This result is precisely what is predicted by the adapting reciprocal inhibition model, removing the only major empirical obstacle to this explanation of rivalry alternations.

As well as providing crucial evidence to validate the adaptation model architecture, this study raises important questions about visual consciousness. When the suppressed eye is probed, observers are apparently able to respond to a stimulus they are not consciously aware of, with accuracy levels above chance. Of course, it is conceivable that the probe presentation itself causes a reversal of dominance, enabling it to be detected, though features of the experimental design make this explanation unlikely. Taken at face value, this effect is very similar to the clinical phenomenon of blindsight [10], but occurring in normal observers (see also [11]). Participants literally communicated information they did not know they had! Such paradoxical behaviour might indicate that visual awareness manifests either after, or in parallel with, the stage at which motor responses are programmed.

The task in the Alais *et al.* [4] study required information about both contrast and spatial location in the suppressed image in order for a correct response. Might other visual attributes, such as colour, orientation, spatiotemporal frequency, motion or higher level properties, also be preserved during suppression? Recent evidence suggests that information about the emotional expression of faces can survive suppression sufficiently to influence subsequent percepts [12]. This suggests that complex processing of visual information can still occur despite complete suppression from conscious awareness. The probe detection technique refined by Alais *et al.* [4] promises to be a powerful tool in unravelling many such aspects of visual consciousness. Perhaps it will encourage a further explosion of research addressing this most elusive aspect of cognitive function.

References

1. Levelt, W.J.M. (1965). On Binocular Rivalry (Assen, The Netherlands: Royal VanGorcum).
2. Crick, F., and Koch, C. (1998). Consciousness and neuroscience. Cereb. Cortex *8*, 97–107.
3. Alais, D., and Blake, R., eds. (2005). Binocular Rivalry (Cambridge, MA: MIT Press).
4. Alais, D., Cass, J., O'Shea, R.P., and Blake, R. (2010). Visual sensitivity underlying changes in visual consciousness. Curr. Biol. *20*, 1362–1367.
5. Wilson, H.R. (2007). Minimal physiological conditions for binocular rivalry and rivalry memory. Vision Res. *47*, 2741–2750.
6. Noest, A.J., van Ee, R., Nijs, M.M., and van Wezel, R.J.A. (2007). Percept-choice sequences driven by interrupted ambiguous stimuli: a low-level neural model. J. Vis. *7*, 10. DOI: 10.1167/7.8.10.
7. Fox, R., and Check, R. (1972). Independence between binocular rivalry suppression duration and magnitude of suppression. J. Exp. Psychol. *93*, 283–289.
8. Stuit, S.M., Cass, J., Paffen, C.L.E., and Alais, D. (2009). Orientation-tuned suppression in binocular rivalry reveals general and specific components of rivalry suppression. J. Vis. *9* (11), 17. DOI: 10.1167/9.11.17.
9. Baker, D.H., and Graf, E.W. (2009). Natural images dominate in binocular rivalry. Proc. Natl. Acad. Sci. USA *106*, 5436–5441.
10. Weiskrantz, L., Warrington, E.K., Sanders, M.D., and Marshall, J. (1974). Visual capacity in the hemianopic field following a restricted occipital ablation. Brain *97*, 709–728.
11. Kolb, F.C., and Braun, J. (1995). Blindsight in normal observers. Nature *377*, 336–338.
12. Adams, W.J., Gray, K.L.H., Garner, M., and Graf, E.W. (2010). High-level face adaptation without awareness. Psych. Science *21*, 205–210.

School of Life and Health Sciences, Aston University, Birmingham, B4 7ET, UK.
E-mail: d.h.baker1@aston.ac.uk

DOI: 10.1016/j.cub.2010.06.010

Cell Polarity: Keeping Worms LeGaL

The protein Lethal giant larvae (LGL) regulates cell polarity in diverse animal models. Now, an LGL orthologue has been identified in the worm *Caenorhabditis elegans* and is shown to function redundantly with a worm-specific polarity protein, PAR-2.

Kenneth E. Prehoda and Bruce Bowerman*

The importance of the nematode *Caenorhabditis elegans* for the study of cell and embryonic polarity is reflected in the naming of several key polarity genes based on their initial identification as *par*titioning-defective mutant loci in these worms [1,2]. These PAR proteins have since been intensely studied over the past decade in several systems, and the mechanisms by which they control cell polarity are becoming clearer. In many systems, the activity of Lethal giant larvae (LGL) is required for PAR-mediated polarity, but so far no orthologue has been identified in worms [3]. Do worms possess LGL or have they developed another polarity mechanism? Recent work by Hoege *et al.* [4] published in *Current Biology* now shows that, surprisingly, the answer to both questions is yes.

Upon fertilization, the one-cell stage *C. elegans* embryo develops cortical polarity along its anterior-posterior axis, which specifies an asymmetric cell division, such that the two daughter cells assume distinct sizes and fates. Polarization of the zygote results in the anterior cortex containing the widely conserved PAR complex, consisting of PAR-3, PAR-6 and an atypical Protein Kinase C (aPKC; called PKC-3 in worms). Two other PAR proteins, PAR-1 and -2, occupy the posterior cortex in *C. elegans,* but their roles are less conserved, with PAR-2 thus far being found only in worms. The anterior PAR complex is used throughout metazoans to polarize diverse cell types, ranging from epithelia and neurons to asymmetrically dividing stem cells [3,5]. Work in many of these systems has shown that the activity of LGL is required for PAR complex mediated polarity. For example, *Drosophila melanogaster* neuroblasts divide asymmetrically by localizing PAR complex proteins to an apical cortical domain [6]. In *Drosophila lgl* mutants, the PAR complex is depolarized, localizing throughout the neuroblast cortex [7]. This phenotype suggests that LGL prevents PAR complex proteins from entering the opposing polarity domain.

As LGL is required for PAR complex-mediated polarity in many systems, it has been surprising that no direct orthologue has been found in worms. Hoege *et al.* [4] have eliminated this curious exception by purifying immunoprecipitated PAR-6 and identifying interacting proteins with mass spectrometry. One PAR-6

interactor had homology to LGL and further investigation showed that it localizes to the posterior cortex, opposite the PAR complex. Surprisingly, however, worm LGL function by itself was not required for polarity or any other process, as LGL mutants display no phenotype. Suspecting that redundancy might mask the function of worm LGL, Hoege *et al.* [4] screened for genetic interactions and found that loss of *C. elegans* LGL function in mutants partially defective for *par-2* resulted in a severe synthetic polarity phenotype with the PAR complex no longer excluded from the posterior cortex, consistent with the LGL loss of function phenotypes observed in other systems. In addition, over-expression of *C. elegans* LGL strongly suppressed the *par-2* mutant phenotype. Thus, worms appear to have developed a redundant method (PAR-2) for restricting PAR complex activity to the anterior cortex, but also retain a role for LGL.

PAR-2 is a RING finger protein that so far appears to be specific to worms. LGL contains a series of WD40 repeats and has no homology with PAR-2. Thus, PAR-2 appears to have evolved convergently to regulate PAR complex polarity. PAR-2 is the dominant pathway for restricting the PAR complex to the anterior cortex, as *lgl* mutants have no apparent defects while *par-2* mutants exhibit a partial depolarization. While PAR-2 has recently been suggested to generate polarity by acting in parallel to Rho-mediated regulation of cortical actomyosin [8], how PAR-2 acts at a molecular level remains unknown. Hoege *et al.* [4], however, do provide new insight into the molecular mechanism by which LGL may influence AP polarity in the worm zygote.

How does LGL regulate polarity? The answer to this question has been difficult to come by. LGL's domain structure provides little insight into function. Early biochemical work in flies identified Myosin II as an interactor [9]. The apparent yeast orthologue, Sro7, regulates exocytosis, a function that would potentially be consistent with a role in polarity [10]. However, neither clue has clarified LGL's role in metazoan polarity. A significant advance in understanding how LGL acts came from its identification as an aPKC substrate in flies and mammals [11–13]. LGL contains three serines at the end of its carboxyl terminus that are phosphorylated by aPKC, and phosphorylation causes LGL to become displaced from the cortex into the cytoplasm. Apparently, phosphorylation induces an intramolecular interaction incompatible with cortical association [14]. A current model for LGL function in *Drosophila* sensory organ precursor (SOP) cells is that LGL holds aPKC in an inactive state until the complex receives an activating signal (Aurora A phosphorylation), at which point LGL is phosphorylated and released from the complex [15]. However, this model is somewhat paradoxical in that LGL and the PAR complex occupy distinct cortical domains and do not overlap at any point in the establishment of PAR polarity. Furthermore, it doesn't explain a key aspect of LGL function as inferred from its loss-of-function phenotype, namely that LGL inhibits Par complex localization.

The aPKC phosphorylation sites are conserved in *C. elegans* LGL, allowing Hoege *et al.* [4] to explore the role of phosphorylation in LGL function. They found when LGL-3A (a non-phosphorylatable variant) was expressed in the one-cell zygote, it was depolarized, localizing to both the anterior and posterior cortex. In contrast, a phosphomimetic construct, LGL-3E, was unable to associate with the cortex. Together these results are consistent with phosphorylation leading to LGL cortical displacement. One might expect the presence of LGL throughout the cortex, as in worms expressing LGL-3A, to displace PAR complex proteins. Instead the anterior PAR complex proteins also became depolarized and present throughout the cortex, overlapping with LGL-3A (when PAR-2 was also inactivated). Overlap of LGL-3A and the Par complex has also been observed in *Drosophila* neuroblasts [16]. This surprising result indicates that the presence of LGL on the cortex appears insufficient to displace the PAR complex — LGL must also be phosphorylatable.

To explain the influence of LGL phosphorylation status on its localization and function, Hoege *et al.* [4] propose a model in which LGL prevents the PAR complex from entering the posterior cortex by recruiting it into the cytoplasm upon phosphorylation. In this model, when

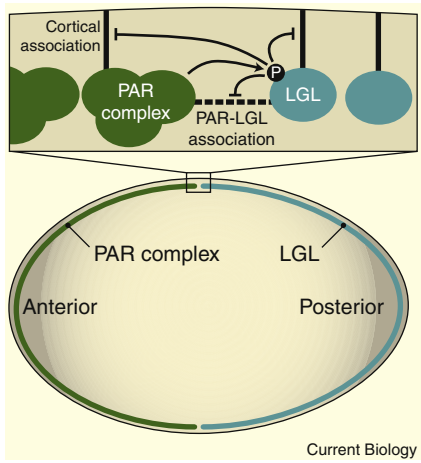

Figure 1. PAR complexes and LGL polarization in *C. elegans*.

The one-cell stage *C. elegans* zygote is polarized into an anterior domain containing the PAR complex and a posterior domain, now shown to contain LGL. The expanded region shows the 'mutual destruction' model proposed by Hoege *et al.* [4]. In this model, phosphorylation of LGL by the PAR complex member aPKC (arrow) causes inhibition of LGL and PAR complex cortical association (thick lines), and inhibition of PAR–LGL association (dashed line).

the PAR complex encounters LGL, which is most likely to occur at the anterior-posterior interface (Figure 1), LGL is phosphorylated, causing their mutual displacement into the cytoplasm. This model is attractive because it explains how LGL might regulate PAR polarity, not just aPKC activity. However, it also has features that will require more work to explain. First, how does LGL phosphorylation lead to mutual displacement of LGL and the PAR complex? While LGL displacement by phosphorylation is likely to occur by a mechanism similar to its fly orthologue, how this would lead to displacement of the PAR complex is entirely unknown. Phosphorylated LGL would have to somehow inhibit the cortical association mechanism of the PAR complex, presumably through direct interaction with the complex as LGL-3E does not associate with the cortex. However, this critical step is complicated by the fact that phosphorylated LGL apparently doesn't bind to the PAR complex. Thus, both the *Drosophila* SOP model for LGL function as an inhibitor of aPKC, and the *C. elegans* model for LGL displacement of the PAR complex into the zygote cytoplasm, each suffer from

a paradox: in *Drosophila* SOP cells, the model does not explain how LGL excludes the PAR complex from the cortex, while in *C. elegans*, LGL must be phosphorylatable to function but phosphorylation decreases its ability to bind the complex it displaces. Thus, while Hoege *et al.* [4] have firmly established a role for LGL in *C. elegans* polarity, how it functions precisely in any system remains an intriguing problem.

References

1. Kemphues, K.J., Priess, J.R., Morton, D.G., and Cheng, N.S. (1988). Identification of genes required for cytoplasmic localization in early C. elegans embryos. Cell *52*, 311–320.
2. Munro, E., and Bowerman, B. (2009). Cellular symmetry breaking during *Caenorhabditis elegans* development. Cold Spring Harb. Perspect. Biol. *1*, a003400.
3. St. Johnston, D., and Ahringer, J. (2010). Cell Polarity in eggs and epithelia: parallels and diversity. Cell *141*, 757–774.
4. Hoege, C., Constantinescu, A.-T., Schwager, A., Goehring, N.W., Kumar, P., and Hyman, A.A. (2010). LGL can partition the cortex of one-cell Caenorhabditis elegans embryos into two domains. Curr. Biol. *20*, 1296–1303.
5. McCaffrey, L.M., and Macara, I.G. (2009). Widely conserved signaling pathways in the establishment of cell polarity. Cold Spring Harb. Perspect. Biol. *1*, a001370.
6. Prehoda, K.E. (2009). Polarization of Drosophila neuroblasts during asymmetric division. Cold Spring Harb. Perspect. Biol. *1*, a001388.
7. Lee, C.Y., Robinson, K.J., and Doe, C.Q. (2006). Lgl, Pins and aPKC regulate neuroblast self-renewal versus differentiation. Nature *439*, 594–598.
8. Zonies, S., Motegi, F., Hao, Y., and Seydoux, G. (2010). Symmetry breaking and polarization of the C. elegans zygote by the polarity protein PAR-2. Development *137*, 1669–1677.
9. Strand, D., Jakobs, R., Merdes, G., Neumann, B., Kalmes, A., Heid, H.W., Husmann, I., and Mechler, B.M. (1994). The Drosophila lethal(2)giant larvae tumor suppressor protein forms homo-oligomers and is associated with nonmuscle myosin II heavy chain. J. Cell Biol. *127*, 1361–1373.
10. Gangar, A., Rossi, G., Andreeva, A., Hales, R., and Brennwald, P. (2005). Structurally conserved interaction of Lgl family with SNAREs is critical to their cellular function. Curr. Biol. *15*, 1136–1142.
11. Betschinger, J., Mechtler, K., and Knoblich, J.A. (2003). The Par complex directs asymmetric cell division by phosphorylating the cytoskeletal protein Lgl. Nature *422*, 326–330.
12. Plant, P.J., Fawcett, J.P., Lin, D.C., Holdorf, A.D., Binns, K., Kulkarni, S., and Pawson, T. (2003). A polarity complex of mPar-6 and atypical PKC binds, phosphorylates and regulates mammalian Lgl. Nat. Cell Biol. *5*, 301–308.
13. Yamanaka, T., Horikoshi, Y., Sugiyama, Y., Ishiyama, C., Suzuki, A., Hirose, T., Iwamatsu, A., Shinohara, A., and Ohno, S. (2003). Mammalian Lgl forms a protein complex with PAR-6 and aPKC independently of PAR-3 to regulate epithelial cell polarity. Curr. Biol. *13*, 734–743.
14. Betschinger, J., Eisenhaber, F., and Knoblich, J.A. (2005). Phosphorylation-induced autoinhibition regulates the cytoskeletal protein Lethal (2) giant larvae. Curr. Biol. *15*, 276–282.
15. Wirtz-Peitz, F., Nishimura, T., and Knoblich, J.A. (2008). Linking cell cycle to asymmetric division: Aurora-A phosphorylates the Par complex to regulate Numb localization. Cell *135*, 161–173.
16. Atwood, S.X., and Prehoda, K.E. (2009). aPKC phosphorylates Miranda to polarize fate determinants during neuroblast asymmetric cell division. Curr. Biol. *19*, 723–729.

Institute of Molecular Biology, University of Oregon, 1370 Franklin Boulevard, Eugene, OR 97403, USA.
*E-mail: bbowerman@molbio.uoregon.edu

DOI: 10.1016/j.cub.2010.06.011