among the immense number of components and then showing that this set is necessary and sufficient for producing the main features of the multicellular system. Engineering multicellular behaviors with such a set of minimal elements is a promising way to test whether we truly understand principles of multicellular systems (You et al., 2004; Liu et al., 2011).

ACKNOWLEDGMENTS

We thank K. Roybal for insightful suggestions. W.A.L. is supported by NIH grants R01 GM55040, R01 GM62583, and P50 GM081879; the NSF Synthetic Biology Engineering Research Center (SynBERC); and the Howard Hughes Medical Institute (HHMI). H.Y. is an HHMI fellow of the Damon Runyon Cancer Research Foundation (DRG-2089-11).

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Monoculture Breeds Poor Social Skills

Gemma L. Staniforth[1,*] and Mick F. Tuite[1,*]

[1]Kent Fungal Group, School of Biosciences, University of Kent, Canterbury, Kent CT2 7NJ, UK

*Correspondence: g.l.staniforth@kent.ac.uk (G.L.S.), m.f.tuite@kent.ac.uk (M.F.T.)

http://dx.doi.org/10.1016/j.cell.2014.08.004

Two studies from Jarosz et al. describe how [*GAR*$^+$], a protein-based epigenetic determinant found mainly in wild yeast strains, can be activated by microbial cross-kingdom communication. With the aid of genetically and ecologically diverse bacteria, yeast can override an ancient regulatory mechanism of glucose repression, promoting both microbial diversity and lifespan extension.

Why some wine fermentations fail has long been a mystery. The resulting bacterial spoilage of the highly valued byproduct of yeast metabolism is typically assumed to be a consequence of the failure of the yeast to produce enough ethanol to give it a growth advantage over the bacteria also found in grape must. Now, two papers in this issue from the Lindquist and Bisson laboratories (Jarosz et al., 2014a, 2014b) indicate that, in fact, the "spoiling" bacteria trigger, rather than passively capitalize on, the arrest in yeast fermentation by secreting a yet-to-be identified chemical messenger that induces the appearance in the wine yeast of a protein-based epigenetic determinant called [*GAR*$^+$] (Figure 1). This chemical dialog with the bacteria is not limited to wine yeasts either but is observed with multiple fruit yeasts and brewing yeasts. The juxtaposition of bacterial contamination on solid nutrients growing yeast is sufficient to induce phenotypic change in the yeast and was the first indication, that was later verified, that a secreted chemical messenger was involved. In every case, the appearance of [*GAR*$^+$] has significant ramifications for the host metabolism, one key change being a >50% reduction in ethanol output, thereby creating a more bacteria-friendly environment.

[*GAR*$^+$] is an unusual epigenetic element. Decades after its original identification based on its non-Mendelian inheritance pattern (Kunz and Ball, 1977), [*GAR*$^+$] was shown to share some, but not all, of the properties expected of a yeast prion (Brown and Lindquist, 2009). Where [*GAR*$^+$] differs from other yeast prions is in its nondependency on the molecular chaperone Hsp104 for its propagation (although it does require a different chaperone, Hsp70). In addition, there are two proteins associated with the [*GAR*$^+$] determinant, neither of which forms amyloids. In the [*gar*$^-$] state, these two proteins have distinct functions and cellular locations—Pma1p is a membrane-bound proton pump, whereas Std1p is a nuclear transcription factor.

The switch to [*GAR*$^+$] turns the yeast from a metabolic specialist devoted to one carbon source—glucose—to a metabolic generalist that can use a wide range of sugars and is no longer subjected to glucose repression. It is presently unclear how this metabolic rewiring is mediated by [*GAR*$^+$]. Attenuation of Pma1p activity may be a contributing factor, as disruption

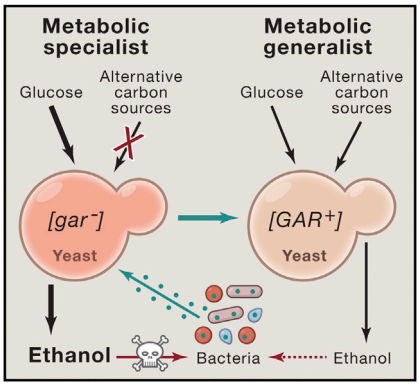

Figure 1. Cross-Kingdom Communication Rewires Yeast Metabolism

In [*gar*$^-$] cells, glucose is preferentially taken up and fermented to ethanol, whereas other carbon sources are ignored. The resulting high levels of ethanol are inhibitory to bacterial growth. However, some bacterial species can secrete a low molecular weight chemical messenger (green dots) that induces the [*GAR*$^+$] state, resulting in a rewiring of glucose metabolism in yeast. As a consequence, the cells switch from metabolic specialists to generalists and are now able to use alternative carbon sources in the presence of glucose, resulting in a drop in ethanol levels, thereby favoring bacterial growth.

of pH homeostasis could impact yeast fermentation properties. In addition, earlier gene expression profiling studies identified a 40-fold downregulation of Hxt3p, a Std1p-regulated hexose transporter, as the only significant transcriptional difference between a [*gar*$^-$] and [*GAR*$^+$] state. Despite Hxt3p downregulation in [*GAR*$^+$] cells, glucose uptake is not decreased, and deletion of the *HXT3* gene does not confer the glucose-repression phenotype of [*GAR*$^+$].

How [*GAR*$^+$] overrides an ancient mechanism of glucose repression found in fungi therefore remains a mystery, as does the relative position of [*GAR*$^+$] within the signaling cascade triggered by exposure to the bacterial chemical messenger. If [*GAR*$^+$] formation were not the primary event, could it occur further downstream, serving to sustain or lock in the cellular response? In this latter sense, [*GAR*$^+$] induction would not give rise to glucose repression per se, but it would be an inseparable marker of the yeast adaptation to the bacterial stimulus.

This remarkable example of cross-kingdom communication also poses many other intriguing questions. For example, the authors attribute the apparent loss of this communication system by domesticated fungal and bacterial strains to the widespread laboratory practices of continuous monoculture. It is conceivable that this mechanism is not lost entirely, as the few known [*GAR*$^+$] modulating proteins are conserved in domesticated yeast, and perhaps the peripheral wiring has been poorly maintained in the absence of social or environmental cues for [*GAR*$^+$] formation. It would therefore be very interesting to follow long-term cocultures of domesticated yeast and nondomesticated bacterial species to determine whether the [*GAR*$^+$] system can be reestablished. Furthermore, what is the chemical nature of the secreted agent that triggers [*GAR*$^+$] formation? Jarosz et al. (2014a) show that it is heat stable and resistant to nucleases and proteases and to extremes of pH. The activity is also distinct from well-established microbial signaling molecules such as an acyl-homoserine lactone involved in quorum sensing in bacteria and farnesol related to fungal-bacteria interactions. We are left to speculate whether we are dealing with complex new chemistries that could be challenging to unravel.

An impressive 30% of the phylogenetically distinct bacterial species tested produce the [*GAR*$^+$]-inducing agent, but what distinguishes these species from the nonproducers? This cohort of bacteria could naturally cohabit the same ecological niches as yeasts, for example. Until we isolate and identify the chemical trigger, it will be difficult to pin down an underlying common metabolic signature or indeed to work out the mechanism of [*GAR*$^+$] induction. What does emerge from these new studies, however, is that the mysterious chemical messenger is unlikely to be a universal prion trigger.

The induction of [*GAR*$^+$] in yeasts by bacterially produced chemical messengers adds to the increasing catalog of productive bacteria-fungal interactions (Frey-Klett et al., 2011). These include the obvious examples of antibacterial agents, such as penicillin produced by fungi, and examples of bacterial metabolites promoting the growth of fungal hyphae. Auxofuran, for instance, produced by certain *Streptomyces* species can promote growth of the hyphae of the fly agaric mushroom *Amanita muscaria* (Riedlinger et al., 2006). There are also other cases of bacteria-fungal interactions during wine fermentations, the most significant being the requirement for nutrients, released by yeast, for the key bacteria-mediated malolactate secondary fermentation that is important for enhancing wine quality and stability (Alexandre et al., 2004). Yeast-produced ethanol may also act as a signaling molecule promoting growth and stress resistance in certain bacteria (Smith et al., 2004). On an evolutionary scale, bacteria have also modified yeast metabolism through horizontal transfer of their genes to yeasts—two strong candidates being the *URA1* gene involved in pyrimidine biosynthesis and the *BDS1* gene related to sulfur metabolism (Hall et al., 2005).

Though [*GAR*$^+$] is a relatively stable epigenetic acquisition, [*GAR*$^+$] cells can switch back to the [*gar*$^-$] state, reinstating their metabolic specialism without fixing a mutational change in the host genome. Indeed, Jarosz et al. (2014b) report the existence of genetic mimics of [*GAR*$^+$] in several wild strains isolated from soils, and as expected, the defining genetic features of prion-based inheritance are missing in this group. However, this metastable existence is a characteristic of all yeast prions and, in some cases, may provide the host organism with advantageous bet-hedging functions (Halfman and Lindquist, 2010). Prion-mediated adaptive strategies have the potential to provide yeast cells with a survival advantage in fluctuating environments and to allow the acquisition of new and beneficial phenotypes without the need to acquire and fix nuclear genetic changes. In the case of [*GAR*$^+$], the metabolic switch gives clear selective advantages to both parties: the bacteria have a significant growth advantage due to lowered ethanol levels while the yeasts obtain an expanded repertoire of carbon sources to use. In addition, the [*GAR*$^+$] yeast cells show increased longevity especially in the presence of high ethanol concentrations and under starvation conditions. The only potential losers in this bacteria-yeast dialog are the wine producers.

The discovery of a highly conserved chemical dialog between yeasts and bacteria in mixed microbial communities is a remarkable finding and will have a profound impact on the way that we

view the benefits of inter-kingdom social communication in constantly varying ecological niches. There may also be more practical benefits; generating wine yeasts that are unable to switch to the [*GAR*$^+$] state would presumably become de rigueur for the wine industry.

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Building a Temperature-Sensitive Ion Channel

Ming-Feng Tsai[1] and Christopher Miller[1,*]

[1]Department of Biochemistry, Howard Hughes Medical Institute, Brandeis University, Waltham, MA 02453, USA

*Correspondence: cmiller@brandeis.edu

http://dx.doi.org/10.1016/j.cell.2014.08.008

The biophysical basis of temperature-sensitive ion channel gating has been a tough nut to crack. Chowdhury, et al. use a protein engineering approach to render a temperature-insensitive voltage-gated channel cold- or heat-responsive to reveal principles for temperature-gating and a plausible model for molecularly enabling this mode of environmental responsiveness.

Thermodynamics is a funny subject, and arguably the one least loved by cell biology students. The generality of classical equilibrium thermodynamics and its independence of specific molecular models means that its assertions are always true—and often useless. But in this issue of *Cell*, Chowdhury and colleagues report thermodynamics-inspired experiments that productively attack a long-lingering problem in sensory neurobiology: how certain ion channel proteins achieve the exquisite thermosensitivity that allows the neurons housing them—and us—to detect changes in temperature (Chowdhury et al., 2014).

The molecular poster children for neuronal temperature sensing are found in the cation-conducting TRP channel family (Clapham, 2003). Among these are TRPV1, which detects painful heat by activating many-fold for just a few degrees rise in temperature above 35°C, and TRPM8, which steeply turns on with cooling below 25°. Many studies have sought to identify the "temperature receptor domain" in such channels, and mutants have been found that profoundly alter or even invert their temperature-activation characteristics (Brauchi et al., 2006; Grandl et al., 2008; Yang et al., 2010; Jabba et al., 2014). But since these studies have fingered widely different protein domains as the culprit, no crisp picture or molecular mechanism has yet emerged.

A few years ago, David Clapham and one of us suggested that a localized thermoreceptor domain need not exist and that a thermodynamic treatment might provide an alternative pathway into the problem (Clapham and Miller, 2011). We based our suggestion on a commonplace of protein physical chemistry: that if a large number of buried hydrophobic groups become exposed to water upon a conformational change, the resulting increase in heat capacity (ΔC_p) of the protein, arising from water-ordering near the nonpolar sidechains (Figure 1A), would confer very steep temperature dependence upon the equilibrium constant of that conformational reaction (Schellman et al., 1981). Grossly oversimplifying TRP channel activation as a simple closed $\leq$ > open equilibrium, we imagined that opening of these channels is accompanied by the exposure to water of hydrophobic residues buried in the closed state—roughly 20 such residues per subunit in the tetrameric channel would do the trick. The residues need not be localized in a particular domain but instead could be distributed throughout the protein. Crucially, this "ΔC_p" idea mathematically entwines enthalpy and entropy components of the conformational free