Evolutionary biology

## Channels of resistance

Two studies reported in this issue provide striking examples of how biologists are getting to grips with adaptive diversification at the molecular level. They deal with two very different animals — one a marine invertebrate and the other a terrestrial vertebrate.

The softshell clam (*Mya arenaria*) occurs around the Atlantic coast of North America. The clams can become contaminated with saxitoxin, the cause of paralytic shellfish poisoning in humans and economic losses to the shellfish industry. The toxin is produced by 'red tide' algae and finds its way into the clams when the algae are ingested. V. Monica Bricelj *et al.* (*Nature* **434,** 763–767; 2005) show that clams from areas subject to recurrent red tides are relatively resistant to the toxin and tend to accumulate it in their tissues. But clams from unaffected areas have low resistance when exposed to the toxin in the laboratory. These differences were mirrored by the sensitivity of isolated clam nerve-trunks exposed to the toxin *in vitro*.

To investigate the underlying molecular mechanism, Bricelj *et al.* sequenced the genomic region encoding a putative voltage-gated sodium channel. Such channels sit in cell membranes and regulate ion flow. The authors found a single mutation that correlated with resistance to the toxin, and that results in replacement of a glutamic acid by aspartic acid at a site previously implicated in the binding of saxitoxin. When introduced into a channel from rat brain, this mutation did not affect ion conductance. But the sensitivity of the channel to saxitoxin was greatly reduced owing to a large decrease in the binding affinity of the toxin at the channel pore.

Saxitoxin produced by red-tide algae probably acts as a potent selective agent on the clams, leading to genetic adaptation, the target of selection being genetic variation at a single site in an ion channel.

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But this phenomenon is not unique to clams. Saxitoxin is related to another nerve poison called tetrodotoxin (TTX). In some populations of the newt *Taricha granulosa*, individuals accumulate large amounts of TTX in their skin as a defence against garter snakes (*Thamnophis sirtalis*; pictured). As a result, the snakes that prey on toxic newts have evolved high levels of resistance to the toxin. Shana L. Geffeney *et al.* (*Nature* **434,** 759–763; 2005) show that variation in the level of resistance of garter snakes co-evolving with their newt prey can be traced to molecular changes that affect the binding of TTX to — you guessed it — a sodium channel.

So similar mechanisms underlie the adaptation of both softshell clams and garter snakes to regular neurotoxin exposure. Evolution is baroque in its many aspects, but is sometimes more predictable than we imagine. **Rory Howlett**

of activation along the head–tail body axis that tightly reflects their order in the cluster[3]. A related phenomenon is observed for the *tinman*, *bagpipe* and *ladybird late* genes in the 93DE cluster of fruitflies[4], hinting that 'temporal collinearity' is a widespread mechanism used to coordinate the expression of clustered genes in time. Quantitative collinearity — collinear variations in activation efficiencies, rather than in the temporal order of expression — is also observed for some Hox genes. In the case of Rhox genes, however, the relative activation efficiencies seem to be coordinated only at their peak values, and are not maintained throughout sperm development. So they may merely reflect the genes' time-delayed activation.

The coordinated activation of the Rhox genes also has features in common with the developmental switches described for genes of the β-globin cluster. Embryonic globins are replaced by fetal and then adult forms[5] in cohorts of maturing blood cells, just as Rhox genes are expressed in a temporal order that coincides with the stages of Sertoli-cell maturation. Thus, both cell types progressively acquire different but related functions as they differentiate.

So how did the Rhox genes evolve? MacLean *et al.* propose[2] that — like other homeobox genes — they arose through the repeated duplication of an ancestral gene, and then adopted distinct properties through positive selection. Why, though, have they remained clustered? Duplication is a powerful way to diversify gene functions. But it may also induce some imbalance in gene dosage, which can be detrimental to biological processes that are regulated by delicate concentration equilibria. Keeping duplicated genes in the same genomic region may help to deal with this problem by allowing better coordination of their activation. This could be achieved, for example, if several genes were to compete for the same enhancer regions (DNA sequences that promote gene expression). Enhancers might subsequently evolve into 'switches', selecting the right gene for a given task at a given time. Such sharing of regulatory elements may have constrained the Rhox genes to remain clustered.

However, MacLean *et al.* point out that DNA sequences near the *Rhox5* gene can specifically direct its expression in Sertoli cells, suggesting that some Rhox genes are individually rather than coordinately controlled — at least as far as their cell-type-specific expression goes. The same is true of some Hox genes: when removed from their cluster and placed elsewhere in the genome, they frequently continue to display fairly good spatial expression specificity. But they often lack precise temporal control. So it seems plausible that a strong evolutionary constraint maintaining both Rhox and Hox gene clustering is the requirement to activate them in a temporal sequence[3], as determined by the architecture of these gene clusters. Further research is needed to determine whether androgens (or other hormones) contribute to this temporal activation.

MacLean and colleagues' discovery[2] of the Rhox genes and their collinear organization and expression provides yet another example of a complex genomic region that somehow coordinates the expression of its genes to regulate a delicate and essential process. It will be interesting to learn more about how the peculiar architecture of the Rhox cluster influences the regulation of these genes and their precise functions, not only throughout germ-cell development, but also in ovaries and the placenta. ■

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