (Liao et al. 2014). Differential expression analysis was performed with DESeq2 (Love et al. 2014). Differentially expressed genes with a Benjamini–Hochberg adjusted P-value cutoff of ≤ 0.05 were considered for further analysis.

To determine the enrichment of Rlm1 targets, promoter analysis of differentially expressed genes (\log_2 fold change < 2 and P-value cutoff of ≤ 0.05) was performed using Yeasttract (www.yeasttract.com) (Monteiro et al. 2020).

Fluorescence microscopy

TRA1 and *tra1_{Q3}* cells expressing yemRFP-SKL were grown to saturation overnight, diluted 10-fold in 2% synthetic minus leucine medium and grown for 4 h at 30°C in a rotating drum. An aliquot of the cells was then spun down, washed, resuspended in PBS, and transferred to a Nunc Lab-Tek chamber slide. Cells were imaged using a Zeiss Axiovert A1 wide-field fluorescence microscope with 63× 1.4 NA oil objective using a Texas Red filter (586 nm excitation/603 nm emission) and an AxioCam ICM1 R1 CCD camera. ImageJ was used to analyze the images (Schneider et al. 2012).

Results

Functional Tra1 is required for chronological aging

Tra1 is essential in yeast (Saleh et al. 1998) with the exception of the fission yeast *Schizosaccharomyce pombe*, which expresses a second TRA2 isoform (Helmlinger et al. 2011; Elías-Villalobos, Fort, et al. 2019). To determine the role of Tra1 in the aging process, we took advantage of a loss-of-function mutant that we previously characterized termed *tra1_{Q3}* (Berg et al. 2018). *tra1_{Q3}* contains 3 glutamine substitutions of arginine residues 3,389, 3,390, and 3,456 proximal or within the putative ATP-binding cleft of the P13K domain. *tra1_{Q3}* results in slow growth, increased stress sensitivity, transcriptional defects and impaired SAGA and NuA4 complex assembly (Berg et al. 2018; Razzaq et al. 2021). CLS was assessed by labeling cells with the viability dye propidium iodide (PI) at different time intervals during aging (Fig. 1a). *tra1_{Q3}* cells display significantly reduced lifespan when compared to their wild-type counterparts (Fig. 1, b–d). Viability of *tra1_{Q3}* cells in exponentially growing culture is comparable to wild type. *tra1_{Q3}* cells begin to decline in viability when cells reach the stationary phase indicating that a functional TRA1 allele is required to extend CLS.

Since caloric restriction efficiently increases longevity in yeast and other organisms (Jiang et al. 2000; Fontana et al. 2010; Choi et al. 2013; Arlia-Ciommo et al. 2014; Leonov et al. 2017; Campos et al. 2018), we then tested whether functional Tra1 is required for lifespan extension by caloric restriction. Caloric restriction is achieved by growing the cells in 0.1% glucose medium whereas complete medium contains 2% glucose. We found that when grown under caloric restriction conditions, *tra1_{Q3}* cells display extended lifespan similar to wild-type TRA1 cells (Fig. 2), suggesting that Tra1 function is dispensable for lifespan extension by caloric restriction.

tra1_{Q3} cells are more sensitive to acetic acid

Culture media acidification is a predominant cell-extrinsic factor associated with cell death during chronological aging (Burtner et al. 2009; Burhans and Weinberger 2009; Hu et al. 2014; Depez et al. 2018; Chaves et al. 2021). During chronological aging, acetic acid is produced following depletion of glucose from the growth media when ethanol is utilized as the main carbon source. Indeed, long-lived mutants, such as *sch9Δ* and *ade4Δ*, show increased resistance to acetic acid (Burtner et al. 2009; Matecic et al. 2010). Therefore, we sought to determine if increased sensitivity to acidification of the cell culture media by acetic acid explains, at least in part, the shorter lifespan of *tra1_{Q3}* cells. To this end, TRA1 and *tra1_{Q3}* cells were grown to stationary phase for 4 days and treated with various concentrations of acetic acid. Indeed, *tra1_{Q3}* increases sensitivity to acetic acid compared with wild-type TRA1 (Fig. 3, a and b). Media acidification during chronological aging is alleviated by buffering the cultures to pH 6 with citrate phosphate or MES, resulting in increased longevity (Burtner et al. 2009; Burhans and Weinberger 2009). We found that buffering the aging media pH with MES significantly extended the lifespan of both TRA1 and *tra1_{Q3}* cells (Fig. 3, c and d), suggesting that the inability to respond to acid stress plays an important role in the reduced longevity phenotype associated with the *tra1_{Q3}* mutation.

Tra1 controls the transcriptional landscape of the aging cell

We previously found that Tra1 regulates the expression of genes associated with several stress responses including cell wall stress and the response to misfolded proteins (Berg et al. 2018; Jiang et al. 2019; Razzaq et al. 2021). Based on findings that *tra1_{Q3}* cells have a shorter lifespan, we next sought to determine how the loss of Tra1 function impacts the transcriptional landscape of the aging cell. We performed a global transcriptome analysis using RNA sequencing for TRA1 and *tra1_{Q3}* cells before chronological aging (day 0) and after 3 days at stationary phase (day 3) to identify genes subjected to different transcriptional regulation. Day 3 was selected because the cells have reached stationary phase and still show modest minimal cell death (Supplementary Fig. 1). We found that ~60% of the variance in the transcriptome data is linked to the aging process and ~20% is linked to the *tra1_{Q3}* mutation (Fig. 4a). The larger distance between TRA1 and *tra1_{Q3}* cells after aging suggests that transcriptional differences between the 2 strains is greater in aged cells. At day 0, 394 genes were statistically differentially regulated (175 downregulated and 219 upregulated) when comparing TRA1 and *tra1_{Q3}* cells ($P < 0.05$, \log_2 fold change > 2) (Supplementary Table 2). At day 3, there were 2,026 statistically differentially expressed genes (978 downregulated and 1,048 upregulated) (Supplementary Table 2). When comparing the age–genotype interaction scores (Smith and Kruglyak 2008; Sardi and Gasch 2018), we found 287 genes (161 negative

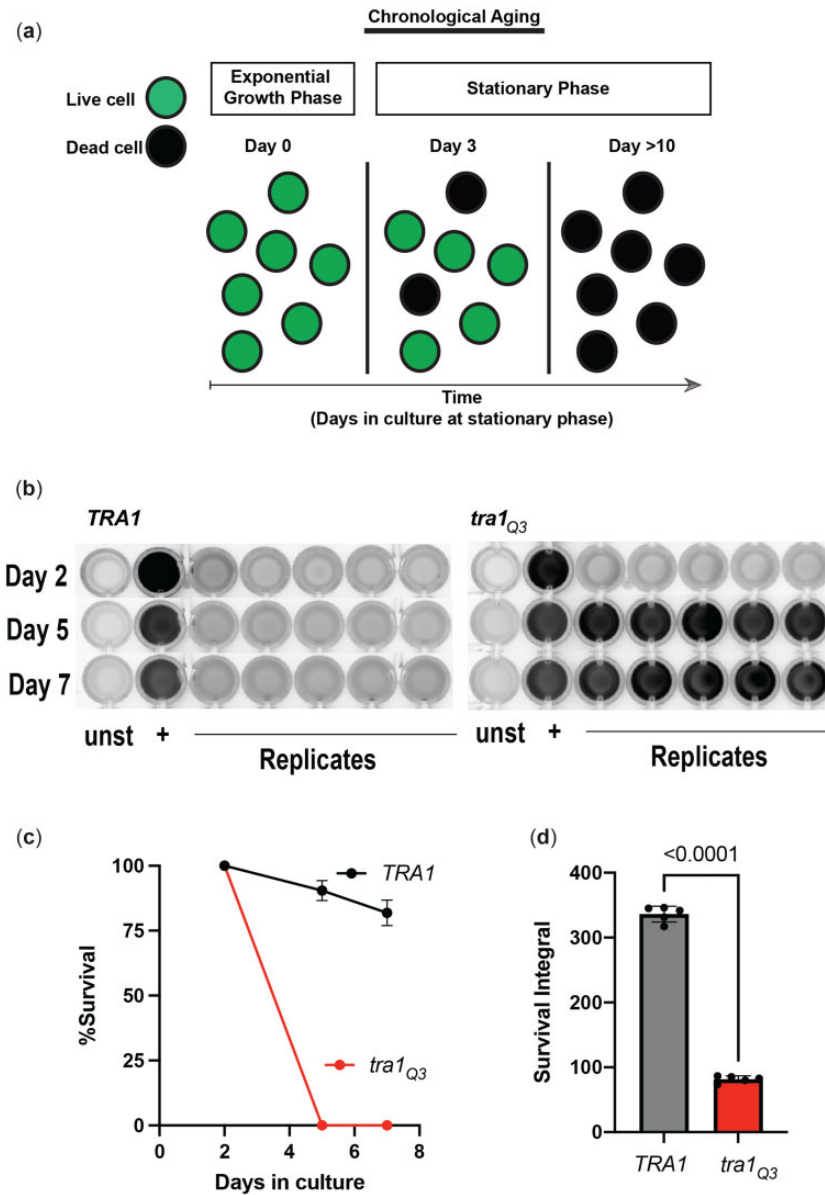


Fig. 1. Functional Tra1 is required for chronological aging. a) CLS is defined as the amount of time yeast cells survive at stationary phase. Experimentally, longevity is assessed by labeling cells with a viability dye such as propidium iodide at various time points during the aging process. Adapted from [Chadwick et al. 2016](#). b) *tra1_{Q3}* cells display reduced CLS. *TRA1* and *tra1_{Q3}* cells were grown for the indicated times in standard synthetic complete medium and stained with propidium iodide to measure cell survival. Stained cells were imaged in a 96-well plate. For each time point, a negative control (unstained cells), a positive control (boiled cells), and 5 replicates were analyzed. c) Normalized survival rates over the aging process and d) survival integral are shown in the bar graph. Significance was assessed using an unpaired Student T-test.

and 126 positive) with a \log_2 score >2.5 and $P < 0.05$, suggesting that these genes differentially respond to the aging process (Fig. 4b) in the 2 genotypes (*TRA1* and *tra1_{Q3}*). Genotype (*TRA1* vs *tra1_{Q3}*) by environment (day 0 vs day 3) interaction scores allow us to decipher how different genotypes specifically modulate gene expression during the aging process. Gene Ontology (GO) analysis revealed significant enrichment in several biological processes, including RNA binding, transmembrane transporter activity, oxidoreductase activity, and unfolded protein binding (Fig. 4c). Genes associated with misfolded protein stress (*SSA1*, *HSP26*, *HSP42*, *SSA4*, *MID1*, *KAR2*, *SSA2*, *CPR6*, *HSP60*, *HSC82*, *APJ1*, *HSP10*, *HSP82*) are upregulated in aged *tra1_{Q3}* cells compared to wild-type suggesting that these cells exhibit increased proteotoxic stress. Genes with a negative age-genotype interaction score were also enriched in Rlm1 target genes, consistent with

our previous results showing that Tra1 controls cell wall integrity (Supplementary Fig. 2) (Berg et al. 2018; Razzaq et al. 2021). Genes with the highest positive and negative interaction scores are presented in Fig. 4d. Genes upregulated in aged *tra1_{Q3}* cells compared to wild type include *RGI1* (involved in metabolism under respiratory conditions) and *BTN2* (a v-snare binding protein). Interestingly, *BTN2* expression is increased under severe ethanol stress (Yamauchi and Izawa 2016), reinforcing the idea that *tra1_{Q3}* cells are subject to increased stress during the aging process. Genes with positive age-genotype interaction are upregulated in wild-type *TRA1* compared to *tra1_{Q3}* in aged cells. Examples of such genes are *SIP18* and *MLS1*. *SIP18* encodes a phospholipid binding hydrophilin involved in vacuolar membrane fusion and is upregulated during replicative lifespan (Chavidel et al. 2018). *MLS1* encodes a malate synthase involved

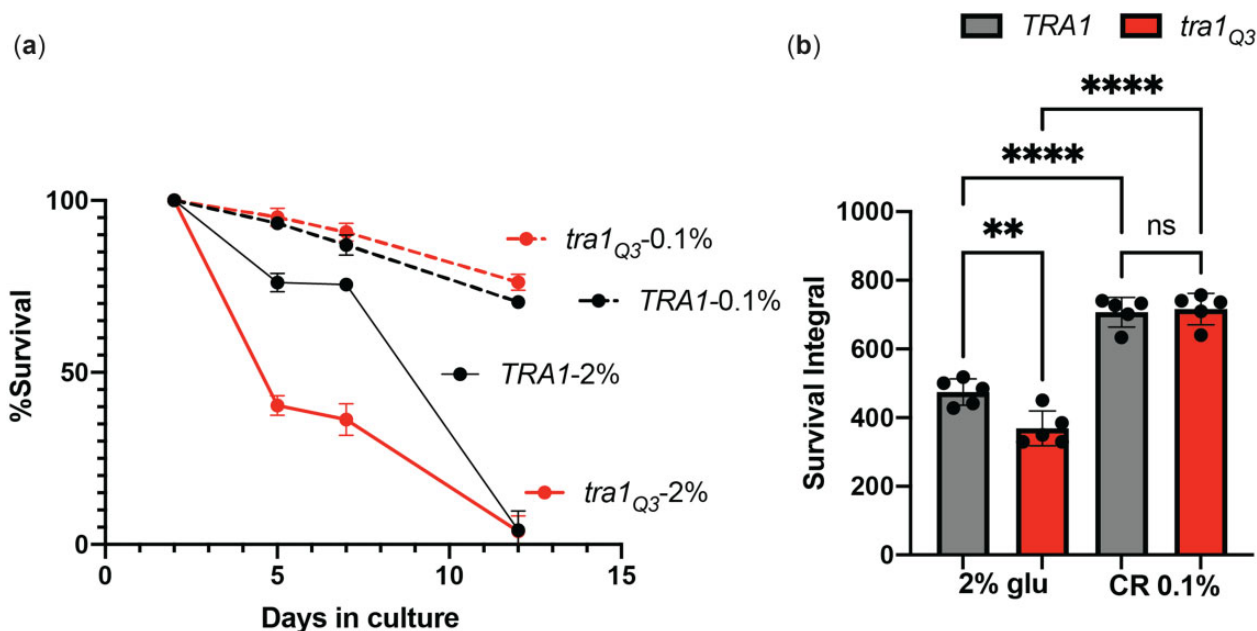


Fig. 2. Tra1 function is dispensable for lifespan extension by caloric restriction. a) TRA1 and *tra1_{Q3}* cells were grown for the indicated time in standard synthetic complete medium containing either 2% glucose or 0.1% glucose (CR) and stained with propidium iodide to measure cell survival. Stained cells were imaged in a 96-well plate. For each time point, a negative control (unstained cells), a positive control (boiled cells), and 5 replicates were analyzed. b) Normalized survival rates over the aging process and calculated survival integral are shown in graphs. Significance was assessed using a 1-way ANOVA followed by a Tukey's multiple comparison test. ** $P < 0.01$ and **** $P < 0.0001$.

in the utilization of non-fermentable carbon sources and is also upregulated in replicatively aged cells (Lesur and Campbell 2004).

We previously showed that cells expressing *tra1_{Q3}* increase expression of TRA1 as a possible compensatory mechanism for the loss of protein function (Berg et al. 2018). Here, we observed this phenomenon as *tra1_{Q3}* mRNA increased ~1.7-fold compared to wild-type at day 0. This difference was exacerbated in aged cells as TRA1 mRNA increased ~3.2-fold in *tra1_{Q3}* cells after aging (Fig. 5a). We also analyzed the mRNA levels of other SAGA and NuA4 components (Fig. 5b). Most striking are the upregulation of ADA2 in aged *tra1_{Q3}* cells and downregulation of components of the SAGA DUB module (*UBP8*, *SUS1*, *SGF73*, *SGF11*). Among the NuA4 components, *ARP4* was upregulated in aged *tra1_{Q3}* cells. Interestingly, mutation of *ARP4* affects CLS in combination with deletion of the linker histone Hho1 (Vasileva et al. 2021).

GO analysis of genes that respond differentially to the aging processing in *tra1_{Q3}* cells revealed an enrichment in genes associated with oxidoreductase activity (Fig. 6a). Among those genes were *CTT1* and *CTA1*, which encode 2 versions of catalase in *S. cerevisiae*. *Ctt1* is cytoplasmic (Seah and Kaplan 1973) while *Cta1* localizes to both the mitochondria and peroxisomes (Petrova et al. 2004). Catalase activity is crucial for hydrogen peroxide detoxification and is an important regulator of oxidative stress resistance associated with various conditions, including aging in yeast (Petriv and Rachubinski 2004; Agarwal et al. 2005; Mesquita et al. 2010; Rona et al. 2015; Guaragnella et al. 2019). Both genes are substantially upregulated in aged wild-type TRA1 cells but not in *tra1_{Q3}* cells (Fig. 6b). Consequently, *tra1_{Q3}* cells were more sensitive to oxidative stress induced by diamide (Fig. 6c). Thus, these data indicate that *tra1_{Q3}* cells have reduced capacity to cope with oxidative damage that is usually associated with the aging process (Pan 2011).

Several differentially expressed genes were also linked to peroxisomal β -oxidation and fatty acid metabolism (*POT1*, *POX1*, *MLS1*, *DCI1*, *ECI1*, *TES1*) (Fig. 7a). The β -oxidation pathway and

peroxisome proliferation are crucial for chronological aging (Lefevre et al. 2013). In yeast, β -oxidation solely occurs in peroxisomes (Hiltunen et al. 2003) and strains with defective peroxisomes fail to grow in presence of oleate due to their incapacity to use fatty acid as a carbon source (Lockshon et al. 2007). We found that *tra1_{Q3}* cells were unable to grow on media containing oleate or myristate, suggesting that they are defective in β -oxidation (Fig. 7b). Because acetyl-CoA produced by β -oxidation serves as an energy source only in respiratory-competent strains, we assessed growth on glycerol. We found that *tra1_{Q3}* cells are unable to grow on plates containing glycerol (Fig. 7b). This phenotype along with the reduced lifespan was also recapitulated in a different strain background (Supplementary Fig. 3). Since β -oxidation in yeast is performed solely in peroxisomes, we also analyzed the expression of *PEX34* and *PEX21* in TRA1 and *tra1_{Q3}* cells during the aging process. *Pex34* regulates peroxisome biogenesis (Tower et al. 2011). *Pex21* regulates import of protein into the peroxisomal matrix (Purdue et al. 1998). Aged *tra1_{Q3}* cells show reduced expression of *PEX34* and *PEX21* compared to aged wild-type cells (Fig. 7c). *tra1_{Q3}* cells consequently display a reduced number of peroxisomes when labeled with the fluorescent reporter *yemRFP-SKL* at both day 0 and day 3 (Fig. 7, d and e). Therefore, our data suggest that *tra1_{Q3}* cells have defective peroxisome function that might contribute to their reduced CLS.

Discussion

A role for Tra1 in chronological aging

Tra1 has been proposed to act as protein-protein hub where it interacts with transcription activators to recruit SAGA and NuA4 coactivator complexes to specific promoters (Elías-Villalobos, Fort, et al. 2019). In both SAGA and NuA4, Tra1 regulates multiple stress responses associated with the aging process such as cell wall integrity, ethanol sensitivity, protein misfolding, lipid synthesis, and the response to DNA damage. Thus, Tra1 regulates

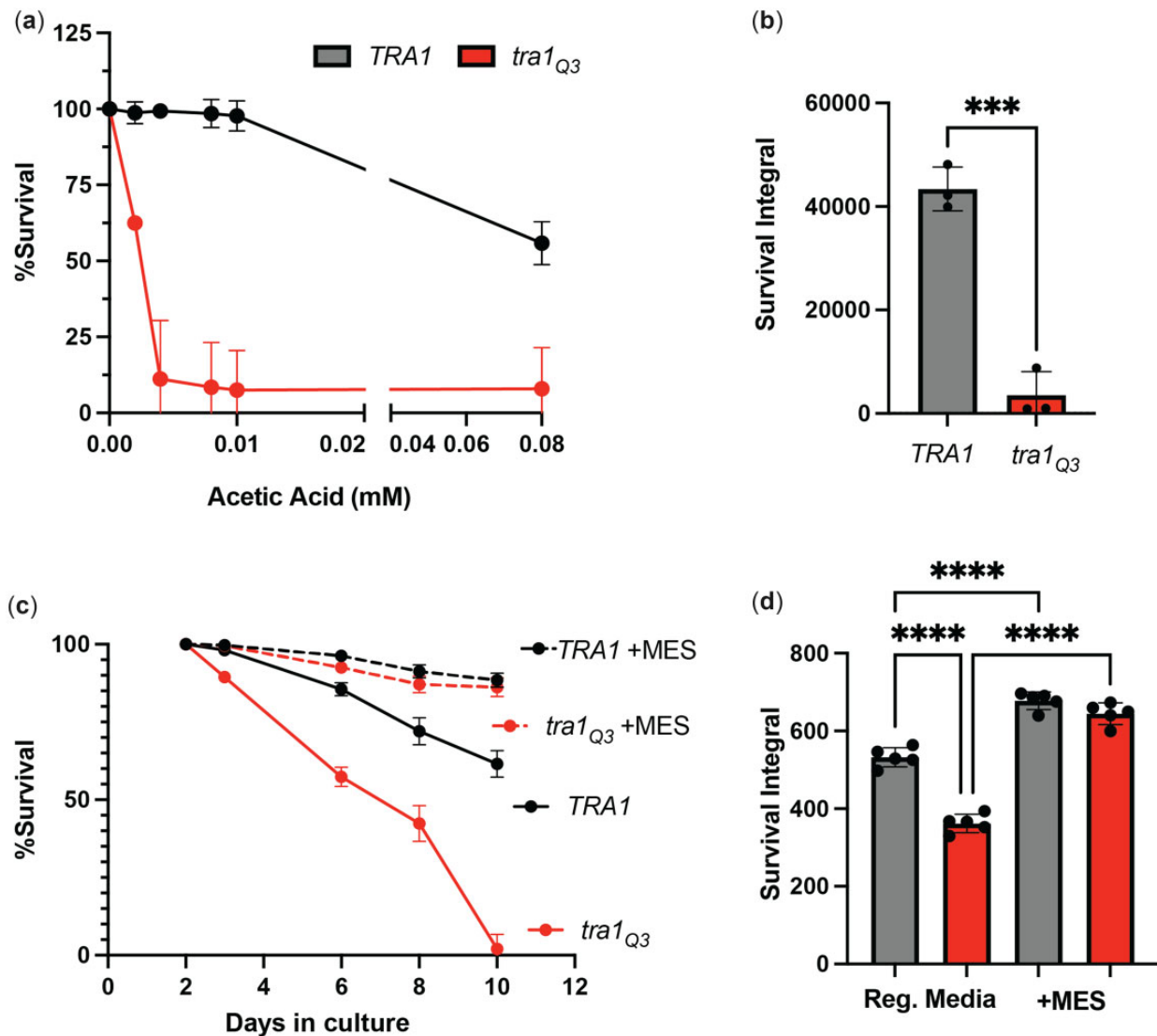


Fig. 3. Increased sensitivity of *tra1*_{Q3} cells to media acidification is linked to shortened lifespan. a) *tra1*_{Q3} cells display increased sensitivity to acetic acid. TRA1 and *tra1*_{Q3} cells were grown for 4 days and subsequently treated with the indicated concentrations of acetic acid for 200 min and stained with propidium iodide to measure cell survival. b) Normalized survival rates upon acetic acid treatment were calculated and survival integrals are shown. c) TRA1 and *tra1*_{Q3} cells were grown for the indicated time in standard synthetic complete medium with or without 0.1 M MES and stained with propidium iodide to measure cell survival. Stained cells were imaged in a 96-well plate. For each time point, a negative control (unstained cells), a positive control (boiled cells) and 5 replicates were analyzed. d) Normalized survival rates over the aging process and calculated survival integrals are shown. Significance was assessed using a 1-way ANOVA followed by a Tukey's multiple comparison test. ****P* < 0.005 and *****P* < 0.0001.

various aspects of the transcriptional response to the aging process. Interestingly, the Tra1 homolog in *Drosophila*, Nipped-A, is required to maintain proliferative capacity of intestinal stem cells through aging, indicating that Tra1 may play a role in aging across species (Tauc et al. 2017).

Caloric restriction impacts multiple stress response pathways that are associated with the aging process, such as those involved in proteotoxic stress (Choi et al. 2013; Arlia-Ciommo et al. 2018). Here, we also show that caloric restriction alleviates the accelerated aging phenotype associated with the *tra1*_{Q3} mutation. Since our data indicate that the *tra1*_{Q3} mutant is subjected to increased stress during aging, it is reasonable to postulate that caloric restriction acts at least in part by alleviating these stresses. Interestingly, while the *tra1*_{Q3} cells do not grow on glycerol, their respiratory defect appears to be less than the threshold required for lifespan extension by caloric restriction (Ocampo et al. 2012).

This study and previous work using the *tra1*_{Q3} mutant (Berg et al. 2018; Razzaq et al. 2021) support an important regulatory role for the Tra1 PI3K domain, despite the absence of key kinase motifs (McMahon et al. 1998; Helmlinger et al. 2011). Our previous work showed that Tra1_{Q3} has decreased association with SAGA and NuA components (Berg et al. 2018). This included Spt20, which is the preferential Tra1 interactor in both fission and budding yeast (Liu et al. 2019; Elías-Villalobos, Toullec, et al. 2019; Wang et al. 2020). Moreover, Tra1 regulates the incorporation of DUB module components into SAGA (Elías-Villalobos, Toullec, et al. 2019). Interestingly, we found decreased expression of DUB components in aged *tra1*_{Q3} cells. Whether this is a response to mis-assembly of the complex remains to be determined. Similarly, Leo et al. (2018) demonstrated that SAGA, through Gcn5, is essential to properly maintain Ubp8 levels under respiratory conditions. Aging also exacerbated the increase in TRA1 expression that we previously observed in *tra1*_{Q3} cells

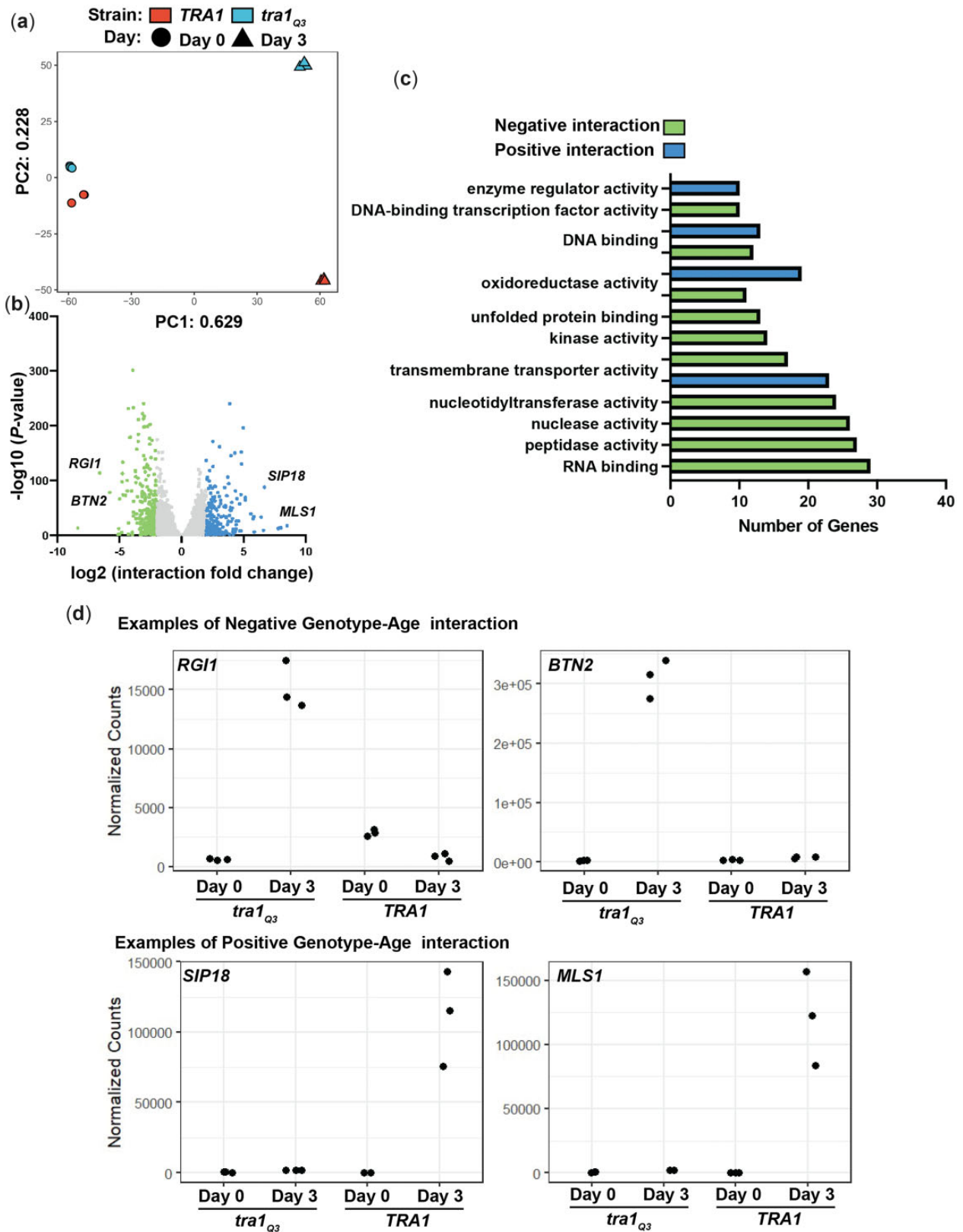


Fig. 4. The *tra1_{Q3}* mutation alters the transcriptional landscape of aging cells. a) Principal component analysis of centered log ratio of normalized reads from *TRA1* and *tra1_{Q3}* cells at day 0 and day 3. Each point represents a single biological replicate ($n = 3$). b) Volcano plot of genes that respond differently to the aging process in the *tra1_{Q3}* cells compared to wild-type *TRA1* (coloured for dark points represent $P < 0.05$, \log_2 fold change > 1). c) Significantly enriched GO biological processes were determined for genes with both positive and negative age-genotype interaction with \log_2 interaction score > 2 ($P < 0.05$) in *tra1_{Q3}* cells compared to wild type. d) Examples of genes with positive (*SIP18*, *MLS1*) and negative (*RGI1*, *BTN2*) age-genotype interaction. Normalized RNA sequencing read counts are shown for *TRA1* and *tra1_{Q3}* cells at day 0 and day 3.

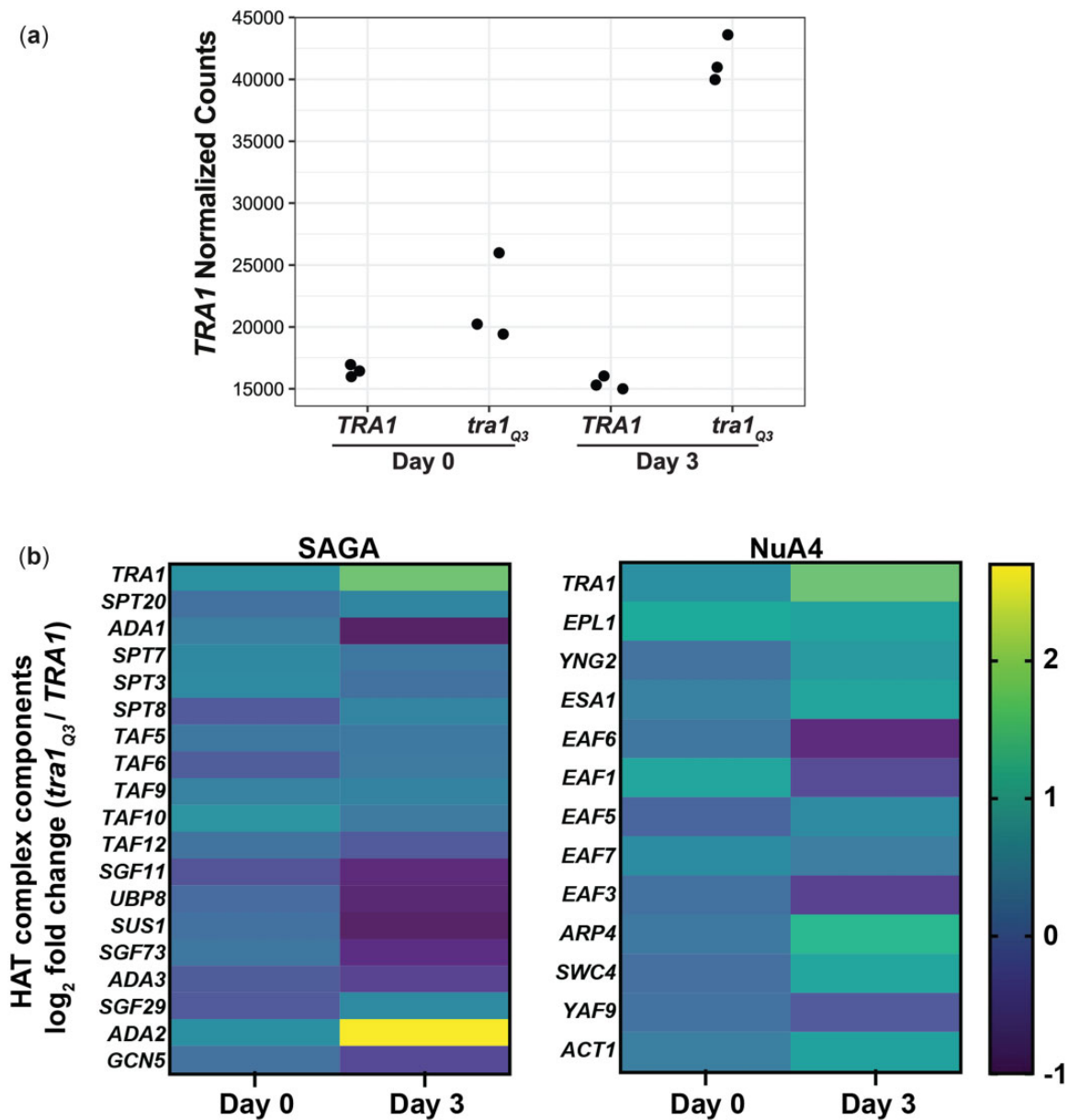


Fig. 5. Aged *tra1*_{Q3} cells display increased TRA1 mRNA abundance. a) Normalized RNA sequencing read counts are shown for TRA1 mRNA in wild-type TRA1 and *tra1*_{Q3} cells at day 0 and day 3. b) Log₂ fold change (*tra1*_{Q3}/TRA1) for the mRNA of genes encoding SAGA and NuA4 components at day 0 and day 3.

(Berg et al. 2018). The nature of this feedback mechanism remains unclear. Upregulation of TRA1 is not observed in response to deleting other components of SAGA and NuA4 (Berg et al. 2018). TRA1 expression is also upregulated upon protein misfolding stress (Jiang et al. 2019). Therefore, it is reasonable to postulate that increased proteotoxic stress associated with the aging *tra1*_{Q3} allele plays a role in this phenotype.

HAT complexes and lifespan regulation

While we show here that compromising Tra1 function shortens CLS, the role of the different components of the SAGA and NuA4 in aging is complex. Deleting GCN5 and SPT20 shortens CLS in winemaking yeast (Orozco et al. 2012; Picazo et al. 2015). Conversely, deleting the SAGA component SGF11 extends CLS (Garay et al. 2014) indicating that members of HAT complexes can differentially impact the aging process. The SAGA DUB

module also regulates replicative aging via its interaction with Sir2 (McCormick et al. 2014; Mason et al. 2017). Deletion of GCN5 and its pharmacological inhibition extends replicative lifespan (Huang et al. 2020). Therefore, different components of SAGA can differentially affect aging. Arp4 (actin-related protein 4), an essential component of the NuA4 complex, is necessary for chronological aging through its interaction with Hho1 (Miloshev et al. 2019). Thus, different components of SAGA and NuA4 can differentially affect aging.

Further studies will be required to determine the role of Tra1 and the impact of the *tra1*_{Q3} mutation on the global acetylation of both histone and non-histone substrates. Interestingly, *tra1*_{Q3} prevents upregulation of ACS1 during aging (Supplementary Table 2). ACS1 encodes a nucleocytoplasmic acetyl-CoA synthetase whose expression increases during chronological aging (Lesur and Campbell 2004; Wierman et al. 2017), presumably to

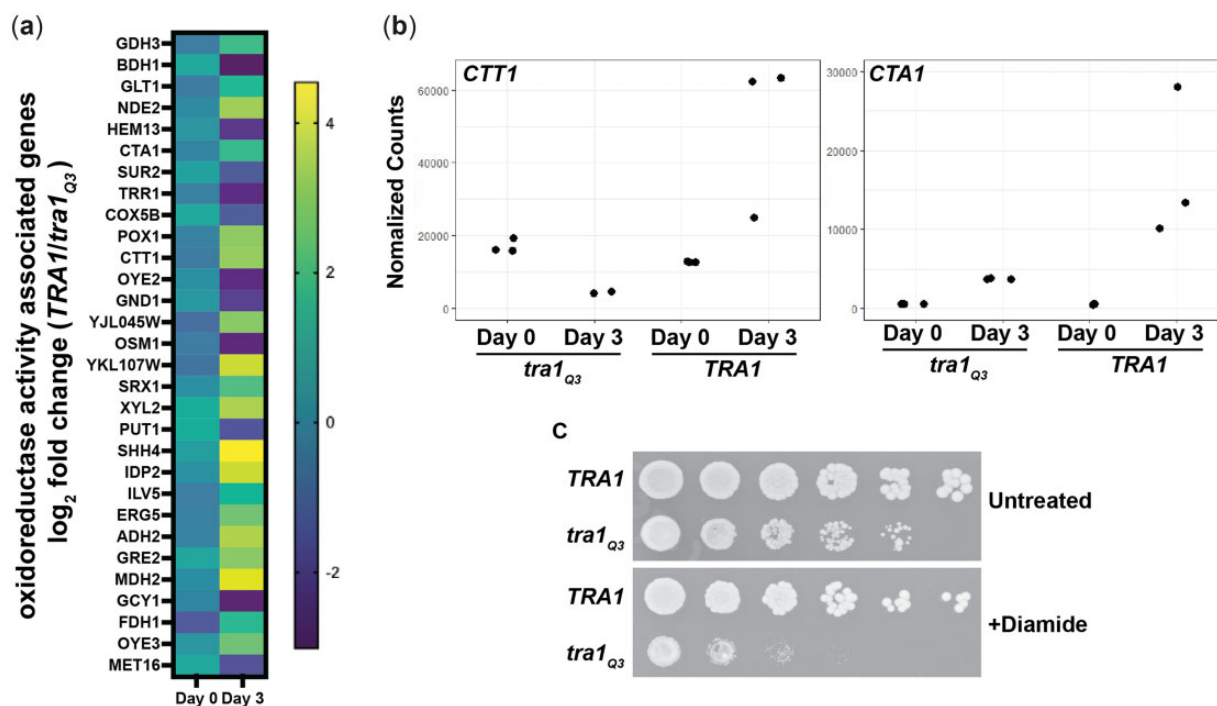


Fig. 6. *tra1_{Q3}* cells are sensitive to oxidative stress. a) Log₂ fold change (TRA1/*tra1_{Q3}*) for mRNA of genes associated with oxidoreductase activity in wild-type TRA1 and *tra1_{Q3}* cells at day 0 and day 3. b) Normalized RNA sequencing read counts are shown for CTT1 and CTA1 in TRA1 and *tra1_{Q3}* cells at day 0 and day 3. c) *tra1_{Q3}* cells are sensitive to diamide. TRA1 and *tra1_{Q3}* cells were spotted onto agar plates without (untreated) or with 1 mM diamide.

maintain the pool of acetyl-CoA required for histone acetylation (Takahashi et al. 2006). ACS1 null cells consequently display reduced CLS (Marek and Korona 2013). This is also in agreement with previous observations that cells deleted for NuA4 and SAGA components (*caf7Δ* and *gcn5Δ*) display decreased levels of acetyl-CoA (Rollins et al. 2017).

Our data also showed that *tra1_{Q3}* cells have increased sensitivity to acetic acid (Fig. 3). This is consistent with studies showing the importance of transcriptional regulation in acetic acid tolerance (Dong et al. 2017; Vall-Llaura et al. 2019; Chaves et al. 2021). A direct link to SAGA and NuA4 was suggested by Dong et al. (2017) who revealed that acetic acid treatment leads to differential expression of multiple SAGA/NuA4 components. Similar to what we observed, increased levels of ADA2 and EPL1 increase sensitivity to acetic acid (Dong et al. 2017). Nonetheless, the impact of acid stress in histone acetylation is complex and involves multiple pathways. Vall-Llaura et al. (2019) found that deacetylation by the Sir2 histone deacetylase regulates acetic acid tolerance. In addition, defects in the Set3 histone deacetylase complex are associated with increased tolerance to acid stress, suggesting that histone acetylation plays a protective role (Sousa et al. 2013). Of note, the most upregulated genes in aged *tra1_{Q3}* cells, *BTN2* and *RGI1*, are strongly upregulated by acetic acid (Dong et al. 2017). This is consistent with these cells experiencing high acid stress and potentially provides an explanation as to why buffering the growth media's pH alleviates the aging phenotype associated with the *tra1_{Q3}* mutation.

Tra1, mitochondria, peroxisomes, and the aging process

Here, we showed that several genes important for β -oxidation are decreased in *tra1_{Q3}* cells after chronological aging. During chronological aging, yeast cells utilize internal fat stores (Goldberg et al. 2009). Ultimately, free fatty acids are the substrate for peroxisomal

β -oxidation. This process allows cells to produce acetyl-CoA that is used to generate the ATP in the mitochondria required for survival at stationary phase. Thus, cells that lack mitochondrial respiration (i.e. incapable of growing on glycerol) cannot grow on oleate (Lockshon et al. 2007). In our case, the absence of growth of the *tra1_{Q3}* strain on medium containing oleate as the carbon source (Fig. 7b) could be linked to deficient mitochondrial respiration. In *S. cerevisiae*, Gcn5 and Ubp8 are also required for respiration (Canzonetta et al. 2016; Leo et al. 2018), a process essential for chronological aging (Pan et al. 2011; Ocampo et al. 2012).

We previously found that a TRA1 mutant displays negative genetic interactions with genes associated with mitochondrial function (Hoke et al. 2008). This could reflect altered regulation of the retrograde response associated with aging (Kim et al. 2004; Jazwinski 2005; Friis et al. 2014; Pogoda et al. 2021). The mitochondrial retrograde pathway signals to the nucleus via the Rtg proteins (Rtg1, 2, and 3) to upregulate genes associated with mitochondrial stress (Liao and Butow 1993; Zdravlević et al. 2015). The canonical retrograde target is *CIT2*, which encodes the peroxisomal isoform of citrate synthase. The retrograde pathway regulates expression of several other peroxisomal proteins (Chelstowska and Butow 1995). Retrograde signaling also upregulates other genes involved in the TCA cycle, such as mitochondrial citrate synthase (*CIT1*), aconitase (*ACO1*), and NAD⁺-dependent isocitrate dehydrogenase (*IDH1/2*). Interestingly, we found that these targets of the retrograde pathway are downregulated in aged *tra1_{Q3}* (Supplementary Table 2). While there is debate concerning the incorporation of Rtg2 in the SAGA-like complex (Pray-Grant et al. 2002; Adamus et al. 2021), our data suggest a role for Tra1 in the regulation of retrograde signaling that could contribute to the changes in peroxisomal gene expression observed in the aged *tra1_{Q3}* cells.

Cells carrying deletions in genes encoding other SAGA components display reduced levels of peroxisomal genes

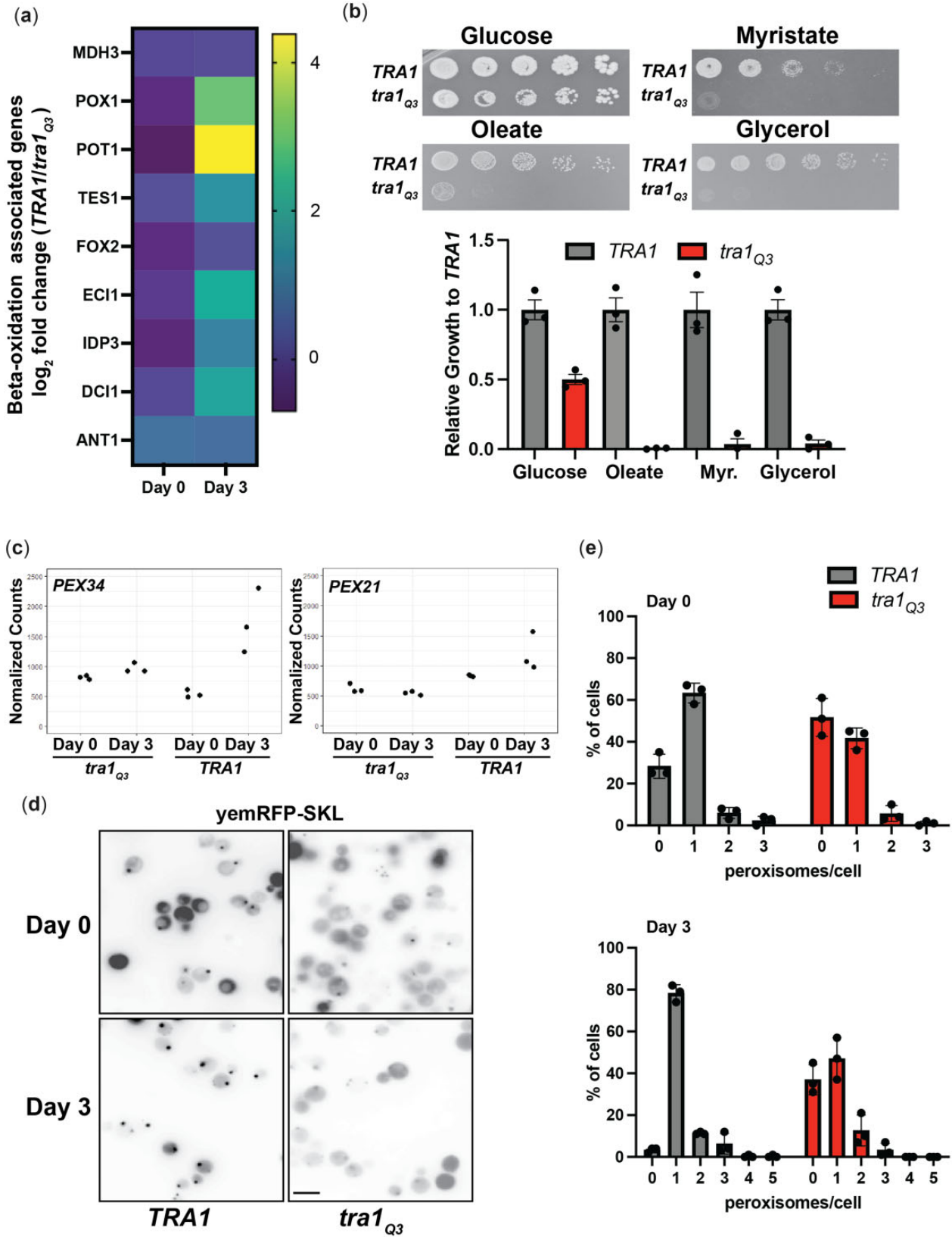


Fig. 7. *tra1_{Q3}* results in defective peroxisomes. a) Log₂ fold change (*TRA1/tra1_{Q3}*) for beta-oxidation genes at day 0 and day 3. b) *tra1_{Q3}* cells show decreased growth in the presence of oleic acid. *TRA1* and *tra1_{Q3}* cells were spotted on agar plates containing either glucose (YPD), oleate, myristate, or glycerol as the carbon source. Growth of *tra1_{Q3}* cells relative to *TRA1* cells was quantified and is shown in the bar graph. c) Normalized RNA sequencing read counts are shown for *PEX34* and *PEX21* in wild-type *TRA1* and *tra1_{Q3}* cells at day 0 and day 3. d) *TRA1* and *tra1_{Q3}* cells expressing *yemRFP-SKL* were imaged using fluorescence microscopy at day 0 and day 3 of the aging process. The number of peroxisomes/cell is shown in bar graphs. Bar: 10 μm.

(Ratnakumar and Young 2010) suggesting a further link between HAT complexes and peroxisomal biogenesis/functions. Interestingly, cells incompetent for β -oxidation have a less severe aging phenotype than cells devoid of peroxisomes, indicating that other peroxisomal functions are important in regulating CLS (Lefevre et al. 2013). Peroxisome proliferation is also associated with the early stage of replicative aging (Deb et al. 2022). Free oxidative radicals have long been suggested to regulate the aging process (Harman 1972). Peroxisomes contain catalase that metabolizes hydrogen peroxide and maintains the cellular redox balance (Lismont et al. 2015). Efficient import of catalase into the peroxisomes improves longevity in human cells (Koepeke et al. 2007). Inhibiting human peroxisomal catalase triggers increased mitochondrial reactive oxygen species (Walton and Pizzitelli 2012). In contrast, deletion and pharmacological inactivation of either form of the yeast catalase (CTT1, cytosolic; CTA1, peroxisomal), is associated with extended CLS (Mesquita et al. 2010). It was proposed that lack of catalase in young cells triggers a sublethal level of oxidative stress that allows hormetic adaptation to oxidative stress and, consequently, lifespan extension (Mesquita et al. 2010). However, overexpressing catalase also extends the CLS of cells lacking superoxide dismutase (*Asod1*) showing that catalase levels are indeed important to alleviate oxidative damage associated with aging (Rona et al. 2015). Therefore, inability of *tra1^{Q3}* cells to properly regulate catalase expression likely has an important role in chronological aging. This is supported by previous studies showing that SAGA plays a major role in the transcriptional response to oxidative stress (Huisinga and Pugh 2004; Sansó et al. 2011; Kim et al. 2019).

Perspectives and conclusions

In conclusion, our work offers insight into how Tra1 regulates gene expression associated with several stress pathways such as regulation of cell wall integrity, mitochondria respiration and peroxisomal function that are crucial for chronological aging. Given the growing evidence that chronologically aged yeast cells form a heterogeneous population of quiescent and nonquiescent cells (Allen et al. 2006; Aragon et al. 2008; Miles et al. 2013; Mohammad et al. 2020), it will be important to define how transcriptional regulators like Tra1 and other SAGA/NuA4 components regulate gene expression, histone acetylation and chromatin accessibility within the aging population.

Data availability

Strains and plasmids are available upon request. Gene expression data are available at GEO with the accession number: GSE206033.

Supplemental material is available at G3 online.

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Conflicts of interest

None declared.

Literature cited

- Adamus K, Reboul C, Voss J, Huang C, Schittenhelm RB, Le SN, Ellisdon AM, Elmlund H, Boudes M, Elmlund D, et al. SAGA and SAGA-like SLIK transcriptional coactivators are structurally and biochemically equivalent. *J Biol Chem.* 2021;296:100671.
- Agarwal S, Sharma S, Agrawal V, Roy N. Caloric restriction augments ROS defense in *S. cerevisiae*, by a Sir2p independent mechanism. *Free Radic Res.* 2005;39(1):55–62.
- Allard S, Utley RT, Savard J, Clarke A, Grant P, Brandl CJ, Pillus L, Workman JL, Côté J. NuA4, an essential transcription adaptor/histone H4 acetyltransferase complex containing Esa1p and the ATM-related cofactor Tra1p. *EMBO J.* 1999;18(18):5108–5119.
- Allen C, Büttner S, Aragon AD, Thomas JA, Meirelles O, Jaetao JE, Benn D, Ruby SW, Veenhuis M, Madeo F, et al. Isolation of quiescent and nonquiescent cells from yeast stationary-phase cultures. *J Cell Biol.* 2006;174(1):89–100.
- Aragon AD, Rodriguez AL, Meirelles O, Roy S, Davidson GS, Tapia PH, Allen C, Joe R, Benn D, Werner-Washburne M, et al. Characterization of differentiated quiescent and nonquiescent cells in yeast stationary-phase cultures. *Mol Biol Cell.* 2008;19(3):1271–1280.
- Arlia-Ciommo A, Leonov A, Piano A, Svistkova V, Titorenko VI. Cell-autonomous mechanisms of chronological aging in the yeast. *Microb Cell.* 2014;1(6):163–178.
- Arlia-Ciommo A, Leonov A, Beach A, Richard VR, Bourque SD, Burstein MT, Kyryakov P, Gomez-Perez A, Koupaki O, Feldman R, et al. Caloric restriction delays yeast chronological aging by remodeling carbohydrate and lipid metabolism, altering peroxisomal and mitochondrial functionalities, and postponing the onsets of apoptotic and liponecrotic modes of regulated cell death. *Oncotarget.* 2018;9(22):16163–16184.
- Atanassov BS, Evrard YA, Multani AS, Zhang Z, Tora L, Devys D, Chang S, Dent SYR. Gcn5 and SAGA regulate shelterin protein turnover and telomere maintenance. *Mol Cell.* 2009;35(3):352–364.
- Beach A, Richard VR, Leonov A, Burstein MT, Bourque SD, Koupaki O, Juneau M, Feldman R, Iouk T, Titorenko VI, et al. Mitochondrial membrane lipidome defines yeast longevity. *Aging.* 2013;5(7):551–574.
- Beach A, Leonov A, Arlia-Ciommo A, Svistkova V, Lutchman V, Titorenko VI. Mechanisms by which different functional states of mitochondria define yeast longevity. *Int J Mol Sci.* 2015;16(3):5528–5554.
- Berg MD, Genereaux J, Karagiannis J, Brandl CJ. The pseudokinase domain of Tra1 is required for nuclear localization and incorporation into the SAGA and NuA4 complexes. *G3 (Bethesda).* 2018;8(6):1943–1957.
- Bhaumik SR, Raha T, Aiello DP, Green MR. In vivo target of a transcriptional activator revealed by fluorescence resonance energy transfer. *Genes Dev.* 2004;18(3):333–343.
- Bird AW, Yu DY, Pray-Grant MG, Qiu Q, Harmon KE, Megee PC, Grant PA, Smith MM, Christman MF. Acetylation of histone H4 by Esa1

- is required for DNA double-strand break repair. *Nature*. 2002; 419(6905):411–415.
- Bolger AM, Lohse M, Usadel B. Trimmomatic: a flexible trimmer for Illumina sequence data. *Bioinformatics*. 2014;30(15):2114–2120.
- Booth LN, Brunet A. The aging epigenome. *Mol Cell*. 2016;62(5): 728–744.
- Bosotti R, Isacchi A, Sonnhammer EL. FAT: a novel domain in PIK-related kinases. *Trends Biochem Sci*. 2000;25(5):225–227.
- Brown CE, Howe L, Sousa K, Alley SC, Carrozza MJ, Tan S, Workman JL. Recruitment of HAT complexes by direct activator interactions with the ATM-related Tra1 subunit. *Science*. 2001; 292(5525):2333–2337.
- Burhans WC, Weinberger M. Acetic acid effects on aging in budding yeast: are they relevant to aging in higher eukaryotes? *Cell Cycle*. 2009;8(14):2300–2302.
- Burtner CR, Murakami CJ, Kennedy BK, Kaerberlein M. A molecular mechanism of chronological aging in yeast. *Cell Cycle*. 2009;8(8): 1256–1270.
- Campos SE, Avelar-Rivas JA, Garay E, Juárez-Reyes A, DeLuna A. Genomewide mechanisms of chronological longevity by dietary restriction in budding yeast. *Aging Cell*. 2018;17(3):e12749.
- Canzonetta C, Leo M, Guarino SR, Montanari A, Francisci S, Filetici P. SAGA complex and Gcn5 are necessary for respiration in budding yeast. *Biochim Biophys Acta*. 2016;1863(12):3160–3168.
- Chadwick SR, Pananos AD, Di Gregorio SE, Park AE, Etedali-Zadeh P, Duennwald ML, Lajoie P. A toolbox for rapid quantitative assessment of chronological lifespan and survival in *Saccharomyces cerevisiae*. *Traffic*. 2016;17(6):689–703.
- Chadwick SR, Lajoie P. Endoplasmic reticulum stress coping mechanisms and lifespan regulation in health and diseases. *Front Cell Dev Biol*. 2019;7:84.
- Chadwick SR, Fazio EN, Etedali-Zadeh P, Genereaux J, Duennwald ML, Lajoie P. A functional unfolded protein response is required for chronological aging in *Saccharomyces cerevisiae*. *Curr Genet*. 2020;66(1):263–277.
- Chaves SR, Rego A, Martins VM, Santos-Pereira C, Sousa MJ, Côte-Real M. Regulation of cell death induced by acetic acid in yeasts. *Front Cell Dev Biol*. 2021;9:642375.
- Chelstowska A, Butow RA. RTG genes in yeast that function in communication between mitochondria and the nucleus are also required for expression of genes encoding peroxisomal proteins. *J Biol Chem*. 1995;270(30):18141–18146.
- Cheng X, Jobin-Robitaille O, Billon P, Buisson R, Niu H, Lacoste N, Abshiru N, Côté V, Thibault P, Kron SJ, et al. Phospho-dependent recruitment of the yeast NuA4 acetyltransferase complex by MRX at DNA breaks regulates RPA dynamics during resection. *Proc Natl Acad Sci U S A*. 2018;115(40):10028–10033.
- Cheng X, Côté V, Côté J. NuA4 and SAGA acetyltransferase complexes cooperate for repair of DNA breaks by homologous recombination. *PLoS Genet*. 2021;17(7):e1009459.
- Cheung ACM, Díaz-Santín LM. Share and share alike: the role of Tra1 from the SAGA and NuA4 coactivator complexes. *Transcription*. 2019;10(1):37–43.
- Choi K-M, Kwon Y-Y, Lee C-K. Characterization of global gene expression during assurance of lifespan extension by caloric restriction in budding yeast. *Exp Gerontol*. 2013;48(12):1455–1468.
- Clarke AS, Lowell JE, Jacobson SJ, Pillus L. Esa1p is an essential histone acetyltransferase required for cell cycle progression. *Mol Cell Biol*. 1999;19(4):2515–2526.
- Deb R, Ghose S, Nagotu S. Increased peroxisome proliferation is associated with early yeast replicative ageing. *Curr Genet*. 2022; 68(2):207–225.
- De Luca V, Leo M, Cretella E, Montanari A, Saliola M, Ciaffi G, Vecchione A, Stoppacciaro A, Filetici P. Role of yUbp8 in mitochondria and hypoxia entangles the finding of human ortholog Usp22 in the glioblastoma pseudo-palisade microlayer. *Cells*. 2022;11(10):1682. <https://doi.org/10.3390/cells11101682>
- Deprez M-A, Eskes E, Wilms T, Ludovico P, Winderickx J. pH homeostasis links the nutrient sensing PKA/TORC1/Sch9 ménage-à-trois to stress tolerance and longevity. *Microb Cell*. 2018;5(3): 119–136.
- Díaz-Santín LM, Lukoyanova N, Aciyan E, Cheung ACM. Cryo-EM structure of the SAGA and NuA4 coactivator subunit Tra1 at 3.7 angstrom resolution. *eLife*. 2017;6:e28384. <https://doi.org/10.7554/eLife.28384>
- Dobin A, Davis CA, Schlesinger F, Drenkow J, Zaleski C, Jha S, Batut P, Chaisson M, Gingeras TR. STAR: ultrafast universal RNA-seq aligner. *Bioinformatics*. 2013;29(1):15–21.
- Dong Y, Hu J, Fan L, Chen Q. RNA-Seq-based transcriptomic and metabolomic analysis reveal stress responses and programmed cell death induced by acetic acid in *Saccharomyces cerevisiae*. *Sci Rep*. 2017;7:42659.
- Downey M. Non-histone protein acetylation by the evolutionarily conserved GCN5 and PCAF acetyltransferases. *Biochim Biophys Acta Gene Regul Mech*. 2021;1864(2):194608.
- Downs JA, Allard S, Jobin-Robitaille O, Javaheri A, Auger A, Bouchard N, Kron SJ, Jackson SP, Côté J. Binding of chromatin-modifying activities to phosphorylated histone H2A at DNA damage sites. *Mol Cell*. 2004;16(6):979–990.
- Eberharter A, Becker PB. Histone acetylation: a switch between repressive and permissive chromatin. Second in review series on chromatin dynamics. *EMBO Rep*. 2002;3(3):224–229. <https://doi.org/10.1093/embo-reports/kvf053>
- Edgar R, Domrachev M, Lash AE. Gene Expression Omnibus: NCBI gene expression and hybridization array data repository. *Nucleic Acids Res*. 2002;30(1):207–210.
- Elías-Villalobos A, Toullec D, Faux C, Séveno M, Helmlinger D. Chaperone-mediated ordered assembly of the SAGA and NuA4 transcription co-activator complexes in yeast. *Nat Commun*. 2019;10(1):5237.
- Elías-Villalobos A, Fort P, Helmlinger D. New insights into the evolutionary conservation of the sole PIKK pseudokinase Tra1/TRRAP. *Biochem Soc Trans*. 2019;47(6):1597–1608.
- Fabrizio P, Pozza F, Pletcher SD, Gendron CM, Longo VD. Regulation of longevity and stress resistance by Sch9 in yeast. *Science*. 2001; 292(5515):288–290.
- Fontana L, Partridge L, Longo VD. Extending healthy life span—from yeast to humans. *Science*. 2010;328(5976):321–326.
- Friis RMN, Glaves JP, Huan T, Li L, Sykes BD, Schultz MC. Rewiring AMPK and mitochondrial retrograde signaling for metabolic control of aging and histone acetylation in respiratory-defective cells. *Cell Rep*. 2014;7(2):565–574.
- Garay E, Campos SE, González de la Cruz J, Gaspar AP, Jinich A, Deluna A. High-resolution profiling of stationary-phase survival reveals yeast longevity factors and their genetic interactions. *PLoS Genet*. 2014;10(2):e1004168.
- Ghavidel A, Baxi K, Prusinkiewicz M, Swan C, Belak ZR, Eskiw CH, Carvalho CE, Harkness TA. Rapid nuclear exclusion of Hcm1 in aging leads to vacuolar alkalization and replicative senescence. *G3 (Bethesda)*. 2018;8(5):1579–1592.
- Goldberg AA, Bourque SD, Kyryakov P, Gregg C, Boukh-Viner T, Beach A, Burstein MT, Machkalyan G, Richard V, Rampersad S, et al. Effect of calorie restriction on the metabolic history of chronologically aging yeast. *Exp Gerontol*. 2009;44(9):555–571.

- Grant PA, Duggan L, Côté J, Roberts SM, Brownell JE, Candau R, Ohba R, Owen-Hughes T, Allis CD, Winston F, et al. Yeast Gcn5 functions in two multisubunit complexes to acetylate nucleosomal histones: characterization of an Ada complex and the SAGA (Spt/Ada) complex. *Genes Dev.* 1997;11(13):1640–1650.
- Grant PA, Schieltz D, Pray-Grant MG, Yates JR, Workman JL. The ATM-related cofactor Tra1 is a component of the purified SAGA complex. *Mol Cell.* 1998;2(6):863–867.
- Guaragnella N, Stirpe M, Marzulli D, Mazzoni C, Giannattasio S. Acid stress triggers resistance to acetic acid-induced regulated cell death through activation which requires in yeast. *Oxid Med Cell Longev.* 2019;2019:4651062.
- Harman D. The biologic clock: the mitochondria? *J Am Geriatr Soc.* 1972;20(4):145–147.
- Helmlinger D, Marguerat S, Villén J, Swaney DL, Gygi SP, Bähler J, Winston F. Tra1 has specific regulatory roles, rather than global functions, within the SAGA co-activator complex. *EMBO J.* 2011;30(14):2843–2852.
- Henry KW, Wyce A, Lo W-S, Duggan LJ, Emre NCT, Kao C-F, Pillus L, Shillatfard A, Osley MA, Berger SL. Transcriptional activation via sequential histone H2B ubiquitylation and deubiquitylation, mediated by SAGA-associated Ubp8. *Genes Dev.* 2003;17(21):2648–2663.
- Hill R, Lee PWK. The DNA-dependent protein kinase (DNA-PK): more than just a case of making ends meet? *Cell Cycle.* 2010;9(17):3460–3469.
- Hiltunen JK, Mursula AM, Rottensteiner H, Wierenga RK, Kastaniotis AJ, Gurvitz A. The biochemistry of peroxisomal beta-oxidation in the yeast *Saccharomyces cerevisiae*. *FEMS Microbiol Rev.* 2003;27(1):35–64.
- Hoke SMT, Guzzo J, Andrews B, Brandl CJ. Systematic genetic array analysis links the *Saccharomyces cerevisiae* SAGA/SLIK and NuA4 component Tra1 to multiple cellular processes. *BMC Genet.* 2008;9:46.
- Hu J, Wei M, Mirzaei H, Madia F, Mirisola M, Amparo C, Chagoury S, Kennedy B, Longo VD. Tor-Sch9 deficiency activates catabolism of the ketone body-like acetic acid to promote trehalose accumulation and longevity. *Aging Cell.* 2014;13(3):457–467.
- Huang B, Zhong D, Zhu J, An Y, Gao M, Zhu S, Dang W, Wang X, Yang B, Xie Z. Inhibition of histone acetyltransferase GCN5 extends lifespan in both yeast and human cell lines. *Aging Cell.* 2020;19(4):e13129.
- Huisinga KL, Pugh BF. A genome-wide housekeeping role for TFIID and a highly regulated stress-related role for SAGA in *Saccharomyces cerevisiae*. *Mol Cell.* 2004;13(4):573–585.
- Huse M, Kuriyan J. The conformational plasticity of protein kinases. *Cell.* 2002;109(3):275–282.
- Jazwinski SM. The retrograde response links metabolism with stress responses, chromatin-dependent gene activation, and genome stability in yeast aging. *Gene.* 2005;354:22–27.
- Jiang JC, Jaruga E, Repnevskaya MV, Jazwinski SM. An intervention resembling caloric restriction prolongs life span and retards aging in yeast. *FASEB J.* 2000;14(14):2135–2137.
- Jiang Y, Berg MD, Genereaux J, Ahmed K, Duennwald ML, Brandl CJ, Lajoie P. Sfp1 links TORC1 and cell growth regulation to the yeast SAGA-complex component Tra1 in response to polyQ proteotoxicity. *Traffic.* 2019;20(4):267–283.
- Kaeberlein M, McVey M, Guarente L. The SIR2/3/4 complex and SIR2 alone promote longevity in *Saccharomyces cerevisiae* by two different mechanisms. *Genes Dev.* 1999;13(19):2570–2580.
- Kaeberlein M. Lessons on longevity from budding yeast. *Nature.* 2010;464(7288):513–519.
- Kane AE, Sinclair DA. Epigenetic changes during aging and their reprogramming potential. *Crit Rev Biochem Mol Biol.* 2019;54(1):61–83.
- Keith CT, Schreiber SL. PIK-related kinases: DNA repair, recombination, and cell cycle checkpoints. *Science.* 1995;270(5233):50–51.
- Kim S, Ohkuni K, Couplan E, Jazwinski SM. The histone acetyltransferase GCN5 modulates the retrograde response and genome stability determining yeast longevity. *Biogerontology.* 2004;5(5):305–316.
- Kim M, Choi Y, Kim H, Lee D. SAGA DUBm-mediated surveillance regulates prompt export of stress-inducible transcripts for proteostasis. *Nat Commun.* 2019;10(1):2458.
- Kingston RE, Bunker CA, Imbalzano AN. Repression and activation by multiprotein complexes that alter chromatin structure. *Genes Dev.* 1996;10(8):905–920. <https://doi.org/10.1101/gad.10.8.905>
- Knutson BA, Hahn S. Domains of Tra1 important for activator recruitment and transcription coactivator functions of SAGA and NuA4 complexes. *Mol Cell Biol.* 2011;31(4):818–831.
- Koepke JI, Nakrieko K-A, Wood CS, Boucher KK, Terlecky LJ, Walton PA, Terlecky SR. Restoration of peroxisomal catalase import in a model of human cellular aging. *Traffic.* 2007;8(11):1590–1600.
- Laun P, Rinnerthaler M, Bogengruber E, Heeren G, Breitenbach M. Yeast as a model for chronological and reproductive aging—a comparison. *Exp Gerontol.* 2006;41(12):1208–1212.
- Lefevre SD, van Roermund CW, Wanders RJA, Veenhuis M, van der Klei IJ. The significance of peroxisome function in chronological aging of *Saccharomyces cerevisiae*. *Aging Cell.* 2013;12(5):784–793.
- Leo M, Fanelli G, Vito SD, Traversetti B, Greca ML. Ubiquitin protease Ubp8 is necessary for *S. cerevisiae* respiration. *Biochim Biophys Acta Mol Cell Res.* 2018;1865(10):1491–1500. <https://doi.org/10.1016/j.bbamcr.2018.07.025>
- Leonov A, Feldman R, Piano A, Arlia-Ciommo A, Lutchman V, Ahmadi M, Elsaser S, Fakim H, Heshmati-Moghaddam M, Hussain A, et al. Caloric restriction extends yeast chronological lifespan via a mechanism linking cellular aging to cell cycle regulation, maintenance of a quiescent state, entry into a non-quiescent state and survival in the non-quiescent state. *Oncotarget.* 2017;8(41):69328–69350.
- Lesur I, Campbell JL. The transcriptome of prematurely aging yeast cells is similar to that of telomerase-deficient cells. *Mol Biol Cell.* 2004;15(3):1297–1312.
- Liao X, Butow RA. RTG1 and RTG2: two yeast genes required for a novel path of communication from mitochondria to the nucleus. *Cell.* 1993;72(1):61–71.
- Liao Y, Smyth GK, Shi W. featureCounts: an efficient general purpose program for assigning sequence reads to genomic features. *Bioinformatics.* 2014;30(7):923–930.
- Lin Y-y, Qi Y, Lu J-y, Pan X, Yuan DS, Zhao Y, Bader JS, Boeke JD. A comprehensive synthetic genetic interaction network governing yeast histone acetylation and deacetylation. *Genes Dev.* 2008;22(15):2062–2074.
- Lin L, Chamberlain L, Zhu LJ, Green MR. Analysis of Gal4-directed transcription activation using Tra1 mutants selectively defective for interaction with Gal4. *Proc Natl Acad Sci U S A.* 2012;109(6):1997–2002.
- Lismont C, Nordgren M, Van Veldhoven PP, Fransen M. Redox interplay between mitochondria and peroxisomes. *Front Cell Dev Biol.* 2015;3:35.
- Liu G, Zheng X, Guan H, Cao Y, Qu H, Kang J, Ren X, Lei J, Dong M-Q, Li X, et al. Architecture of *Saccharomyces cerevisiae* SAGA complex. *Cell Discov.* 2019;5(1):
- Lockshon D, Surface LE, Kerr EO, Kaeberlein M, Kennedy BK. The sensitivity of yeast mutants to oleic acid implicates the

- peroxisome and other processes in membrane function. *Genetics*. 2007;175(1):77–91.
- Longo VD. Mutations in signal transduction proteins increase stress resistance and longevity in yeast, nematodes, fruit flies, and mammalian neuronal cells. *Neurobiol Aging*. 1999;20(5):479–486.
- Longo VD, Fabrizio P. Chronological aging in *Saccharomyces cerevisiae*. *Subcell Biochem*. 2012;57:101–121.
- Love MI, Huber W, Anders S. Moderated estimation of fold change and dispersion for RNA-seq data with DESeq2. *Genome Biol*. 2014;15(12):550.
- MacLean M, Harris N, Piper PW. Chronological lifespan of stationary phase yeast cells; a model for investigating the factors that might influence the ageing of postmitotic tissues in higher organisms. *Yeast*. 2001;18(6):499–509.
- Marek A, Korona R. Restricted pleiotropy facilitates mutational erosion of major life-history traits. *Evolution*. 2013;67(11):3077–3086.
- Mason AG, Garza RM, McCormick MA, Patel B, Kennedy BK, Pillus L, La Spada AR. The replicative lifespan-extending deletion of SGF73 results in altered ribosomal gene expression in yeast. *Aging Cell*. 2017;16(4):785–796.
- Matecic M, Smith DL, Pan X, Maqani N, Bekiranov S, Boeke JD, Smith JS. A microarray-based genetic screen for yeast chronological aging factors. *PLoS Genet*. 2010;6(4):e1000921.
- McCormick MA, Mason AG, Guyenet SJ, Dang W, Garza RM, Ting MK, Moller RM, Berger SL, Kaerberlein M, Pillus L, et al. The SAGA histone deubiquitinase module controls yeast replicative lifespan via Sir2 interaction. *Cell Rep*. 2014;8(2):477–486.
- McMahon SB, Van Buskirk HA, Dugan KA, Copeland TD, Cole MD. The novel ATM-related protein TRRAP is an essential cofactor for the c-Myc and E2F oncoproteins. *Cell*. 1998;94(3):363–374.
- Medkour Y, Titorenko VI. Mitochondria operate as signaling platforms in yeast aging. *Aging*. 2016;8(2):212–213.
- Mesquita A, Weinberger M, Silva A, Sampaio-Marques B, Almeida B, Leão C, Costa V, Rodrigues F, Burhans WC, Ludovico P, et al. Caloric restriction or catalase inactivation extends yeast chronological lifespan by inducing H₂O₂ and superoxide dismutase activity. *Proc Natl Acad Sci U S A*. 2010;107(34):15123–15128.
- Miles S, Li L, Davison J, Breeden LL. Xbp1 directs global repression of budding yeast transcription during the transition to quiescence and is important for the longevity and reversibility of the quiescent state. *PLoS Genet*. 2013;9(10):e1003854.
- Miloshev G, Staneva D, Uzunova K, Vasileva B, Draganova-Filipova M, Zagorchev P, Georgieva M. Linker histones and chromatin remodelling complexes maintain genome stability and control cellular ageing. *Mech Ageing Dev*. 2019;177:55–65.
- Mirisola MG, Longo VD. Yeast chronological lifespan: longevity regulatory genes and mechanisms. *Cells*. 2022;11(10):1714. <https://doi.org/10.3390/cells11101714>
- Mohammad K, Baratang Junio JA, Tafakori T, Orfanos E, Titorenko VI. Mechanisms that link chronological aging to cellular quiescence in budding yeast. *Int J Mol Sci*. 2020;21(13):4717. <https://doi.org/10.3390/ijms21134717>
- Monteiro PT, Oliveira J, Pais P, Antunes M, Palma M, Cavalheiro M, Galocha M, Godinho CP, Martins LC, Bourbon N, et al. YEASTRACT: a portal for cross-species comparative genomics of transcription regulation in yeasts. *Nucleic Acids Res*. 2020;48(D1):D642–D649.
- Mordes DA, Glick GG, Zhao R, Cortez D. TopBP1 activates ATR through ATRIP and a PIKK regulatory domain. *Genes Dev*. 2008;22(11):1478–1489.
- Morgan MT, Haj-Yahya M, Ringel AE, Bandi P, Brik A, Wolberger C. Structural basis for histone H2B deubiquitination by the SAGA DUB module. *Science*. 2016;351(6274):725–728.
- Mortimer RK, Johnston JR. Life span of individual yeast cells. *Nature*. 1959;183(4677):1751–1752.
- Mumberg D, Müller R, Funk M. Yeast vectors for the controlled expression of heterologous proteins in different genetic backgrounds. *Gene*. 1995;156(1):119–122.
- Murakami CJ, Burtner CR, Kennedy BK, Kaerberlein M. A method for high-throughput quantitative analysis of yeast chronological life span. *J Gerontol A: Biol Sci Med Sci*. 2008;63(2):113–121.
- Mutiu AI, Hoke SMT, Genereaux J, Hannam C, MacKenzie K, Jobin-Robitaille O, Guzzo J, Côté J, Andrews B, Haniford DB, et al. Structure/function analysis of the phosphatidylinositol-3-kinase domain of yeast tra1. *Genetics*. 2007;177(1):151–166.
- Nelson JD, Denisenko O, Bomsztyk K. Protocol for the fast chromatin immunoprecipitation (ChIP) method. *Nat Protoc*. 2006;1(1):179–185.
- Ning Z, Guo X, Liu X, Lu C, Wang A, Wang X, Wang W, Chen H, Qin W, Liu X, et al. USP22 regulates lipidome accumulation by stabilizing PPAR γ in hepatocellular carcinoma. *Nat Commun*. 2022;13(1):2187.
- Ocampo A, Liu J, Schroeder EA, Shadel GS, Barrientos A. Mitochondrial respiratory thresholds regulate yeast chronological life span and its extension by caloric restriction. *Cell Metab*. 2012;16(1):55–67.
- Orozco H, Matallana E, Aranda A. Wine yeast sirtuins and Gcn5p control aging and metabolism in a natural growth medium. *Mech Ageing Dev*. 2012;133(5):348–358.
- Pan Y, Schroeder EA, Ocampo A, Barrientos A, Shadel GS. Regulation of yeast chronological life span by TORC1 via adaptive mitochondrial ROS signaling. *Cell Metab*. 2011;13(6):668–678.
- Pan Y. Mitochondria, reactive oxygen species, and chronological aging: a message from yeast. *Exp Gerontol*. 2011;46(11):847–852.
- Pavletich NP, Yang H. mTOR kinase structure, mechanism and regulation. *Nature*. 2013;497:217–223.
- Petriv OI, Rachubinski RA. Lack of peroxisomal catalase causes a progeric phenotype in *Caenorhabditis elegans*. *J Biol Chem*. 2004;279(19):19996–20001.
- Petrovskiy AA, Tauro MG, Lajoie P, Duennwald ML. A quantitative imaging-based protocol for yeast growth and survival on agar plates. *STAR Protoc*. 2020;1(3):100182.
- Petrova VY, Drescher D, Kujumdzieva AV, Schmitt MJ. Dual targeting of yeast catalase A to peroxisomes and mitochondria. *Biochem J*. 2004;380(Pt 2):393–400.
- Picazo C, Orozco H, Matallana E, Aranda A. Interplay among Gcn5, Sch9 and mitochondria during chronological aging of wine yeast is dependent on growth conditions. *PLoS One*. 2015;10(2):e0117267.
- Pogoda E, Tutaj H, Pirog A, Tomala K, Korona R. Overexpression of a single ORF can extend chronological lifespan in yeast if retrograde signaling and stress response are stimulated. *Biogerontology*. 2021;22(4):415–427.
- Pray-Grant MG, Schieltz D, McMahon SJ, Wood JM, Kennedy EL, Cook RG, Workman JL, Yates JR, Grant PA. The novel SLIK histone acetyltransferase complex functions in the yeast retrograde response pathway. *Mol Cell Biol*. 2002;22(24):8774–8786.
- Prokakis E, Dyas A, Grün R, Fritzsche S, Bedi U, Kazerouni ZB, Kosinsky RL, Johnsen SA, Wegwitz F. USP22 promotes HER2-driven mammary carcinoma aggressiveness by suppressing the unfolded protein response. *Oncogene*. 2021;40(23):4004–4018.
- Purdue PE, Yang X, Lazarow PB. Pex18p and Pex21p, a novel pair of related peroxins essential for peroxisomal targeting by the PTS2 pathway. *J Cell Biol*. 1998;143(7):1859–1869.
- Rando OJ, Winston F. Chromatin and transcription in yeast. *Genetics*. 2012;190(2):351–387.

- Ratnakumar S, Young ET. Snf1 dependence of peroxisomal gene expression is mediated by Adr1. *J Biol Chem*. 2010;285(14):10703–10714.
- Razzaq I, Berg MD, Jiang Y, Genereaux J, Uthayakumar D, Kim GH, Agyare-Tabbi M, Halder V, Brandl CJ, Lajoie P, et al. The SAGA and NuA4 component Tra1 regulates *Candida albicans* drug resistance and pathogenesis. *Genetics*. 2021;219(2). <https://doi.org/10.1093/genetics/iyab131>
- Roberts SM, Winston F. Essential functional interactions of SAGA, a *Saccharomyces cerevisiae* complex of Spt, Ada, and Gcn5 proteins, with the Snf/Swi and Srb/Mediator complexes. *Genetics*. 1997; 147(2):451–465.
- Rollins M, Huard S, Morettin A, Takuski J, Pham TT, Fullerton MD, Côté J, Baetz K. Lysine acetyltransferase NuA4 and acetyl-CoA regulate glucose-deprived stress granule formation in *Saccharomyces cerevisiae*. *PLoS Genet*. 2017;13(2):e1006626.
- Rona G, Herdeiro R, Mathias CJ, Torres FA, Pereira MD, Eleutherio E. CTT1 overexpression increases life span of calorie-restricted *Saccharomyces cerevisiae* deficient in Sod1. *Biogerontology*. 2015; 16(3):343–351.
- Saleh A, Schieltz D, Ting N, McMahon SB, Litchfield DW, Yates JR, Lees-Miller SP, Cole MD, Brandl CJ. Tra1p is a component of the yeast Ada.Spt transcriptional regulatory complexes. *J Biol Chem*. 1998;273(41):26559–26565.
- Sansó M, Vargas-Pérez I, Quintales L, Antequera F, Ayté J, Hidalgo E. Gcn5 facilitates Pol II progression, rather than recruitment to nucleosome-depleted stress promoters, in *Schizosaccharomyces pombe*. *Nucleic Acids Res*. 2011;39(15):6369–6379.
- Sardi M, Gasch AP. Genetic background effects in quantitative genetics: gene-by-system interactions. *Curr Genet*. 2018;64(6):1173–1176.
- Schneider CA, Rasband WS, Eliceiri KW. NIH Image to ImageJ: 25 years of image analysis. *Nat Methods*. 2012;9(7):671–675.
- Seah TC, Kaplan JG. Purification and properties of the catalase of bakers' yeast. *J Biol Chem*. 1973;248(8):2889–2893.
- Sharov G, Voltz K, Durand A, Kolesnikova O, Papai G, Myasnikov AG, Dejaegere A, Ben Shem A, Schultz P. Structure of the transcription activator target Tra1 within the chromatin modifying complex SAGA. *Nat Commun*. 2017;8(1):1556.
- Shiloh Y, Ziv Y. The ATM protein kinase: regulating the cellular response to genotoxic stress, and more. *Nat Rev Mol Cell Biol*. 2013; 14(4):197–210.
- Smith GCM, Jackson SP. The PIKK family of protein kinases. In: Bradshaw R, Dennis E, editors. *Handbook of Cell Signaling*. Academic Press; 2003. p. 557–561.
- Smith EN, Kruglyak L. Gene-environment interaction in yeast gene expression. *PLoS Biol*. 2008;6(4):e83.
- Sousa M, Duarte AM, Fernandes TR, Chaves SR, Pacheco A, Leão C, Côrte-Real M, Sousa MJ. Genome-wide identification of genes involved in the positive and negative regulation of acetic acid-induced programmed cell death in *Saccharomyces cerevisiae*. *BMC Genomics*. 2013;14:838.
- Steinkraus KA, Kaeberlein M, Kennedy BK. Replicative aging in yeast: the means to the end. *Annu Rev Cell Dev Biol*. 2008;24:29–54.
- Steunou A-L, Rossetto D, Côté J. Regulating chromatin by histone acetylation. In: Workman J, Abmayr S, editors. *Fundam Chromatin*. New York (NY): Springer. 2014;147–212.
- Takahashi H, McCaffery JM, Irizarry RA, Boeke JD. Nucleocytoplasmic acetyl-coenzyme a synthetase is required for histone acetylation and global transcription. *Mol Cell*. 2006;23(2):207–217.
- Tauc HM, Tasdogan A, Meyer P, Pandur P. Nipped-A regulates intestinal stem cell proliferation in *Drosophila*. *Development*. 2017; 144(4):612–623.
- Taylor SS, Kornev AP. Protein kinases: evolution of dynamic regulatory proteins. *Trends Biochem Sci*. 2011;36(2):65–77.
- Tissenbaum HA, Guarente L. Model organisms as a guide to mammalian aging. *Dev Cell*. 2002;2(1):9–19.
- Tower RJ, Fagarasanu A, Aitchison JD, Rachubinski RA. The peroxin Pex34p functions with the Pex11 family of peroxisomal divisional proteins to regulate the peroxisome population in yeast. *Mol Biol Cell*. 2011;22(10):1727–1738.
- Vall-Llaura N, Mir N, Garrido L, Vived C, Cabisco E. Redox control of yeast Sir2 activity is involved in acetic acid resistance and longevity. *Redox Biol*. 2019;24:101229.
- Vasileva B, Staneva D, Krasteva N, Miloshev G, Georgieva M. Changes in chromatin organization eradicate cellular stress resilience to UVA/B light and induce premature aging. *Cells*. 2021; 10(7):1755. <https://doi.org/10.3390/cells10071755>
- Walton PA, Pizzitelli M. Effects of peroxisomal catalase inhibition on mitochondrial function. *Front Physiol*. 2012;3:108.
- Wang H, Dienemann C, Stützer A, Urlaub H, Cheung ACM, Cramer P. Structure of the transcription coactivator SAGA. *Nature*. 2020; 577(7792):717–720.
- Wierman MB, Maqani N, Strickler E, Li M, Smith JS. Caloric restriction extends yeast chronological life span by optimizing the Snf1 (AMPK) signaling pathway. *Mol Cell Biol*. 2017;37. <https://doi.org/10.1128/MCB.00562-16>
- Yamauchi Y, Izawa S. Prioritized expression of BTN2 of *Saccharomyces cerevisiae* under pronounced translation repression induced by severe ethanol stress. *Front Microbiol*. 2016;7:1319.
- Ždravčić M, Guaragnella N, Giannattasio S. Yeast as a tool to study mitochondrial retrograde pathway en route to cell stress response. *Methods Mol Biol*. 2015;1265:321–331.

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