GENE REGULATION

Infiltrating the network

An inventive approach to the engineering of gene expression networks represents an important step in modeling such complex processes as embryonic pattern formation.

Embryonic patterning is vital to the developmental process, and anybody who has studied developmental biology is aware of the byzantine temporal and spatial regulatory processes involved, with extensive genetic cross-talk and multiple gradients of expression interacting with each other to establish boundaries where other expression events can take place. Modeling these systems is essential to deciphering them, according to Mark Isalan, an investigator in the lab of Luis Serrano at the European Molecular Biology Laboratory (EMBL). "You've got all these beautiful networks," he says, "but maybe the limit of whether you understand them is whether you can rebuild them from first principles."

Isalan's team recently demonstrated a new breakthrough on this front, assembling an artificial gene network that replicates some key processes of *Drosophila* embryo pattern formation (Isalan *et al.*, 2005a). They designed several bead-conjugated transcription units using a technique for the linkage of expression-ready PCR products to magnetic beads, with each unit containing a T7 or SP6 promoter and a gene encoding one of three different synthetic transcription factors.

To simulate the early-stage *Drosophila* embryo, Isalan's group magnetically arranged their beads into discrete zones in a chamber filled with low–melting point agarose, permitting the passive diffusion of transcription factors. The arrangement was designed to spatially restrict expression for each transcription factor to different regions of the 'embryo', in a manner that roughly mirrors normal expression of the developmental genes *hunchback*, *giant* and *Krüppel*. In initial experiments, the only other restriction on the range of expression resulted from the distribution of T7 and SP6 RNA polymerases and diffusion within the chamber. This resulted in the emergence of some crude patterns; however, the addition of further layers of regulation considerably sharpened and extended the longevity of these patterns. By modifying the transcription units so that the transcription factors could regulate each other's expression (for instance, expression of gene *A* represses expression of genes *B* and *C*) and reducing protein half-life by adding proteases to the chamber, the patterns were considerably sharpened. A computer simulation, designed as a companion to the *in vitro* system, closely mirrored the patterns they observed under various experimental conditions.

The authors found that this system effectively illustrated several key principles of pattern formation, including the importance of cross-repression. Boston University researcher James Collins, whose own work has dealt extensively with genetic networks, expressed admiration for this technique: "I think that the [focus of this] field, going forward, is not going to be creating novel circuits with useful functions in biotechnology, but more… designing isolated circuits with particular designs to explore natural biological phenomena underlying gene expression and in cellular function. And in this case, I think [Serrano's team] did a really nice job."

Although these *in vitro* studies have yielded interesting results, Isalan's real hope is to promptly move this system into cell-based studies, using a related technique described by his group in another recent paper (Isalan *et al.*, 2005b). "I think it will be much more relevant to try and engineer networks in multicellular systems," he says, "and you can use this as a tool to program individual cells to do things in space, and make more physiologically relevant systems." In the meantime, however, he hopes that this will demonstrate some of the potential applications of his team's magnetic bead–based strategy. "I hope that people might try playing with it, because it's quite a neat little tool to localize genes in particular places, and I think you can do a lot with it."

Michael Eisenstein

RESEARCH PAPERS

Isalan, M. *et al.* Engineering gene networks to emulate *Drosophila* embryonic pattern formation. *PLoS Biol.* **3**, 1–9 (2005a).

Isalan, M. *et al.* Localized transfection on arrays of magnetic beads coated with PCR products. *Nat. Methods* **2**, 113–118 (2005b).