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Review article

Cellular responses of *Saccharomyces Cerevisiae* Against Arsenic

Saccharomyces Cerevisiae'nin Arsenik'e Karşı Hücresel Tepkileri

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Abstract

Arsenic is a metalloid member of heavy metals associated with many health problems from various cancers to skin diseases. Due to mankind activities and natural sources, arsenic contamination seen globally. More than 150 million people globally face with arsenic via arsenic polluted ground water. It is well known that speciation of arsenic is important for its actions inside of the exposed organism. *Saccharomyces cerevisiae*, is one of six model organisms, provides an general answer for the question "What eukaryotes do?". So assessing some questions on budding yeast gives a general idea about potential results in other eukaryotes including human. One of the issues investigated on this yeast is that impacts and metabolism of arsenic. Arsenic is well studied on *Saccharomyces cerevisiae* and consequently much data became available. In this review, cellular impacts of arsenic and response of the yeast towards arsenic exposure is covered.

Keywords: *Saccharomyces cerevisiae*, Arsenic, Signaling Pathways, Cytotoxicity, Protein Aggregation.

Özet

Arsenik, çeşitli kanserlerden cilt hastalıklarına kadar birçok sağlık problemiyle bağlantılı ağır metallerin bir metaloid üyesidir. İnsanlık faaliyetleri ve doğal kaynaklar nedeniyle, küresel olarak arsenik kirliliği görülmektedir. Dünyada 150 milyondan fazla insan arsenik kirli yeraltı suyu ile arsenikle karşı karşıya kalmaktadır. Hangi arsenik türüne maruz kalındığı arseniğin organizmanın içindeki eylemleri için önemli olduğu iyi bilinmektedir. Saccharomyces cerevisiae, altı model organizmadan biridir, "Ökaryotlar ne yapar?" sorusuna genel bir cevap sağlar. Bu nedenle, bazı soruların bu maya üzerinde değerlendirilmesi, insan dahil diğer ökaryotlardaki potansiyel sonuçlar hakkında genel bir fikir verir. Bu mayada araştırılan konulardan biri arseniğin etki ve metabolizmasıdır. Arsenik, Saccharomyces cerevisiae' da iyi çalışılmıştır ve sonuç olarak birçok veri mevcut olmuştur. Bu derlemede, arseniğin hücresel etkileri ve mayanın arsenik maruziyetine tepkisi ele alınmaktadır.

Anahtar Kelimeler: *Saccharomyces cerevisiae*, arsenik, sinyal yolakları, sitotoksisite, protein aggregasyonu.

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INTRODUCTION

There are two main approaches accepted to explain the term "heavy metal". One of them is based on density while the other one utilizes atomic mass as the major criteria. Explanations employed density as primary factor differ each other via using distinct threshold numbers of density so as to assess an element as "heavy" . Description based on atomic mass accepts mass of calcium (Ca) atom as minimum. According to Bánfalvi, (2011), 3 g/cm^3 employed as a threshold density (Bánfalvi, 2011). Heavy metals can be grouped under two major category according to biology, namely biologically essentials and non-essentials. Essential heavy metals are required for the normal functioning of cells. In high doses, essentials ones are harmful as even low doses of non-essential heavy metals are (Bánfalvi, 2011).

Physicochemical characteristics and ligand choice of metal are main determinants of its toxicity. While metals like cadmium (Cd) and mercury (Hg) are classified as "soft" transition metals, chromium (Cr), manganese (Mn), arsenic (As), antimony (Sb) and selenium (Se) are categorized as "hard". Hard ones prefer oxygen in their higher oxidation states and sulfur in their lower oxidation states as their ligand. However, soft transition metals favor sulfur in terms of their ligand preference. Lead (Pb), iron (Fe), cobalt (Co), nickel (Ni), copper (Cu) and zinc (Zn) may use oxygen, sulfur or nitrogen as ligands (Da Silva & Williams, 2001; Lemire et al., 2013).

Source of heavy metal pollution is not always mankind activities but also natural geology. As a soft heavy metal/metalloid arsenic is globally famous because of its pollution in tap water which makes this metalloid available for each consumer of the contaminated drinking water (Bhattacharya et al., 2002; Halem et al., 2009). As cells of lung, colon, prostate and skin constantly face with arsenic, they can be transformed into cancerous tissues (Kim et al., 2011). As compared to others, most frequently skin cells are giving rise to malignant cancers when exposure to the metalloid occurs via oral route (Nathaniel, 2005; Smith et al., 1992). If rate of risk of external organs to become malignant are ignored, bladder cells remains to be the one experiencing the utmost risk (Chu & Crawford-Brown, 2006). Moreover, cancers of kidney, liver, and prostate found to relate with exposure to the metalloid (Bates et al., 1992; Chen et al., 1992). It is revealed, when the metalloid exposure occurs at antenatal period, the risk of emergence of negative impacts arises at the period of childhood. Major underlying mechanisms of these negative impacts include reduction in DNA methylation, endocrinological changes, immune suppression, neural toxicity and alteration in enzymes significant for development of fetus (Vahter, 2008).

Heavy metals, especially arsenic and cadmium is well studied on *Saccharomyces cerevisiae* (Tamás, et al., 2018; Tamás, et al., 2014). In the following text accumulated scientific data about impacts of arsenic on budding yeast, at molecular level, is presented.

Arsenic and *Saccharomyces cerevisiae*

Livings face with arsenic mostly in the forms of As (V), AsO_4^{3-}, As (III), $As(OH)_3$. As it is easily seen, prevalent species are pentavalent and trivalent ones (Wysocki & Tamás, 2011). AsO_4^{3-} creates complexes with phosphate and thus acts on phosphate transportation, metabolism and phosphate dependent signaling machineries. Arsenic compounds known to interfere with cellular events occurring in mitochondria (Ralph, 2008; Thorsen et al., 2009; Vujcic et al., 2007) as it can be concluded from the inhibitory effect of As (V) on ATP production (Cortés et al., 2000). As (III) is thought to be more hazardous than As (V) as As (III) is able to connect with -SH sites which influences structural and/or functional characteristics of proteins. Moreover, oxidative alterations sourced from As (III) presence affects these characteristics, too (Aposhian & Aposhian, 2006; Kitchin & Wallace, 2008). As an instance for As (III) induced protein damage, hazards on actin and tubilin, basic cytoskeleton elements, can be shown (Thorsen et al., 2009). In addition, As (III) triggers P-body and stress granule formation (Jacobson et al., 2012).

Arsenic in solution commonly found as $As(OH)_3$ (Ramírez-Solís et al., 2004) whose structure is similar to glycerol (Porquet & Filella, 2007). Based on the structural similarity, it is suggested that $As(OH)_3$ can be recognized as a substrate by Fps1, a yeast aquagyceroporin, like it is done for glycerol (**Fig. 1**). This condition also applies for Sb (III) whose general form in neutral pH is $Sb(OH)_3$ (Wysocki & Tamás, 2011). As (III) competes with glucose to be transported via hexose permeases (Hxt) (**Fig. 1**). When glucose is absent, expression rate of this type of permeases rises thus even Fps1 lacking cells bioaccumulate As (III) in large amounts. It is hypothesized that the enzyme recognize arsenic as a substrate when three $As(OH)_3$ gathers to construct a six membered ring species structurally similar to hexoses (Liu et al., 2004). Due to high glucose contented natural environment of yeast, expression of these permeases kept at their normal level. For this reason, instead of hexose permeases, Fps1 seems to be prevalent transporter of As (III)/Sb (III) as physiological conditions are supplied (Liu et al., 2006).

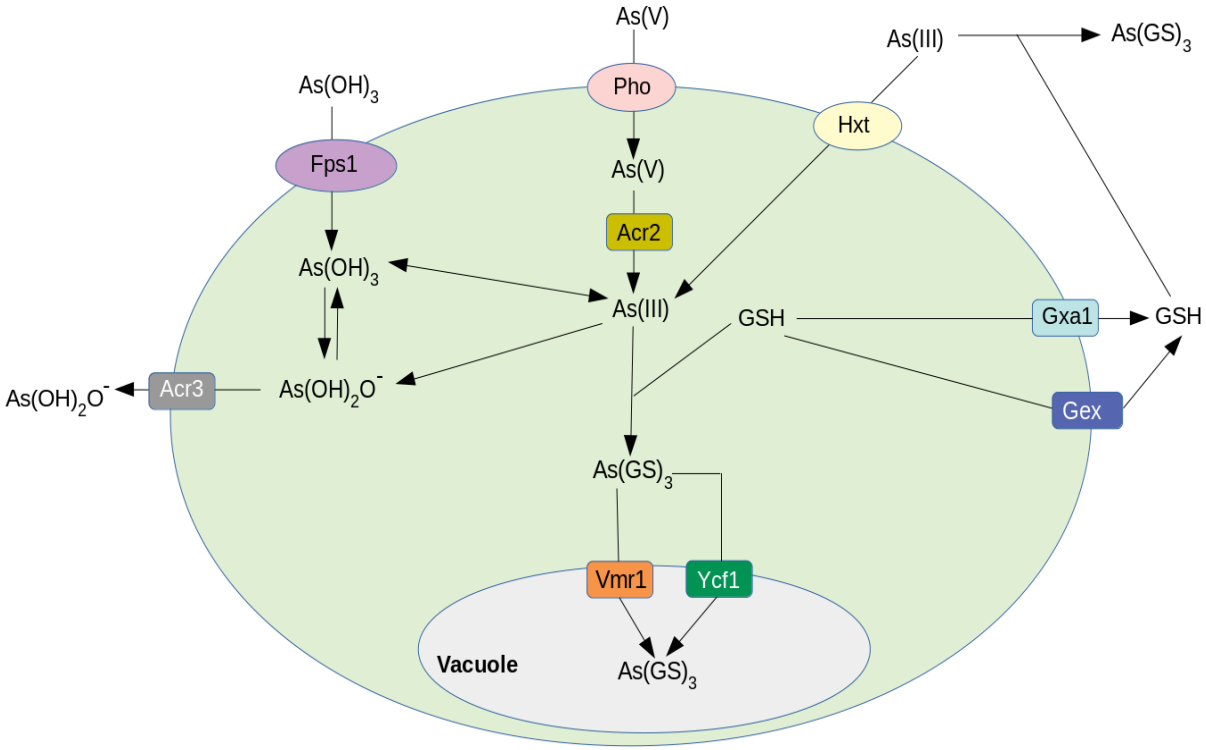

Fig. 1 This figure describes arsenic detoxification routes of yeast *Saccharomyces cerevisiae* (inspired from Maciaszczyk-Dziubinska et al., 2012; Wysocki & Tamás, 2011 ; Kiriyama et al., 2012). Hxt: Hxt1-Hxt17, Gal2; Pho: Pho84p and Pho87p

As demonstrated in **Fig. 1**, As(V) is transported into cell by means of phosphate transporters. There are five determined phosphate transporters (Pho) for *Saccharomyces cerevisiae*, namely Pho84, Pho87, Pho89, Pho90 and Pho91. While Pho84 and Pho89 have high affinity, the other three of five have low affinity (Maciaszczyk-Dziubinska et al., 2012). Leastways, Pho84 and Pho87 are found to be responsible for influx of As (V) (Bun-ya et al., 1996; Yompakdee et al., 1996). After As (V) reaches cytoplasm, it encounters with the arsenate reductase Acr2, and converted to As (III) by the action of Acr2. GSH (glutathione) is utilized as electron donor for the reduction reaction (Mukhopadhyay & Rosen, 1998; Mukhopadhyay et al., 2000). Converted As (III) has two main route, it can generate complex with GSH and the complex directed to be sequestered into the vacuole via Ycf1 and Vmr1 (Maciaszczyk-Dziubinska et al., 2012; Wysocki & Tamás, 2011) or it could be extracted to extracellular environment via the activity of Acr3, an arsenite permeases. Acr3 is reported to be one of the most significant arsenic detoxification elements of *Saccharomyces cerevisiae* (Ghosh et al., 1999; Wysocki et al., 1997) as well as of some other fungi and some prokaryotes (Aaltonen & Silow, 2008; Fu et al., 2009). In addition to all of these, *Saccharomyces cerevisiae* is able to prevent As (III) entry by extracellular chelation via using GSH. Formed GS-As (III) complex lacks the ability to enter into the cells (Wysocki & Tamás, 2011). Together with Gex1/Gex2, Gax1 is reported to be responsible for efflux

of GSH to the environment (Kiriyama et al., 2012). When exposure time is long, most of As (III) is chelated by extracellular agents like GSH, amount of free As (III) outside of the cell become less than intracellular As (III) thus Fps1 channel pumps $As(OH)_3$ towards the environment and, again as a consequence of long exposure time, Fps1 is upregulated (Wysocki & Tamás, 2011).

Arsenic exposure known to provide activation of Yap1 in the nucleus, by an unknown mechanism, where it promotes transcription of oxidative stress tolerance genes (Wysocki & Tamás, 2011). Among these genes, *GSH1* (Glutathione Synthase), *GLR1* (Glutathione Reductase), *TRX2* (Thioredoxin) and *TRR1* (Thioredoxin Reductase) are found (Kuge & Jones, 1994; Jaekwon Lee et al., 1999; Morgan et al., 1997). Yap1 is not the only transcription factor manages tolerance against As exposure but also Yap8 stands for transcription of both *ACR2* and *ACR3* genes which provide detoxification of arsenic via distinct ways. It is modulated at the level of protein (Wysocki & Tamás, 2011). In the presence of arsenic, it is stabilized and transcription of *ACR2* and *ACR3* occurs. However in the absence of arsenic, Yap8 remains unstable and targeted by Ubc4 dependent degradation mechanism (Di & Tamás, 2007). Yap8 senses the presence of As (III) via directly binding to As (III) (Kumar et al., 2016). When it is activated, it promotes employment of Mediator complex to the *ACR2/ACR3* promoter by interacting with the tail subunit Med2 of Mediator. The Mediator complex recruits elements of core transcriptional machinery involving TBP. However, these actions are not sufficient to invoke upregulation of *ACR2*. Nucleosome remodeling activity of SWI/SNF and SAGA are mandatory for an efficient induction of upregulation of *ACR2* (Menezes et al., 2017). Another Yap8-arsenic relation takes place indirectly through Ufd2. Ufd2, an E4-ubiquitin ligase, is positively regulated in response to arsenicals both at translational and post-translational levels. In the presence of arsenic, Ufd2 interacts with Yap8 to invoke its stabilization. Consequently, expression of *ACR3* and potency of yeast to acclimatize to arsenic-triggered hazards are modulated. Despite Ufd2 U-box domain, plays roles in ubiquitin ligation, it is not indispensable for Yap8 stability and has no importance in terms of arsenic resistance (Ferreira et al., 2015).

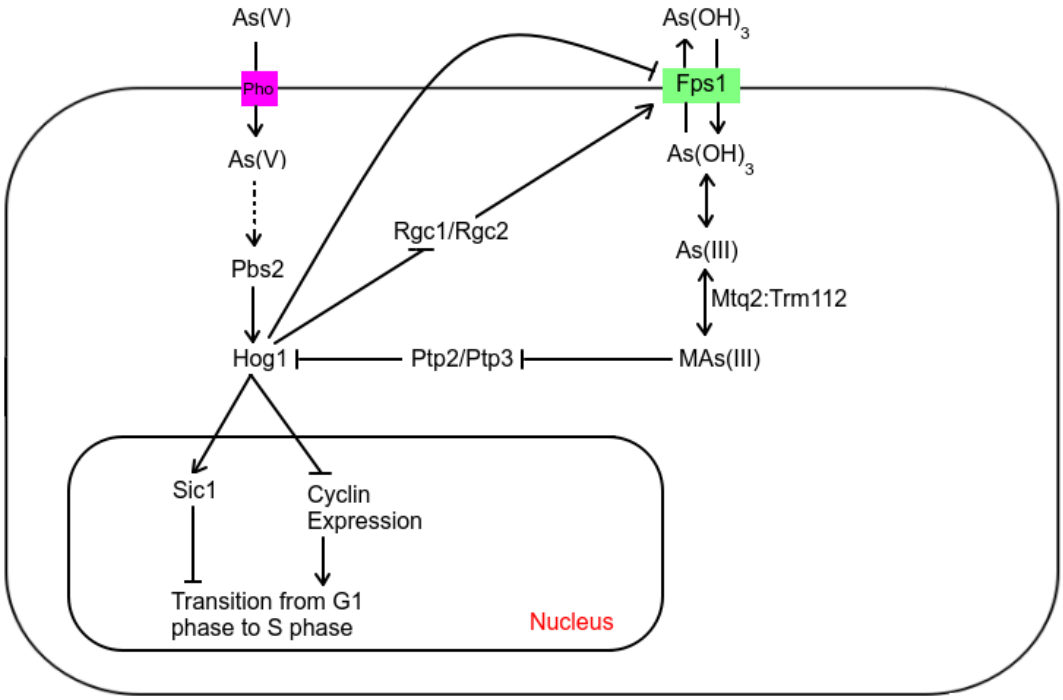

Fig. 2 Hog1 activation by arsenic exposure ends up with the delayed G_1 checkpoint (Migdal et al., 2008) and decreased uptake of As (III) (Wysocki & Tamás, 2011).

Fig. 2 shows Hog pathway, **H**igh **O**smolarity **G**lycerol pathway, is a stress activated signaling route for budding yeast. Its major component is Hog1. Hog1 can be activated by means of arsenic. In order to activate Hog1, As (III) must enter into cell by the action of Fps1. Entered As (III) must be processed via means of Mtq2:Trm112 which functions as a methyl ligation machine. Methylated As (III) prevents activities of Ptp2 and Ptp3. Because Ptp2 and Ptp3 are not functioning, they lack ability to inhibit Hog1 by dephosphorylation. Therefore Hog1 remains free to enter into nucleus and cause changes in nuclear actions. Story for Hog1 activation by the help of As (V) is different. It acts on Hog1 by providing activation of Pbs2. Activated Pbs2 causes phosphorylation of Hog1 thus makes Hog1 functional (Jongmin Lee & Levin, 2018). Functioning Hog1 can modulate Fps1 closing. It achieves this by phosphorylation of positive regulators of Fps1, namely Rgc1 and/or Rgc2. Phosphorylation of these two regulators results in dissociation of them from Fsp1 so the closure of Fsp1 (Jongmin Lee & Levin, 2015). Alternatively Hog1 can directly phosphorylate and negatively regulate Fsp1. As Fps1 closed, As (III) uptake goes downward (Wysocki & Tamás, 2011). If Hog1 pass into nucleus, it gains ability to interfere with cyclin expression (González-Novo et al., 2015) while it is able to stabilize Sic1 by phosphorylation, as well (Escoté et al., 2004). Stabilized Sic1 could escape from ubiquitin dependent degradation and interfere with B-type cyclin-Cdk which is significant for phase transition from G_1 to S phase (Yang et al., 2013). Functional Sic1 together with downregulation of cyclin expression lead to

delays in G_1 phase check point (Escoté et al., 2004; Migdal et al., 2008). However this is not the single way that As (III) interferes with cell cycle (Wysocki & Tamás, 2011).

Besides Hog1 pathway, TOR and PKA pathways are reported to be affected from As (III) exposure, too. These two pathways are known to mediate signaling and nutritional events (Wysocki & Tamás, 2011). Their actions in response to As (III) includes upregulation of stress tolerance genes and downregulation of ribosomal protein genes. The mechanism behind reduction in ribosomal biogenesis involves inhibition of TORC1. Inhibited TORC1 could not activate Sfp1, the transcription factor managing ribosome biogenesis. By this way reduction in ribosome biogenesis happens (Hosiner et al., 2009). Another report stated that Slt2 MAPK pathway, essential for cell integrity, is activated with arsenate exposure. One of changes on this pathway because of the exposure is the phosphorylation of Slt2 (Matia-González & Rodríguez-Gabriel, 2011). Whilst Hog1 prevents arsenite entry through phosphorylation at T231 of Fps1, Slt2 invokes arsenite efflux via phosphorylation at S537 (Ahmadpour et al., 2016). Additionally, rise at the level of cytosolic calcium divalent ion (Ca^{2+} or Ca^{++}) is observed in response to arsenite triggered stress. Consequently, Crz1 exposed to dephosphorylation and become free to enter into nucleus. Dephosphorylated Crz1 stimulates genes encoding the Ca^{2+} transporters Pmr1 and Pmc1, and expressing a protein play role in synthesis of the cell wall, Gsc2 (Ferreira et al., 2012). While Pmr1 is pumping Ca^{++} towards inside of Golgi complex (Sorin et al., 1997), Pmc1 stands for pumping Ca^{++} into Endoplasmic reticulum (Cunningham & Fink, 1994).

A study in 2013 elicited the repressive impact of arsenate on high affinity Fe uptake by downregulating iron transporter Fet3-Ftr1 complex (Batista-Nascimento et al., 2013). Downregulation involves inactivation of Aft1, Fe sensing transcription factor (Outten & Albetel, 2013). While both of these proteins are downregulated at the level of mRNA, Fet3 additionally modulated via internalization into ER and degradation (Batista-Nascimento et al., 2013).

Another aspects of As (III) exposure is a significant rise in GSH synthesis. Thus cytosolic GSH level increases, too. This rise in GSH level achieved via upregulation of components of sulfur assimilation pathway and directing the assimilation products of the assimilation pathway into production of GSH (Thorsen et al., 2007). Formed GSH is capable of chelating metals for later imprisoning of them into the vacuole and/or providing protection to cell from metal-triggered oxidation and/or inhibiting creation of irreversibly bound metal-protein pairs and/or preventing of oxidative damages on proteins (Wysocki & Tamás, 2011). If As (III) achieves to disrupt folding and functioning of the protein (Aposhian & Aposhian, 2006; Kitchin & Wallace, 2008), the yeast protects itself by the help of ubiquitin-proteasome mechanism which destroys these damaged structures (Goldberg, 2003).

As (III) is found to lead protein aggregation. However the aggregation is not sourced from mistranslation because it is known that As (III) does not induce efficient mistranslation. Some chaperones discovered to be linked with As (III)-stimulated aggregation which implies that arsenic

provokes protein misfolding. Arsenite provokes aggregation of peptide chains which are not folded yet and directly prevents functioning of chaperone. Protein aggregates, formed because of As (III) exposure, are capable of disrupting labile proteins to make them aggregated and misfolded. Accumulation of these aggregates further contributes to arsenic cytotoxicity despite they are cleared by proteosomal system whose enhancement is important for As (III) resistance (Jacobson et al., 2012). Budding yeast faced with As (III) displays increased expression of chaperones and transcriptional upregulation of proteasomal elements via Rpn4. Rpn4 deficient yeasts suffer from As (III)-sensitivity (Haugen et al., 2004; Thorsen et al., 2007, 2009) while this protein is also needed for Cd (Thorsen et al., 2009) and Cr tolerance (Holland et al., 2007). In terms of protein aggregation, As (III) seem to be more aggressive than Cd and Cr (Jacobson et al., 2012).

Conclusion

Arsenic is a soft metal (loid) and it attracts interest of researcher's because of its toxic properties and impacts on human and other livings. This metalloid is well-studied on budding yeast. In this review we presented accumulated scientific data regarding impacts of arsenic on budding yeast, at molecular level. Species of arsenic differs in the strategy that creates insults on cells. Speciation of arsenic may be the most significant determinant of its cellular effects. Negative impacts of arsenic involve changes in cellular signaling, protein folding and expression, calcium homeostasis and overall antioxidative capacity. *Saccharomyces cerevisiae* has various cellular systems, some of which are extracellular, to overcome arsenic and arsenic-mediated damages. To encounter with intracellular defense arsenic should into cell which is provided by several transporters. Once entered into cell, according to its speciation, as can experience some reactions and/or flux in the direction of out of the cell. Arsenic is known to act on expression of arsenic tolerance genes.

Recently developed YARG is a database containing 3396 arsenic-associated genes of *S. cerevisiae* that are gained from 13 phenotypic screening and 7 transcriptional profiling datasets (Rathod et al., 2018). The database is available at http://cosbi4.ee.ncku.edu.tw/YARG/search. This yeast database could be useful for future arsenic specific investigations.

REFERENCES

Aaltonen, E. K. J., & Silow, M. (2008). Transmembrane topology of the Acr3 family arsenite transporter from *Bacillus subtilis*. *Biochimica et Biophysica Acta (BBA)-Biomembranes*, *1778*(4), 963–973.

Ahmadpour, D., Maciaszczyk-Dziubinska, E., Babazadeh, R., Dahal, S., Migocka, M., Andersson, M., … Hohmann, S. (2016). The mitogen-activated protein kinase Slt2 modulates arsenite transport through the aquaglyceroporin Fps1. *FEBS Letters*, *590*(20), 3649–3659.

Aposhian, H. V., & Aposhian, M. M. (2006). Arsenic toxicology: five questions. *Chemical Research in Toxicology*, *19*(1), 1–15.

Bánfalvi, G. (2011). Heavy metals, trace elements and their cellular effects. In *Cellular effects of heavy metals* (pp. 3–28). Springer.

Bates, M. N., Smith, A. H., & Hopenhayn-Rich, C. (1992). Arsenic ingestion and internal cancers: a review. *American Journal of Epidemiology*, *135*(5), 462–476.

Batista-Nascimento, L., Toledano, M. B., Thiele, D. J., & Rodrigues-Pousada, C. (2013). Yeast protective response to arsenate involves the repression of the high affinity iron uptake system. *Biochimica et Biophysica Acta (BBA)-Molecular Cell Research*, *1833*(5), 997–1005.

Bhattacharya, P., Frisbie, S. H., Smith, E., Naidu, R., Jacks, G., & Sarkar, B. (2002). Arsenic in the environment: a global perspective. *Handbook of Heavy Metals in the Environment. Marcell Dekker Inc., New York*, 147–215.

Bun-ya, M., Shikata, K., Nakade, S., Yompakdee, C., Harashima, S., & Oshima, Y. (1996). Two new genes, *PHO86* and *PHO87*, involved in inorganic phosphate uptake in Saccharomyces cerevisiae. *Current Genetics*, *29*(4), 344–351.

Chen, C. J., Chen, C. W., Wu, M. M., & Kuo, T. L. (1992). Cancer potential in liver, lung, bladder and kidney due to ingested inorganic arsenic in drinking water. *British Journal of Cancer*, *66*(5), 888–892.

Chu, H.-A., & Crawford-Brown, D. (2006). Inorganic Arsenic in Drinking Water and Bladder Cancer: A Meta-Analysis for Dose-Response Assessment. *International Journal of Environmental Research and Public Health*, *3*(4), 316–322. https://doi.org/10.3390/ijerph2006030039.

Cortés, P., Castrejón, V., Sampedro, J. G., & Uribe, S. (2000). Interactions of arsenate, sulfate and phosphate with yeast mitochondria. *Biochimica et Biophysica Acta (BBA)-Bioenergetics*, *1456*(2–3), 67–76.

Cunningham, K. W., & Fink, G. R. (1994). Calcineurin-dependent growth control in *Saccharomyces cerevisiae* mutants lacking PMC1, a homolog of plasma membrane Ca2+ ATPases. *The Journal of Cell Biology*, *124*(3), 351–363.

Da Silva, J. J. R. F., & Williams, R. J. P. (2001). *The biological chemistry of the elements: the inorganic chemistry of life*. Oxford University Press.

Di, Y., & Tamás, M. J. (2007). Regulation of the arsenic-responsive transcription factor Yap8p involves the ubiquitin-proteasome pathway. *Journal of Cell Science*, *120*(2), 256–264.

Escoté, X., Zapater, M., Clotet, J., & Posas, F. (2004). Hog1 mediates cell-cycle arrest in G1 phase by the dual targeting of Sic1. *Nature Cell Biology*, *6*(10), 997.

Ferreira, R. T., Menezes, R. A., & Rodrigues-Pousada, C. (2015). E4-Ubiquitin ligase Ufd2 stabilizes Yap8 and modulates arsenic stress responses independent of the U-box motif. *Biology Open*, bio-010405.

Ferreira, R. T., Silva, A. R. C., Pimentel, C., Batista-Nascimento, L., Rodrigues-Pousada, C., & Menezes, R. A. (2012). Arsenic stress elicits cytosolic Ca2+ bursts and Crz1 activation in Saccharomyces cerevisiae. *Microbiology*, *158*(9), 2293–2302.

Fu, H.-L., Meng, Y., Ordóñez, E., Villadangos, A. F., Bhattacharjee, H., Gill, J. A., … Rosen, B. P. (2009). Properties of arsenite efflux permeases (Acr3) from Alkaliphilus metalliredigens and *Corynebacterium glutamicum*. *Journal of Biological Chemistry*, jbc-M109.

Ghosh, M., Shen, J., & Rosen, B. P. (1999). Pathways of As (III) detoxification in *Saccharomyces cerevisiae*. *Proceedings of the National Academy of Sciences*, *96*(9), 5001–5006.

Goldberg, A. L. (2003). Protein degradation and protection against misfolded or damaged proteins. *Nature*, *426*(6968), 895.

González-Novo, A., Jiménez, J., Clotet, J., Nadal-Ribelles, M., Cavero, S., de Nadal, E., & Posas, F. (2015). Hog1 targets Whi5 and Msa1 transcription factors to down-regulate cyclin expression upon stress. *Molecular and Cellular Biology*, MCB-01279.

Halem, D., Bakker, S. A., Amy, G., & Dijk, J. (2009). *Arsenic in drinking water: A worldwide water quality concern for water supply companies*. *Drinking Water Engineering and Science* (Vol. 2). https://doi.org/10.5194/dwes-2-29-2009

Haugen, A. C., Kelley, R., Collins, J. B., Tucker, C. J., Deng, C., Afshari, C. A., … Van Houten, B. (2004). Integrating phenotypic and expression profiles to map arsenic-response networks. *Genome Biology*, *5*(12), R95.

Holland, S., Lodwig, E., Sideri, T., Reader, T., Clarke, I., Gkargkas, K., … Avery, S. V. (2007). Application of the comprehensive set of heterozygous yeast deletion mutants to elucidate the molecular basis of cellular chromium toxicity. *Genome Biology*, *8*(12), R268.

Hosiner, D., Lempiäinen, H., Reiter, W., Urban, J., Loewith, R., Ammerer, G., … Schüller, C. (2009). Arsenic toxicity to Saccharomyces cerevisiae is a consequence of inhibition of the TORC1 kinase combined with a chronic stress response. *Molecular Biology of the Cell*, *20*(3), 1048–1057.

Jacobson, T., Navarrete, C., Sharma, S. K., Sideri, T. C., Ibstedt, S., Priya, S., … Tamás, M. J. (2012). Arsenite interferes with protein folding and triggers formation of protein aggregates in yeast. *J Cell Sci*, jcs-107029.

Kim, K.-W., Chanpiwat, P., Hanh, H. T., Phan, K., & Sthiannopkao, S. (2011). Arsenic geochemistry of groundwater in Southeast Asia. *Frontiers of Medicine*, *5*(4), 420–433. https://doi.org/10.1007/s11684-011-0158-2

Kiriyama, K., Hara, K. Y., & Kondo, A. (2012). Extracellular glutathione fermentation using engineered Saccharomyces cerevisiae expressing a novel glutathione exporter. *Applied Microbiology and Biotechnology*, *96*(4), 1021–1027.

Kitchin, K. T., & Wallace, K. (2008). The role of protein binding of trivalent arsenicals in arsenic carcinogenesis and toxicity. *Journal of Inorganic Biochemistry*, *102*(3), 532–539.

Kuge, S., & Jones, N. (1994). YAP1 dependent activation of TRX2 is essential for the response of Saccharomyces cerevisiae to oxidative stress by hydroperoxides. *The EMBO Journal*, *13*(3), 655–664.

Kumar, N. V., Yang, J., Pillai, J. K., Rawat, S., Solano, C., Kumar, A., … Tamás, M. J. (2016). Arsenic directly binds to and activates the yeast AP-1-like transcription factor Yap8. *Molecular and Cellular Biology*, *36*(6), 913–922.

Lee, J., Godon, C., Lagniel, G., Spector, D., Garin, J., Labarre, J., & Toledano, M. B. (1999). Yap1 and Skn7 control two specialized oxidative stress response regulons in yeast. *Journal of Biological Chemistry*, *274*(23), 16040–16046.

Lee, J., & Levin, D. E. (2015). Rgc2 Regulator of Glycerol Channel Fps1 Functions as Homo-and Hetero-dimers with Rgc1. *Eukaryotic Cell*, EC-00073.

Lee, J., & Levin, D. E. (2018). Intracellular mechanism by which arsenite activates the yeast stress MAPK Hog1. *Molecular Biology of the Cell*, *29*(15), 1904–1915.

Lemire, J. A., Harrison, J. J., & Turner, R. J. (2013). Antimicrobial activity of metals: mechanisms, molecular targets and applications. *Nature Reviews. Microbiology*, *11*(6), 371–384. https://doi.org/10.1038/nrmicro3028

Liu, Z., Boles, E., & Rosen, B. P. (2004). Arsenic trioxide uptake by hexose permeases in *Saccharomyces cerevisiae*. *Journal of Biological Chemistry*, *279*(17), 17312–17318.

Liu, Z., Sanchez, M. A., Jiang, X., Boles, E., Landfear, S. M., & Rosen, B. P. (2006). Mammalian glucose permease GLUT1 facilitates transport of arsenic trioxide and methylarsonous acid. *Biochemical and Biophysical Research Communications*, *351*(2), 424–430.

Maciaszczyk-Dziubinska, E., Wawrzycka, D., & Wysocki, R. (2012). Arsenic and antimony transporters in eukaryotes. *International Journal of Molecular Sciences*, *13*(3), 3527–3548.

Matia-González, A. M., & Rodríguez-Gabriel, M. A. (2011). Slt2 MAPK pathway is essential for cell integrity in the presence of arsenate. *Yeast*, *28*(1), 9–17.

Menezes, R. A., Pimentel, C., Silva, A. R. C., Amaral, C., Merhej, J., Devaux, F., & Rodrigues-Pousada, C. (2017). Mediator, SWI/SNF and SAGA complexes regulate Yap8-dependent transcriptional activation of ACR2 in response to arsenate. *Biochimica et Biophysica Acta (BBA)-Gene Regulatory Mechanisms*, *1860*(4), 472–481.

Migdal, I., Ilina, Y., Tamás, M. J., & Wysocki, R. (2008). Mitogen-activated protein kinase Hog1 mediates adaptation to G1 checkpoint arrest during arsenite and hyperosmotic stress. *Eukaryotic Cell*, *7*(8), 1309–1317.

Morgan, B. A., Banks, G. R., Toone, W. M., Raitt, D., Kuge, S., & Johnston, L. H. (1997). The Skn7 response regulator controls gene expression in the oxidative stress response of the budding yeast Saccharomyces cerevisiae. *The EMBO Journal*, *16*(5), 1035–1044.

Mukhopadhyay, R., & Rosen, B. P. (1998). *Saccharomyces cerevisiae* ACR2 gene encodes an arsenate reductase. *FEMS Microbiology Letters*, *168*(1), 127–136.

Mukhopadhyay, R., Shi, J., & Rosen, B. P. (2000). Purification and Characterization of Acr2p, the *Saccharomyces cerevisiae* Arsenate Reductase. *Journal of Biological Chemistry*, *275*(28), 21149–21157.

Nathaniel, M. M. (2005). Arsenic: In Search of an Antidote to a Global Poison. *Environmental Health Perspectives*, *113*(6), A378–A386. https://doi.org/10.1289/ehp.113-a378

Outten, C. E., & Albetel, A.-N. (2013). Iron sensing and regulation in *Saccharomyces cerevisiae*: ironing out the mechanistic details. *Current Opinion in Microbiology*, *16*(6), 662–668.

Porquet, A., & Filella, M. (2007). Structural evidence of the similarity of Sb (OH) 3 and As (OH) 3 with glycerol: implications for their uptake. *Chemical Research in Toxicology*, *20*(9), 1269–1276.

Ralph, S. J. (2008). Arsenic-based antineoplastic drugs and their mechanisms of action. *Metal-Based Drugs*, *2008*.

Ramírez-Solís, A., Mukopadhyay, R., Rosen, B. P., & Stemmler, T. L. (2004). Experimental and Theoretical Characterization of Arsenite in Water: Insights into the Coordination Environment of As− O. *Inorganic Chemistry*, *43*(9), 2954–2959.

Rathod, J., Tu, H.-P., Chang, Y.-I., Chu, Y.-H., Tseng, Y.-Y., Jean, J.-S., & Wu, W.-S. (2018). YARG: A repository for arsenic-related genes in yeast. *PloS One*, *13*(7), e0201204.

Smith, A. H., Hopenhayn-Rich, C., Bates, M. N., Goeden, H. M., Hertz-Picciotto, I., Duggan, H. M., … Smith, M. T. (1992). Cancer risks from arsenic in drinking water. *Environmental Health Perspectives*, *97*, 259–267. https://doi.org/10.1289/ehp.9297259

Sorin, A., Rosas, G., & Rao, R. (1997). PMR1, a Ca2+-ATPase in yeast Golgi, has properties distinct from sarco/endoplasmic reticulum and plasma membrane calcium pumps. *Journal of Biological Chemistry*, *272*(15), 9895–9901.

Tamás, M. J., Fauvet, B., Christen, P., & Goloubinoff, P. (2018). Misfolding and aggregation of nascent proteins: a novel mode of toxic cadmium action in vivo. *Current Genetics*, *64*(1), 177–181.

Tamás, M. J., Sharma, S. K., Ibstedt, S., Jacobson, T., & Christen, P. (2014). Heavy metals and metalloids as a cause for protein misfolding and aggregation. *Biomolecules*, *4*(1), 252–267.

Thorsen, M., Lagniel, G., Kristiansson, E., Junot, C., Nerman, O., Labarre, J., & Tamás, M. J. (2007). Quantitative transcriptome, proteome, and sulfur metabolite profiling of the *Saccharomyces cerevisiae* response to arsenite. *Physiological Genomics*, *30*(1), 35–43.

Thorsen, M., Perrone, G. G., Kristiansson, E., Traini, M., Ye, T., Dawes, I. W., … Tamás, M. J. (2009). Genetic basis of arsenite and cadmium tolerance in *Saccharomyces cerevisiae*. *BMC Genomics*, *10*(1), 105.

Vahter, M. (2008). Health effects of early life exposure to arsenic. *Basic & Clinical Pharmacology & Toxicology*, *102*(2), 204–211. https://doi.org/10.1111/j.1742-7843.2007.00168.x

Vujcic, M., Shroff, M., & Singh, K. K. (2007). Genetic determinants of mitochondrial response to arsenic in yeast *Saccharomyces cerevisiae*. *Cancer Research*, *67*(20), 9740–9749.

Wysocki, R., Bobrowicz, P., & Ułaszewski, S. (1997). The Saccharomyces cerevisiae ACR3 gene encodes a putative membrane protein involved in arsenite transport. *Journal of Biological Chemistry*, *272*(48), 30061–30066.

Wysocki, R., & Tamás, M. J. (2011). *Saccharomyces cerevisiae* as a model organism for elucidating arsenic tolerance mechanisms. In *Cellular Effects of Heavy Metals* (pp. 87–112). Springer.

Yang, X., Lau, K.-Y., Sevim, V., & Tang, C. (2013). Design principles of the yeast G1/S switch. *PLoS Biology*, *11*(10), e1001673.

Yompakdee, C., Bun-ya, M., Shikata, K., Ogawa, N., Harashima, S., & Oshima, Y. (1996). A putative new membrane protein, Pho86p, in the inorganic phosphate uptake system of *Saccharomyces cerevisiae*. *Gene*, *171*(1), 41–47.

Yu, M.-H., & Tsunoda, H. (2004). *Environmental toxicology: biological and health effects of pollutants*. crc press.