their major threat from illegal hunters, who kill them for their tusks and for food. Leader-Williams and co-authors argue the case for legal hunting of male elephants maintaining elephant numbers. Although serious concerns are raised elsewhere over the implications of losing older males from endangered elephant populations because of their particular importance in breeding (*2*), male elephants were not the subjects of our paper. However, we emphasize in general terms the danger of removing older, more experienced individuals from social groups in endangered populations of advanced social mammals, because the situation for female elephants has obvious parallels elsewhere (*1, 3*). In many whale species, for example, large-brained, long-lived females also form closely bonded social groups (*3, 4*), and examination of the size of individuals in commercial catches suggests that the largest may have been selectively taken (*5*). Given that our results indicate that groups may rely on older members for their store of social knowledge, in the absence of information on specific cases we would urge caution over any activity that results in their removal from endangered populations.

KAREN MCCOMB,[1,2]* CYNTHIA MOSS,[2] SARAH DURANT[3]

[1]Experimental Psychology, School of Biological Sciences, University of Sussex, Brighton BN1 9QG, UK. [2]Amboseli Elephant Research Project, African Wildlife Foundation, Box 48177, Nairobi, Kenya. [3]Institute of Zoology, Zoological Society of London, Regent's Park, London NW1 4RY, UK

*To whom correspondence should be addressed. E-mail: karenm@biols.susx.ac.uk

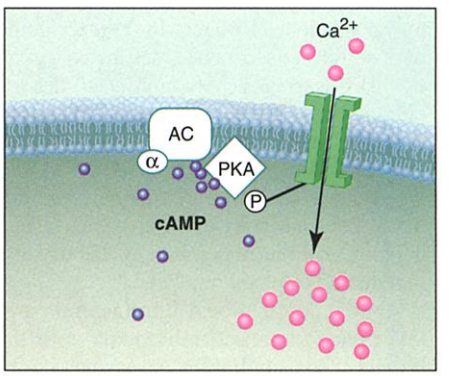
References and Notes

1. K. McComb, C. Moss, S.M. Durant, L. Baker, S. Sayialel, *Science* **292**, 491 (2001).
2. A. Dobson, J. Poole, in *Behavioural Ecology and Conservation Biology*, T. M. Caro, Ed. (Oxford Univ. Press, Oxford, 1998), pp. 193–208.
3. E. Pennisi, *Science* **292**, 417 (2001).
4. R. C. Connor, in *Cetacean Societies, Field Studies of Dolphins and Whales*, J. Mann *et al.*, Eds. (Univ. of Chicago Press, Chicago, 2000), pp. 199–218.
5. P.T. Stevick, *Mar. Mamm. Sci.* **15**, 725 (1999).

The Fourth Dimension in Cellular Signaling

A KEY QUESTION IN CELL BIOLOGY IS HOW signaling specificity is achieved. The authors of two Perspectives (*1, 2*) reflected on the difficulties of relaying information from a staggering number of extracellular receptors through a small number of intracellular signaling molecules. A classic example is the cAMP (cyclic adenosine 3′,5′-monophosphate) pathway, in which the binding of hormones, neurotransmitters, and odorants to receptors triggers the production of cAMP. This soluble messenger activates downstream effectors such as cAMP-dependent protein kinase (PKA), which differentially phosphorylates hundreds of cellular targets. In the past few years, the primary focus of research into specificity has been on macromolecular signaling complexes that effectively organize proteins (from receptors to targets) into two-dimensional arrays at the surface membrane. In their report, M. A. Davare and colleagues provide an elegant example, demonstrating that β_2 adrenergic receptors assemble in complexes with L-type Ca^{2+} channels, and that signals from the receptor to the channels (transmitted through the cAMP pathway) are localized (Reports, "A β_2 adrenergic receptor signaling complex assembled with the Ca^{2+} channel $Ca_v1.2$," 6 Jul., p. 98).

Although this sort of organization is essential for signaling specificity, a pressing question remains, what happens to cAMP?



Keeping cAMP close by. Perhaps cAMP is "channeled" between adenylyl cyclase (AC) and protein kinase (PKA) in certain macromolecular signaling complexes (see the response by Hall and Hell). (α, a G protein; P, phosphate)

Laporte *et al.* (*2*) touch on the problem: "[I]t is commonly assumed that activation of ion channels through second messenger–dependent kinases such as PKA can be sensed anywhere within the cell because of the rapid diffusion of small second messenger molecules. The findings from Devare *et al.* certainly challenge the generality of this assumption." We would like to point out that this vital issue, diffusional spread of small molecules, has received some attention. In two cell types studied by Devare and colleagues, rat hippocampal neurons and human embryonic kidney cells, there are multiple lines of evidence for diffusional restrictions under the surface membranes, giving rise to chemical compartmentalization (*3, 4*). The latter study dealt specifically with cAMP. Diffusional restrictions have also been observed in cardiac myocytes (*5*). It is worth noting that without these restrictions, cAMP concentrations right next to adenylyl cyclase (the enzyme that produces cAMP) would not be high enough to activate PKA, unless the entire cell filled with cAMP (*4*). This, of course, would acti-

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vate every PKA molecule in the cell. The morphological basis for diffusional restrictions is a fascinating subject for future study. With many of the necessary tools at hand, the time is right to expand our focus beyond the bounds of two-dimensional complexes to actual, four-dimensional signals.

JEFFREY W. KARPEN,* THOMAS C. RICH
Department of Physiology and Biophysics, University of Colorado Health Sciences Center, Denver, CO 80262, USA
*To whom correspondence should be addressed. E-mail: jeffrey.karpen@uchsc.edu

References and Notes
1. C. R. Weston, R. J. Davis, *Science* **292**, 2439 (2001).
2. S. A. Laporte, R. H. Oakley, M. G. Caron, *Science* **293**, 62 (2001).
3. K. Svoboda, D. W. Tank, W. Denk, *Science* **272**, 716 (1996).
4. T. C. Rich *et al.*, *J. Gen. Physiol.* **116**, 147 (2000).
5. W. J. Lederer, E. Niggli, R. W. Hadley, *Science* **248**, 283 (1990); N. Leblanc, J. R. Hume, *Science* **248**, 372 (1990); J. H. Bridge, J. R. Smolley, K. W. Spitzer, *Science* **248**, 376 (1990).

Response

A macromolecular signaling complex is formed between the L-type Ca^{2+} channel $Ca_v1.2$ and the β_2 adrenergic receptor, heterotrimeric G proteins, an adenylyl cyclase (AC), and PKA, which regulate the activity of this channel by means of cAMP in a highly localized manner (*1*). Karpen and Rich perceptively ask: How can signaling by diffusible cAMP be localized so precisely?

As they showed earlier by mathematical analysis (*2*), simple diffusion cannot account for activation of PKA, even if it is close to the AC. Assuming a maximal catalytic rate of 59 per second for cAMP production by AC and a diffusion rate of 3 x 10^{-6} cm^2 per second, cAMP concentration will not exceed 5 nM in a distance of 10 nm from the catalytic site of the fully active AC if diffusion is unrestricted (*2*). Half-maximal activation of the various PKA isoforms is usually observed with a cAMP concentration in the range of 100 nM. ACs and PKA holoenzymes have molecular masses above 100 kilodaltons. The size of a protein with such a molecular mass is typically in the range of 5 nm. It is, therefore, likely that the distance between the catalytic site of the AC and the cAMP binding sites of PKA in our channel complex is 10 nm, if not larger.

The authors hypothesized in their work on cAMP-gated ion channels that elements of the endoplasmic reticulum might be localized underneath the plasma membrane, thereby limiting the diffusion of cAMP away from the AC and their channels in their system (*2*). This is a valid hypothesis that may also be true for the $Ca_v1.2$ channel complex in the heart. Cardiac $Ca_v1.2$ is precisely juxtaposed to elements of the sarcoplasmic reticulum (*3*) that could substantially limit cAMP diffusion by restricting it to two dimensions. However, the situation might be different in the system we studied, the neuronal cell body, and we propose here an alternative model for diffusional restriction of cAMP. We hypothesize that the AC and PKA might be arranged in such a way that cAMP is "channeled" from the AC to PKA by a molecular mechanism, thereby dramatically increasing the likelihood that newly synthesized cAMP will bind to PKA. Models of molecular product channeling are well established for metabolic enzymes, and mechanisms range from the formation of actual tunnels by proteins to electrostatic channeling, due to surface charges on the proteins that attract the substrate (*4*). A molecular mechanism of cAMP channeling would make activation of PKA by a neighboring AC more effective and spatially more restricted.

DUANE D. HALL, JOHANNES W. HELL
Department of Pharmacology, University of Iowa, Iowa City, IA 52242, USA. E-mail: johannes-hell@uiowa.edu

References and Notes
1. M. A. Davare *et al.*, *Science* **293**, 97 (2001).
2. T. C. Rich *et al.*, *J. Gen. Physiol.* **116**, 147 (2000).
3. D. R. L. Scriven, P. Dan, E. D. W. Moore, *Biophys. J.* **79**, 2682 (2000).
4. C. C. Hyde *et al.*, *J. Biol. Chem.* **263**, 17857 (1988); A. H. Elcock *et al.*, *J. Mol. Biol.* **262**, 370 (1996); C. Velot, P. A. Srere, *J. Biol. Chem.* **275**, 12926 (2000).

CORRECTIONS AND CLARIFICATIONS

REPORTS: "A multispecies overkill simulation of the end-Pleistocene megafaunal mass extinction" by J. Alroy (8 Jun., p. 1893). In figure 1, some of the black or gray lines representing extant or extinct species, respectively, were the wrong color. The number and shape of the lines are correct, and the statistics are unaffected by the color error. The correct figure appears here.