development result in short forelimbs[7].

The diverse mechanisms underlying flightlessness that have been identified in these genomic studies are not necessarily incompatible with each other. Indeed, an emerging perspective is that the genetic mechanisms that lead to changes in wing shape and length might be as diverse as the ecological contexts in which flight loss has occurred. Perhaps this is not surprising. Studies of digit reduction in mammals have shown similarly diverse mechanisms[8,9], and different genetic mechanisms underlie adaptations to high altitude in closely related hummingbird species[10]. More work with museum collections[11], and in developmental biology and anatomy, is needed to advance our understanding of the genetic changes that underpin traits such as flightlessness. ■

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METABOLISM

A stress-coping strategy for yeast cells

Stressed yeast cells take up the amino acid lysine and reprogram their metabolism to free up supplies of a stress-relieving molecule. Lysine uptake therefore increases the tolerance of yeast cells to stress. SEE LETTER P.249

JENS NIELSEN

Metabolism is crucial for all living cells: it provides energy as well as the molecular building blocks required for growth. Some metabolic pathways protect cells against different types of stress, including the oxidative stress caused by other metabolic processes in the cell or by external factors. On page 249, Olin-Sandoval *et al.*[1] describe how yeast cells (*Saccharomyces cerevisiae*) can reprogram their metabolism so that they are better equipped to handle the oxidative stress that is caused by the accumulation of chemically reactive molecules known as reactive oxygen species (ROS).

Understanding how the many different metabolic pathways in a cell interact and ensure its proper functioning under varying environmental conditions is necessary for designing cell 'factories' — genetically engineered cells that can be cultured to produce fuels, chemicals, foods or pharmaceuticals. It is also important for gaining insight into the molecular mechanisms that underlie various human diseases, because metabolic changes are associated not only with disorders such as diabetes and cardiovascular disease that have conventionally been considered to be metabolic disorders, but also with conditions such as cancer and Alzheimer's disease[2].

Cell factories and human cells undergoing pronounced metabolic changes (such as cancer cells) experience different types of stress, including oxidative stress, which can be caused by the accumulation of ROS. These molecules disrupt many cellular processes: for example, they cause DNA damage and problems with protein folding. Cells have therefore evolved various defence mechanisms to cope with ROS accumulation.

The dominant pathway that cells use to combat accumulated ROS is to chemically reduce them with the thiol group (SH) of the antioxidant peptide glutathione; this reaction results in the formation of a sulfur bridge between two glutathione molecules (Fig. 1). To replenish the levels of glutathione, certain enzymes break the sulfur bridge apart, using the cofactor molecule NADPH as an electron acceptor to promote the reaction. Thus, when cells experience oxidative stress and must deplete accumulated ROS, they have a higher demand for NADPH than do non-stressed cells. However, NADPH is sometimes required for rapid cell growth; therefore, in growing cells, there might be less NADPH available for handling accumulated ROS than in non-growing cells. Olin-Sandoval *et al.*[1] demonstrate that, in the presence of the amino acid lysine, yeast cells can reprogram their metabolism such that they can allocate more NADPH for dealing with accumulated ROS.

The authors found this mechanism while studying a previously reported, yet largely unexplained, phenomenon: that yeast cells lacking Tpo1, an exporter protein that removes a group of chemicals called polyamines from the cell, are more sensitive to oxidative stress than wild-type cells[3]. Olin-Sandoval *et al.* used protein-expression analyses to demonstrate that, compared with wild-type yeast cells, yeast

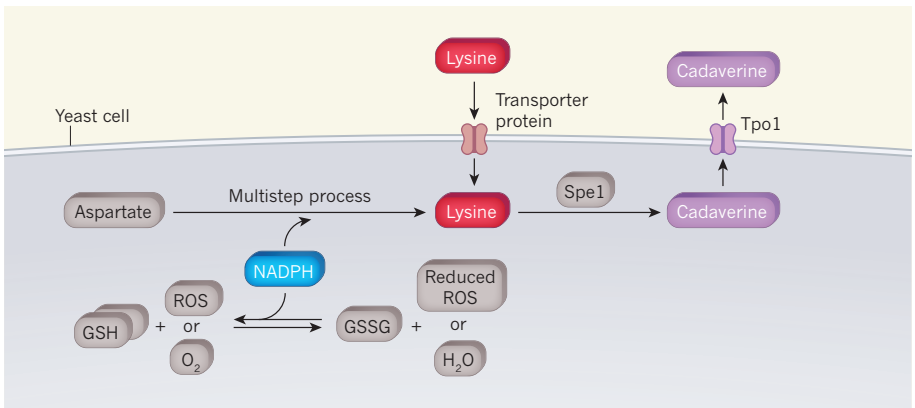

Figure 1 | Yeast cells reprogram their metabolism to reduce stress. In yeast cells, reactive oxygen species (ROS) and molecular oxygen (O_2) are chemically reduced by reaction with pairs of glutathione (GSH) peptides, which become linked by a sulfur bridge to form glutathione disulfide (GSSG). The enzyme-cofactor molecule NADPH is needed to replenish the levels of glutathione in the cell, and also for the multistep production of the amino acid lysine from another amino acid, aspartate. Olin-Sandoval *et al.*[1] found that yeast cells can harvest large amounts of lysine from outside the cell. The enzyme Spe1 converts lysine to the polyamine cadaverine, which is removed from the cell by the exporter protein Tpo1. Lysine harvesting results in an inhibition of lysine production (not shown), probably through a feedback mechanism. Thus, lysine harvesting reduces the use of NADPH for lysine synthesis, freeing it up for its role in handling accumulated ROS.

cells lacking Tpo1 show increased expression of enzymes that are involved in producing lysine, one of the amino acids used as building blocks to make proteins. This finding led the authors to speculate that polyamine export might be involved in protecting cells against oxidative stress.

Removing the carboxyl group (COOH) from lysine — that is, decarboxylation of lysine — produces a polyamine called cadaverine. Although yeast cells have not previously been reported to produce cadaverine, they make another polyamine, putrescine, by decarboxylating the amino acid ornithine[4]. Putrescine is a precursor molecule that is needed to make the polyamines spermidine and spermine, which activate a quality-control process called autophagy in which cell components are degraded and recycled[5]; therefore, putrescine is also crucial for protecting cells against stress.

Lysine and ornithine have very similar molecular structures, and the authors found that Spe1, the enzyme that decarboxylates ornithine, can also decarboxylate lysine to generate cadaverine. Although experiments with purified Spe1 revealed that the enzyme has a much lower affinity for lysine than it does for ornithine, Olin-Sandoval *et al.* showed that yeast cells can take up lysine from the surrounding culture medium to increase its concentration inside the cell, such that Spe1 will then convert it to cadaverine. Thus, harvested lysine is decarboxylated to cadaverine, which, in turn, is exported from the cell by Tpo1 (Fig. 1). Although the production of cadaverine by Spe1 was accompanied by increased production of putrescine, the levels of spermidine and spermine were not affected, suggesting that lysine uptake and cadaverine production do not affect autophagy.

The increase in lysine levels in the yeast cells that harvest the amino acid from outside the cell might elicit feedback mechanisms to inhibit lysine production. In yeast cells, the generation of lysine from the amino acid aspartate requires NADPH (Fig. 1). Therefore, the harvesting of lysine from outside the cell results in NADPH being spared from use in the production of lysine. Using mathematical modelling of cellular metabolism, the authors found that decreases in the production of lysine would result in an attenuated flux of molecules through the oxidative part of the pentose phosphate pathway — the metabolic pathway that is the dominant source of NADPH. Thus, harvesting high levels of lysine enables stressed cells to produce more NADPH through the pentose phosphate pathway to deal with accumulated ROS. Consistent with this, the authors found that lysine-harvesting cells were less sensitive to the oxidative-stress agent diamide than were cells that did not harvest lysine.

Supplementation of methionine, another amino acid whose synthesis requires large amounts of NADPH, also improves the tolerance of cells to diamide[6]. But Olin-Sandoval and colleagues found that lysine harvesting is even better than methionine supplementation at improving diamide tolerance. Furthermore, cells that harvested lysine showed less ROS accumulation than did cells that did not harvest lysine. Whether cadaverine (and putrescine) improves the tolerance of cells to stress, in addition to the other effects of lysine harvesting on metabolism, remains unclear.

Many metabolic processes are conserved in evolution between yeast and mammalian cells, and yeast is a widely used model organism for studying human cells[7]. The authors therefore also evaluated whether lysine harvesting is a general mechanism for improving stress tolerance, and indeed found this to be the case in several other species of yeast, as well as in *Bacillus subtilis*, a species of the Gram-positive group of bacteria. However, although the authors demonstrated that several different human cell lines can harvest lysine, lysine harvesting did not make these cells more tolerant to oxidative stress. This is probably because human cells cannot synthesize lysine themselves, and therefore do not need a supply of NADPH to support biosynthesis of lysine; in these cells, harvesting lysine would not free up the capacity to create NADPH.

"Lysine-harvesting cells were less sensitive to oxidative stress than cells that did not harvest lysine."

Despite the lack of protective effects of lysine harvesting in human cells, the findings are still exciting because they provide an excellent demonstration of how apparently unrelated metabolic pathways interact. Moreover, they illustrate how a seemingly simple process — that is, the harvesting and decarboxylation of lysine — can drive major metabolic reprogramming that leads to an increase in the cell's capacity for producing NADPH. Besides these general findings, the study is also relevant for efforts to use yeast as a cell-factory system, because oxidative stress presents major problems for many production processes in such cell factories[7]. The findings of this study indicate that supplementing yeast-cell medium with lysine could help the cells to overcome this stress, and thus improve the efficiency of production of various valuable chemicals. ■

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HYDROLOGY

Groundwater resilience in sub-Saharan Africa

An analysis of aquifer replenishment in sub-Saharan Africa shows that reduced precipitation does not always deplete groundwater reserves, challenging the idea that these reserves will decrease in response to global warming. SEE LETTER P.230

RICHARD W. HEALY

The population of sub-Saharan Africa is currently about 1 billion, and is predicted to double by 2050 (go.nature.com/2zj9kca), whereas the region's climate is predicted to become drier during the same period[1]. Clearly, the demand for fresh water will increase. Whether groundwater can satisfy this demand is a looming question. Little is known about the rates at which water is replenished to groundwater aquifers[2,3] in that region, and thus the rate of sustainable withdrawal. On page 230, Cuthbert *et al.*[4] identify the processes involved, and examine the long-term trends of aquifer replenishment in sub-Saharan Africa. The authors conclude that future drying climatic trends could affect surface-water supplies, but might not decrease groundwater supplies.

Aquifer replenishment occurs naturally in two general forms: precipitation that infiltrates at the land surface and percolates to the water table (most common in humid regions), and infiltration from streams and other surface-water bodies (most common in arid regions).