economical approach to defense. In infections, the damage caused by pathogenicity must be offset against the costs of deploying an immune response (*18*). Rather than striving to completely eliminate infections, the immune system might manage a persistent infection at a low and nondamaging level (*19*). This is analogous to the concept of "economic injury level" in agricultural pest control, whereby pests are not eradicated but are suppressed to a threshold where the cost of pest-driven damage is lower than the cost of further control. MAMPs indicate the presence of microbes, but if the microbes are doing little or no damage to the host, the cost of immune activity may exceed the benefit of clearing the infection. The presentation of damage-triggered danger signals in conjunction with MAMPs, however, indicates a severe infection that justifies the expense of a defense response.

The immune system cannot afford to be rampantly stimulated by benign foreign molecules, but needs to determine whether a signal indicates microbial nonself or danger. Insights from insect immunity point to the possibility that both types of elicitors may be important in combination. Perhaps neither MAMPs nor danger signals are by themselves a sufficient cue for optimal regulation of host immunity, but together they constitute a reliable indicator for modulating the immune response to yield both effective defense and homeostatic regulation of commensal microbial communities (see the figure). In this scenario, the two models of immune activation (*1*, *2*) as triggered by nonself versus by danger signals need not be considered mutually exclusive, but could be merged into a single model where the host reads the balance of signals to mount an appropriate immunological reaction.This measuring of signals may allow the host to effectively fight an infection, while maintaining healthy relationships with commensals.

**References and Notes**


1. R. Medzhitov, C. A. Janeway Jr., *Science* **296**, 298 (2002).
2. P. Matzinger, *Science* **296**, 301 (2002).
3. E. Ragan, C. An, H. Jiang, M. Kanost, in *Insect Infection and Immunity*, J. Rolff, S. Reynolds, Eds. (Oxford Univ. Press, Oxford, 2009), pp. 34–48.
4. R. J. Dillon, V. M. Dillon, *Annu. Rev. Entomol.* **49**, 71 (2004).
5. R. E. Vance, R. R. Isberg, D. A. Portnoy, *Cell Host Microbe* **6**, 10 (2009).
6. J. H. Ryu, S. H. Kim, H. Y. Lee, J. Y. Bai, Y. D. Nam, J. W. Bae, D. G. Kee, S. C. Shin, E. M. Ha, W. J. Lee, *Science* **319**, 777 (2008).
7. N. H. Salzman, K. Hung, D. Haribhai, H. Chu, J. Karlsson-Sjöberg, E. Amir, P. Teggatz, M. Barman, M. Hayward, D. Eastwood, M. Stoel, Y. Zhou, E. Sodergren, G. M. Weinstock, C. B. Williams, N. A. Bos, *Nat. Immunol.* **11**, 76 (2010).
8. A. Zaidman-Rémy, M. Hervé, M. Poidevin, S. Pili-Floury, M. S. Kin, D. Blanot, B. H. Oh, R. Ueda, D. Mengin-Lecreulx, B. Lemaitre, *Immunity* **24**, 463 (2006).
9. C. Anselme, V. Pérez-Brocal, A. Vallier, C. Vincent-Monegat, D. Charif, A. Latorre, A. Moya, A. Heddi, *BMC Biol.* **6**, 43 (2008).
10. J. Wang, Y. Wu, G. Yang, S. Aksoy, *Proc. Natl. Acad. Sci. U.S.A.* **106**, 12133 (2009).
11. P. Liehl, M. Blight, N. Vodovar, F. Boccard, B. Lemaitre, *PLoS Pathog.* **2**, e56 (2006).
12. E.-M. Ha, C.-T. Oh, Y. S. Bae, W.-J. Lee, *Science* **310**, 847 (2005).
13. N. Buchon, N. A. Broderick, M. Poidevin, S. Pradervand, B. Lemaitre, *Cell Host Microbe* **5**, 200 (2009).
14. A. Vilcinskas, *Virulence* **1**, 206 (2010).
15. R. Lande, J. Gregorio, V. Facchinetti, B. Chatterjee, Y. H. Wang, B. Homey, W. Cao, Y. H. Wang, B. Su, F. O. Nestle, T. Zal, I. Mellman, J. M. Schröder, Y. J. Liu, M. Gilliet, *Nature* **449**, 564 (2007).
16. S. Gambaryan, A. Kobsar, N. Rukoyatkina, S. Herterich, S. Geiger, A. Smolenski, S. M. Lohmann, U. Walter, *J. Biol. Chem.* **285**,18352 (2010).
17. B. Lemaitre, J. M. Reichhart, J. A. Hoffmann, *Proc. Natl. Acad. Sci. U.S.A.* **94**, 14614 (1997).
18. B. P. Lazzaro, T. J. Little, *Philos. Trans. R. Soc. Lond. B Biol. Sci.* **364**, 15 (2009).
19. E. R. Haine, Y. Moret, M. T. Siva-Jothy, J. Rolff, *Science* **322**, 1257 (2008).

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CELL BIOLOGY

# Phosphatase Inhibition Delays Translational Recovery

A small molecule, guanabenz, increases survival of cells under stress.

**R. Luke Wiseman[1] and Jeffery W. Kelly[1,2,3]**

In cells, various signaling pathways help to maintain proteostasis—the proper concentrations, folding, and function of proteins. When a cell is under stress, upstream "stress sensors" within these pathways are activated, initiating a signaling cascade that minimizes the misfolding and aggregation of proteins, which can lead to disease (*1*–*3*). Stress sensors often respond to the accumulation of misfolded proteins within specific cell compartments by activating the transcription of proteostasis components, such as enzymes and "chaperone" proteins that assist with folding or by attenuating new protein synthesis. The propagation of stress-response signaling is often mediated by phosphorylation, or the addition of a phosphate group to the stress sensor and/or downstream signaling components. Because of the central importance of stress signaling pathways in maintaining the integrity of the cellular proteome, manipulating these pathways has become an attractive strategy for preventing the protein misfolding linked to numerous human diseases (*4*, *5*).

On page 91 of this issue, Tsaytler *et al.* take a step toward this goal. They demonstrate that the selective inhibition of a stress-induced phosphatase complex involved in a stress-signaling pathway that controls proteostasis in the endoplasmic reticulum (ER) increases cellular survival (*6*). This novel approach demonstrates the potential for manipulating stress-signaling cascades through direct targeting of a property that emerges from these complex signaling cascades (an emergent property), allowing for specific manipulation of stress signaling that is independent of pathways involved in general cellular homeostasis.

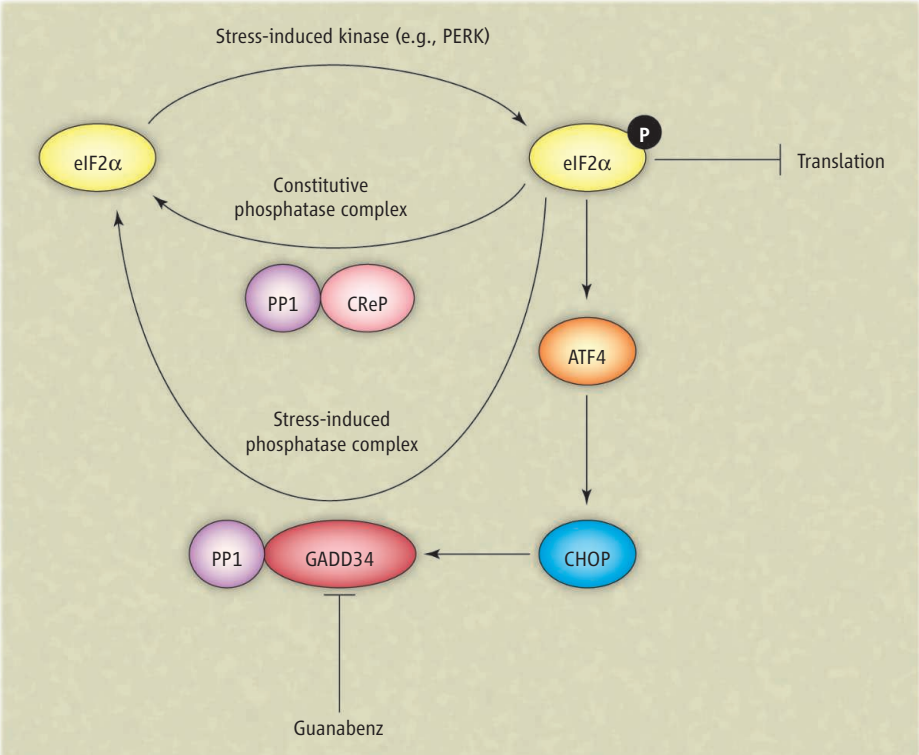
One of the best-characterized stress-responsive signaling pathways is called the unfolded protein response. It maintains proteostasis in the ER, where the secreted proteome is folded (*1*, *7*). The unfolded protein response comprises integrated signaling pathways that emanate from three transmembrane stress sensors localized in the ER: IRE1, ATF6, and PERK. These sensors are activated by the accumulation of misfolded proteins within the ER lumen. Activation of IRE1 and ATF6 enhances protein folding capacity within the ER lumen

[1]Department of Molecular and Experimental Medicine, Scripps Research Institute, La Jolla, CA 92037, USA. [2]Department of Chemistry, Scripps Research Institute, La Jolla, CA 92037, USA. [3]Skaggs Institute for Chemical Biology, Scripps Research Institute, La Jolla, CA 92037, USA. E-mail: wiseman@scripps.edu; jkelly@scripps.edu

through transcriptional up-regulation of ER chaperones, folding enzymes, and components of the ER-associated degradation pathway. Activation of PERK induces the selective phosphorylation of the α subunit of eukaryotic initiation factor 2 (eIF2α), which results in attenuation of ribosomal translation, thus reducing the load on the ER proteostasis network (see the figure). Phosphorylated eIF2α also selectively promotes translation of the transcription factor ATF4, which targets stress-responsive genes, including the transcription factor CHOP. In turn, CHOP induces transcription of the protein phosphatase 1 regulatory subunit GADD34 (also called PPP1R15A), which binds to the catalytic subunit of protein phosphatase 1. A heterodimer consisting of the protein phosphatase 1 catalytic subunit and GADD34 regulatory subunit selectively dephosphorylates eIF2α, thus providing a negative feedback loop in the PERK signaling cascade. This loop turns off signaling and facilitates restoration of ribosomal translation after ER stress.

Through expert detective work, Tsaytler *et al*. correctly suspected that the small molecule guanabenz selectively binds to GADD34. This prevents the assembly of the active protein phosphatase 1–GADD34 heterodimer and delays recovery from stress-induced translational attenuation (see the figure). Although guanabenz binds the stress-induced regulatory subunit GADD34, it does not bind the constitutively expressed eIF2α regulatory subunit CReP (PPP1R15B), which similarly forms a heterodimer with protein phosphatase 1 and catalyzes eIF2α dephosphorylation. Thus, guanabenz only slows eIF2α dephosphorylation during stress. In addition, guanabenz does not induce cytotoxicity on its own, as do other inhibitors of eIF2α phosphatases, such as calyculin A (*8*). Instead, guanabenz provides a boost to the endogenous PERK signaling pathway, extending the duration of translational attenuation and enabling the recovery from ER stress by targeting an emergent property of the PERK signaling cascade.

Research has shown that PERK signaling is critical for maintaining ER proteostasis in pancreatic beta cells expressing high levels of insulin (*9*). Thus, modulation of PERK signaling may alleviate ER stress associated with increased insulin production. Consistent with this hypothesis, Tsaytler *et al*. demonstrated that guanabenz dose-dependently protects cells against ER stress induced by the overexpression of a destabilized mutant insulin protein. Similarly, recent evidence suggests that increased activity of IRE1 and/or PERK signaling enhances mutant enzyme folding capacity within the ER, increasing trafficking and function of destabilized mutant proteins associated with lysosomal storage diseases (*10*). It would be interesting to see whether prolonged PERK signaling, enabled by guanabenz, is sufficient to restore mutant lysosomal enzyme proteostasis. Guanabenz also has the potential to modulate translational attenuation in response to other cellular stresses, such as amino acid deprivation, heme deficiencies, and oxidative insults that activate alternative eIF2α kinases (*11*, *12*). If so, it could potentially enhance cellular survival in a manner analogous to that observed by Tsaytler *et al*.




**Enhancing proteostasis.** Guanabenz selectively inhibits the GADD34-mediated negative feedback loop of PERK signaling through direct binding to GADD34, preventing its association with protein phosphatase 1 (PP1). As a result, guanabenz extends translational attenuation in response to ER stress, increasing chaperone-to-substrate ratios and alleviating protein misfolding within the ER lumen.

Multiple groups have sought, or are seeking, small molecules that can activate a specific arm or arms of the unfolded protein response (*4*), and there is optimism that these strategies will lead to the development of drugs targeting numerous maladies. Molecules such as guanabenz could represent an interesting category of these so-called proteostasis regulators, because they might be able to make up for insufficient stress-responsive signaling by prolonging its duration. The regulation of stress-response pathways is only partially understood, and it is likely that other strategies for extending the duration of stress responses exist, such as extending the duration of the cytosolic heat-shock response by modulating histone deacetylase (*13*). These agents could exhibit additive or synergistic properties with other stress-signaling pathway activators (*10*). This strategy, targeting an emergent property of stress-responsive signaling, represents an elegant opportunity for adapting proteostasis to treat human diseases.

**References**

1. D. Ron, P. Walter, *Nat. Rev. Mol. Cell Biol.* **8**, 519 (2007).
2. R. I. Morimoto, *Genes Dev.* **22**, 1427 (2008).
3. C. M. Haynes, D. Ron, *J. Cell Sci.* **123**, 3849 (2010).
4. W. E. Balch, R. I. Morimoto, A. Dillin, J. W. Kelly, *Science* **319**, 916 (2008).
5. E. T. Powers, R. I. Morimoto, A. Dillin, J. W. Kelly, W. E. Balch, *Annu. Rev. Biochem.* **78**, 959 (2009).
6. P. Tsaytler, H. P. Harding, D. Ron, A. Bertolotti, *Science* **332**, 91 (2011); 10.1126/science/science.1201396.
7. M. Schröder, R. J. Kaufman, *Annu. Rev. Biochem.* **74**, 739 (2005).
8. M. Boyce *et al*., *Science* **307**, 935 (2005).
9. A. Volchuk, D. Ron, *Diabetes Obes. Metab.* **12** (suppl. 2), 48 (2010).
10. T. W. Mu *et al*., *Cell* **134**, 769 (2008).
11. R. C. Wek, H.-Y. Jiang, T. G. Anthony, *Biochem. Soc. Trans.* **34**, 7 (2006).
12. H. P. Harding *et al*., *Mol. Cell* **11**, 619 (2003).
13. S. D. Westerheide, J. Anckar, S. M. Stevens Jr., L. Sistonen, R. I. Morimoto, *Science* **323**, 1063 (2009).

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# Phosphatase Inhibition Delays Translational Recovery

R. Luke Wiseman and Jeffery W. Kelly

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