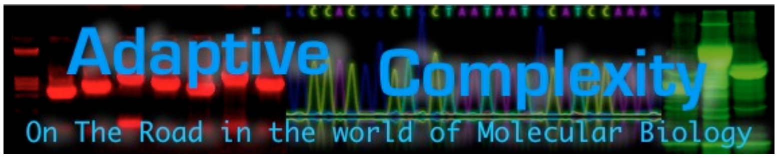

Hi-resolution Evolution: A Repeat Performance

By Michael White | January 29th 2010 10:41 AM | 13 comments | Print | E-mail | Track Comments

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Michael White

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What kinds of genetic changes are required to evolve significant changes in body shape and size? The availability of affordable, state-of-the-art DNA sequencing and array technology has made it possible to study evolution at a level of molecular detail inaccessible just a decade ago.

Researchers are uncovering the details of molecular evolution, and answering some interesting questions along the way, such as the relative importance of changing genes themselves versus changing how those genes are regulated. Recall how genetically similar we are to chimps, and yet how morphologically different we are. Or think of the fact that humans have ~22,000 genes (give or take a few thousand) - not much more than a nematode worm that has only 1000 cells. Intuitively, this seems to suggest that the key difference between worms and chimps and humans is not what genes you have, but how those genes are regulated. However, we can't just assume this conclusion; we have to go look and see whether protein-coding or non-protein-coding DNA changes are the favored means of achieving evolutionary change. (Not surprsingly, the answer so far seems to be *both*.)

In any case, there are many more questions like this about molecular evolution that we can now begin to answer with the modern experimental and computational tools available to biologists. But even with the slew of modern tools we have available, many of today's biologists are studying evolution by taking a page out of Darwin's playbook: they are studying evolutionary change at its most incipient point, at the level of differences between members of the same species. In other words, variation between populations of the same species is just evolution on a small scale, because different populations of the same species are what, in some cases, give rise to new species.

What's the advantage to studying variation within a species? In a word, sex. You can cross-breed two different members of the same species, but not, in most cases two members of different species. (For one approach to getting around the problem of species barriers, check out this research.)

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ABOUT MICHAEL

Welcome to Adaptive Complexity, where I write about genomics, systems biology, evolution, and the connection between science and literature,

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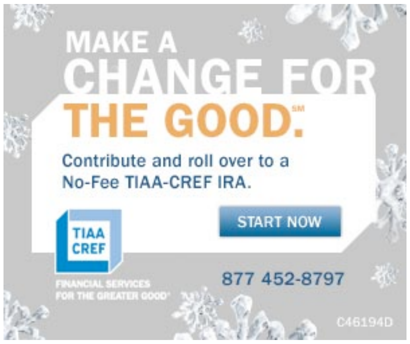

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The Benefits of Breeding

Why is cross breeding so critical? It's a powerful way to identify those genetic changes that actually make a difference. To illustrate this, imagine that you want to know what genetic changes make humans different from chimps. You can compare the two genomes and note the differences, but it's difficult to tell which of those changes are actually meaningful functional changes. We do have ways of guessing at which changes might be selectively neutral changes, and which ones are under the pressure of natural selection, but even if we can identify which regions of the human genome have been under selective pressure to diverge from the chimp genome, we have no idea what the functional effects of those changes are.

What we want to do is the classical problem of genetics: we want to connect genotype with phenotype, genetic variation with variation in physical traits. To do this, we turn to cross-breeding within species. The idea is that you cross two different animals (or plants or yeast) with big differences in your trait of interest, to generate offspring. Those offspring will resemble their parents to different degrees. If you cross a lean breed of pig with a fat breed, you'll get offspring that are lean, fat, or something in between.

Next, you genotype the offspring. Each of the offspring has inherited a different selection of genetic material from the parents. (Remember that each parent, at least in the case of pigs and other diploid organisms, has two complete copies of each chromosome, but passes on only one copy to its offspring.) You determine, at many different points in the genome, which copy each offspring inherited. For example, at position *z* on chromosome 11, one piglet got Dad's copy #1, and Mom's copy #2, the next piglet got Dad's copy #1 and Mom's copy #1, and another piglet got Dad's copy #2 and Mom's copy #1.

In other words, each offspring has received a different genetic inheritance from each parent. The next step is to correlate that genetic inheritance with the phenotype, with the traits in the offspring. Let's say that all of the fattest piglets, in our hypothetical cross, received Dad's copy #1 at position *z* on chromosome 11. We would strongly suspect, then, that Dad's #1 variant of whatever gene is at that position contributes to being a fat pig.

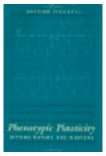
Looking for correlations like this after a cross is a little more complex than I've laid out here, but you get the idea. The result, at the end of this process, is a set of *quantitative trait loci*, or QTL; regions of the genome (usually somewhat large regions, not yet narrowed down to single genes or single genetic variants) that genetically contribute to a particular trait.

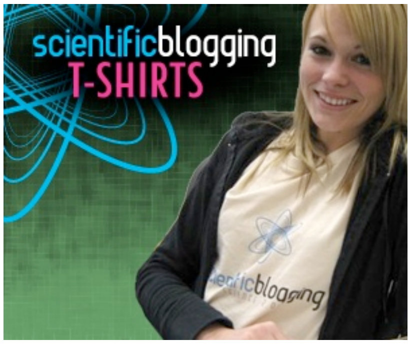
Finding QTL is the reason why studying evolution by studying variation within a single species is so powerful: at the end of the process, you have focused in on only those regions of the genome, only those genetic differences between physically different individuals, that actually contribute to variation in a particular trait. The genomes of any two parents from the same species will be filled with genetic differences, but which differences matter? QTL hunting answers that question.

Repeatable Evolution

A recent study (actually, the lastest in a long series of studies) uses this technique to ask what kinds of genetic changes are used in evolution to remodel skeletons? The animal used here is the three-spine stickleback, a fish that dwells in a variety of aquatic habitats, including marine and freshwater ones. Different populations of sticklebacks have evolved different skeletal morphologies. In particular, marine sticklebacks are heavily armored, a useful trait in their predator-rich environment. Some populations of freshwater sticklebacks have lost much of their armoring, and in fact, this loss of armor has evolved independently multiple times in different populations of marine sticklebacks.

One particular focus of study has been the protective, bony pelvic girdle, which has been lost on some freshwater populations. Researchers in the lab of David Kingsley, at Stanford, are interested in understanding what kinds of evolutionary genetic changes lead to the evolution of pelvic reduction. Following the QTL-mapping procedure I outlined above, they've zeroed in on one particular gene called *Pitx1*. Changes in this gene explain much of the evolutionary changes in pelvic girdle observed





in different stickleback populations. Evolution as acted on the same gene, again and again, to reduce the pelvic armoring that is important for survival in marine environments, but appears to be a hindrance in freshwater ones.

Pitx1 is a developmental regulator gene that is expressed in the developing hindlimbs of many different vertebrate species. The Kingsley lab discovered that populations with reduced pelvic girdles were missing a key regulatory region near the gene that controls expression of *Pitx1*. Without this regulatory region, *Pitx1* is poorly expressed and the fish don't develop a pelvis.

One of the amazing results of this study is repeat performance of evolution: in 9 of 13 pelvic-reduced populations, this key regulatory region was missing, but it was deleted by different mutations. That is, evolution independently hit upon the very same genetic solution for pelvic reduction in these fish 9 out of 13 times. And in 21 stickleback populations with no pelvic reduction, the *Pitx1* regulatory region was always intact, which means that it's critical for maintaining pelvic armor.

Because sticklebacks are so manipulable, the scientists could directly test the effects of this *Pitx1* regulatory region. When they put that *Pitx1* regulatory sequence back into eggs from fish with no pelvis, the eggs developed into fish with pelvic armor.

The lessons of this this research are clear: we can watch evolution in action, at high molecular resolution, by studying variation within species; evolution can hit upon the same solutions over and over, and finally, sometimes regulatory changes are the key to phenotypic change.

Photo by Mike Shapiro

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COMMENTS

Good article.

> *...but even if we can identify which regions of the human genome have been under selective pressure...*

It would seem that this becomes a potentially unanswerable question since we don't necessarily have enough information regarding the environmental selection and sexual selection pressures that any individual would have experienced.

Reply to This » Link **Gerhard Adam** | 01/29/10 | 16:41 PM

You don't need to know what the specific pressures are. You look for genome sequence that has changed less rapidly (negative/purifying selection) or more rapidly (positive selection) than a model of neutral sequence change.

We can't say what the pressures were, or do much more than guess about the effects of these changes, but we can, just by comparing DNA sequence, find regions that show signatures of selection.

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Michael White | 01/29/10 | 17:58 PM

I guess I'm thinking a bit more philosophically about it. Considering something like the question of the human evolution of bigger brains, we can see that changes must've occurred, and we can certainly argue that a change in fitness must have accompanied it so that it became pervasive and fixed in the population. However, we must obviously have something that could've been selected in the first place. So, whether it was a coincidence, a mutation, drift, etc. are probably impossible to pin down. In fact, it seems that it may well simply be a fortuitous combination of normal variations with environmental factors that may create the circumstances to push a species (or us) down a particular path. After which any advantages that a change in the trait conveyed would have to be coupled with environmental circumstances that ensured such an advantage resulted in tangible fitness differences.

In any case, I guess I was thinking more along the lines that regardless of what traits are expressed or present from the parents, in the absence of an environmental factor that can act as a selection "filter", the only thing we can see is the "mechanics" of the process.

Reply to This »   Link

Gerhard Adam | 01/29/10 | 18:15 PM

> *However, we must obviously have something that could've been selected in the first place. So, whether it was a coincidence, a mutation, drift, etc. are probably impossible to pin down.*

This is another justification for studying genetic and phenotypic variation within species - you can gain an understanding of what is out there to be acted on by selection.

> *in the absence of an environmental factor that can act as a selection "filter", the only thing we can see is the "mechanics" of the process.*

You're right - generally, we can only speculate regarding the specific nature of the selection pressure or filter, although there have been instances where people have directly tested the effects of hypothesized environmental pressures.

In the case of the sticklebacks, the situation isn't so complex - we can largely pin the causes of the changes in pelvic girdle morphology on the change in the types of predators in the freshwater environment, and perhaps the lower availability of bone-making calcium in the environment.

Reply to This »   Link

Michael White | 01/29/10 | 20:45 PM

I understand and you're right, that this is a good way to gain more insight into the process, as well as some instances of where we can evaluate the environmental pressures.

I've been stuck lately on the issue of human brain size increase, ultimately leading (presumably) to the evolution of "reason". Obviously everything that is thought of is essentially a story, but I've been trying to think about whether there's any way to come up with a plausible scenario that could explain the branching off point. In thinking about it, it seems impossible to escape the conclusion that it was largely a matter of dumb luck. A particular set of circumstances that are unique enough to where they could occur with some degree of frequency, coupled with the coincidence of a relatively low human population. This probably resulted in a higher degree of inbreeding which would have been prone to accelerate the selection process, thereby amplifying the increase in brain size.

Just a story, but I'm struck by the possibility.

Reply to This » Link **Gerhard Adam** | 01/29/10 | 21:27 PM

> *Obviously everything that is thought of is essentially a story, but I've been trying to think about whether there's any way to come up with a plausible scenario that could explain the branching off point. In thinking about it, it seems impossible to escape the conclusion that it was largely a matter of dumb luck.*

Dumb luck, or, as I'd put it more opaquely, contingency, always plays a role. But don't forget that the primate lineage, and the ape lineage had already experienced big expansions in brain size/intelligence. So apes were already exploiting a niche where bigger brains served them well.

You can also speculate about the rapid increase in brain size in the human lineage in terms of a positive feedback loop. What ever initiated the need for bigger brains beyond what hominoids already had (such as adaptation to a more unpredictable climate during a time of transition), the process of brain size expansion could have been accelerated by the increasing social needs:

Bigger brains enabled social interactions to become a more complex and important element of survival in the environment of our ancestors 3 million years ago, and a more complex social scene kept up the pressure for bigger brains.

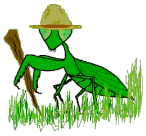
Reply to This » Link **Michael White** | 01/30/10 | 10:44 AM

I just read an interesting article in Scientific American that suggested part of the impetus for developing bigger brains was the changing climate, which caused hominids to traverse larger territories in their quest for food. However, because of the larger ranges, there was a real danger of over-heating, so there was a selection pressure to lose our hair. In conjunction with longer legs which allowed for farther distances being traveled, the loss of hair (and subsequent increase in glands to facilitate sweating) suggests that there was a trend to accommodate other mechanisms for cooling under more physically demanding situations.

There are obviously more elements involved with this, but it seemed like an interesting idea.

Reply to This » Link **Gerhard Adam** | 01/31/10 | 00:14 AM

> *we can largely pin the causes of the changes in pelvic girdle morphology on the change in the types of predators in the freshwater environment, and perhaps the lower availability of bone-making calcium in the environment.*



So, has anyone investigated whether or not Calcium has any regulatory control of the expression of these *Pitx1* genes?

One reason some of this stuff is so complicated for such dull measurers like us is there are so many confounding variables. For instance, if these fish were introduced into a (fresh water) environment that lacked Calcium, and Calcium was a regulatory mechanism for the gene, they wouldn't express it.

The presumption that they have 'stopped making' a bony pelvic structure because there aren't as many predators, so why waste the resources is one possible explanation. But it also could be that in the absence of Calcium in the environment, the Sticklebacks changed their behavior to better avoid whatever predators are there.

I would have supposed something like more readily, since if there is a nice, juicy fish, that is succulent and has hardly any bones, chances are some predator will figure it out. And I know there has been a lot of work on the plastic (read: fickle) courtship and nesting behaviors of these guys. Susan Foster at Clark University worked on evolutionary relationships on characters like that:
http://www.clarku.edu/activelearning/departments/biology/foster/fosterD.cfm

(Okay, I know I could google it myself, but I figured someone may know already!)

Reply to This »   Link   **Mark Berman** | 01/30/10 | 14:07 PM

> *if there is a nice, juicy fish, that is succulent and has hardly any bones, chances are some predator will figure it out.*

And we did, Mark, long since!

Predator #1 - great diving beetle
UK The great diving beetle is one of the most ferocious predators
in various habitats in UK and US. In the UK it is known to kill sticklebacks by sucking out the most fluid parts, leaving skin and bones.

Predator #2 - man.

> *In some areas, sticklebacks occur in great numbers and they have been turned into oil, animal feed and agricultural fertiliser.*

http://www.ypte.org.uk/animal/stickleback-three-spined-/178

Related, may be of interest: Valery V. Ziuganov and Alexey A. Zotin
Pelvic Girdle Polymorphism and Reproductive Barriers in the Ninespine Stickleback Pungitius pungitius (L.) from Northwest Russia
http://www.jstor.org/pss/4535322

Reply to This »   Link   **Patrick Lockerby** | 01/30/10 | 19:40 PM

A very interesting article Mike, but I can't believe you wrote this: **we have to go look and see whether protein-coding or non-protein-coding DNA changes are the favored means of achieving evolutionary change.**
Surely you're not implying purpose to evolution?

Reply to This »   Link   **Steve Davis** | 02/02/10 | 18:37 PM

And this has me worried Mike. **"Evolution (h)as acted on the same gene, again and again, to reduce the pelvic armoring that is important for survival..."** Evolution is an outcome, surely, not an active agent.

Reply to This » Link **Steve Davis** | 02/02/10 | 18:55 PM

Steve, this is a just a colloquial way of talking about the effects on selection, used in order to avoid excessively long and awkward sentences. Even in a professional setting, we often talk about selection 'acting' on something.

Reply to This » Link **Michael White** | 02/03/10 | 10:30 AM

Even so Mike, I think some of the professionals get themselves confused when language is not as precise as it could be.
Precise language is even more important in your case here, where you regularly go into battle against "creation science", and do a great job. By giving the impression, rightly or wrongly, that evolution is an active agent or that natural selection has purpose, you've inadvertantly wandered into the realms of the supernatural and given ammunition to those who claim that science has become a religion.

Reply to This » Link **Steve Davis** | 02/04/10 | 00:35 AM

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