Forget fossils, the key to understanding the explosion of animal life half a billion years ago lies in our genes. *Bob Holmes* climbs down the evolutionary tree

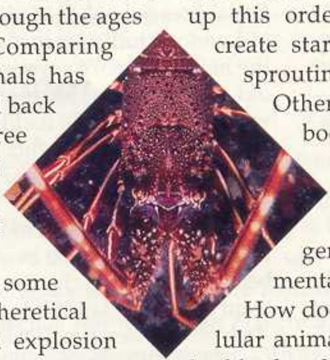
When we were worms

GLASS skyscrapers, Gothic cathedrals, yurts, Georgian terraces, Shinto shrines, wattle and daub, Victorian railway stations, Bauhaus, igloos, mock-Tudor. Imagine that all the architectural styles that human ingenuity could ever devise appeared during one 35-year period, sometime in the middle of the 15th century. Imagine how today's historians would be trampling over each other in their eagerness to learn what made that window of profound creativity possible. That's roughly how palaeontologists feel about the Cambrian explosion.

In just 35 million years, the blinking of an eye for evolution, animal life erupted in an explosion of inventiveness that far outshines anything the planet has seen before or since. During the preceding three billion years of life on Earth, the most sophisticated creatures evolution could come up with were the organismal equivalent of mud huts—algae, the mysterious Ediacarans (see "The Garden of Ediacara", p 32), and perhaps flatworms. Then, with little warning, at least little that left any trace in the fossil record, all hell broke loose. Early in the Cambrian period about 535 million years ago, large, diverse and lavishly elaborate animals suddenly appeared.

They had heads, middles, rear ends, segments and guts. Some had four legs, some a dozen. Some a shell, some antennae, some gills. In short, almost all the body forms familiar in modern animals, and several more that have long since become extinct, sprung up in this one great orgy of creativity. Then it all stopped, as abruptly as it had started. And in over 500 million years since the end of the Cambrian, evolution has rested on its laurels, content to spin off variations on old themes.

The palaeontologists struggling to explain the Cambrian explosion face a tough task—500 million years separate them from their subject. Until recently their only option was to study the animal fossil record. But now, more and more researchers are taking a different path to enlightenment: scrutinising the genetic record that has been handed down through the ages to today's creatures. Comparing the genes of living animals has enabled biologists to crawl back down the evolutionary tree to deduce which genes were present back then and what roles they might have played.



This exploration has led some researchers to make the heretical claim that the Cambrian explosion never happened, and others to say it certainly did, and that they have found the likely mechanism: the genes that help the cells of a developing embryo know front from back, top from bottom, and near from far. They believe that these genes were a necessary prerequisite for the explosion—though they may not have been what set it off.

The most famous of these genes belong to a set known as the *hox* cluster. *Hox* genes are the mapmakers that tell the embryo's cells where they are on the body's front-to-back axis and thus what they should become. In fruit flies, in which they were first discovered, eight *hox* genes line up on their chromosome like a train of boxcars. The gene at the front of the train tells cells there to make a head and other paraphernalia characteristic of that part of the body, number two takes over a bit further back, and so on back to the guard's van (Americans call it the caboose), which holds sway over the hindmost end of the animal. By mucking up this orderly sequence, researchers create startling freaks such as flies sprouting legs from their heads. Other sets of genes lay out the body's up-down axis or distinguish the base of a leg or a wing from its tip.

The discovery of these genes solved one of developmental biology's central puzzles. How does the embryo of a multicellular animal know what structures to build where? And about two years ago, three palaeontologists began to suspect that these genes could be the answer to another great enigma, the Cambrian explosion. The modular way in which the

Illustration: Charlie Ward   Planet Earth Pictures

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'Was giga-great-grandmother a roundish flatworm, little more than an oo

genes map body regions would have provided evolution with a mechanism to modify one part of the body without changing the rest, to duplicate segments, or to add new appendages where none existed before. This is the line taken by James Valentine, of the University of California at Berkeley, David Jablonski of the University of Chicago and Douglas Erwin of the National Museum of Natural History in Washington DC. Just as spoken language (also modular) is flexible enough to support the whole of human culture, says Valentine, this genetic language is so basic, so powerful, and so adaptable that it could underlie the whole amazing diversity of animal life.

Sounds great, but making bold claims is the easy bit—to convince your colleagues, you usually need some evidence. Valentine, Jablonski and Erwin needed to show that these mapmaking genes actually existed in the Cambrian. That posed a problem—*Jurassic Park* notwithstanding, genes don't fossilise, least of all for half a billion years and more.

Fortunately, living organisms hold much of the secret. "The present is the key to the past," says Rudolf Raff, an evolutionary developmental biologist at Indiana University in Bloomington. Raff's reasoning is simple. Living organisms are the tips of branches in the evolutionary tree. If the same gene occurs in two of these animals, that gene must also have been present in their common ancestor, back where their lineages first split apart. By comparing ever more distantly related organisms, biologists can move down toward the earliest branches near the tree's root.

Planet Earth Pictures   The Natural History Museum

Fruit flies and frogs—two veterans of the biology labs—have very different body designs. And they occupy twigs on two of the most fundamental branches of the animal tree, the groups known to biologists as protostomes and deuterostomes respectively (named after the way in which the embryonic mouth forms). Palaeontologists know from the fossil record that these two lineages diverged no later than the early Cambrian, 535 million years ago. They could scarcely be more distant cousins.

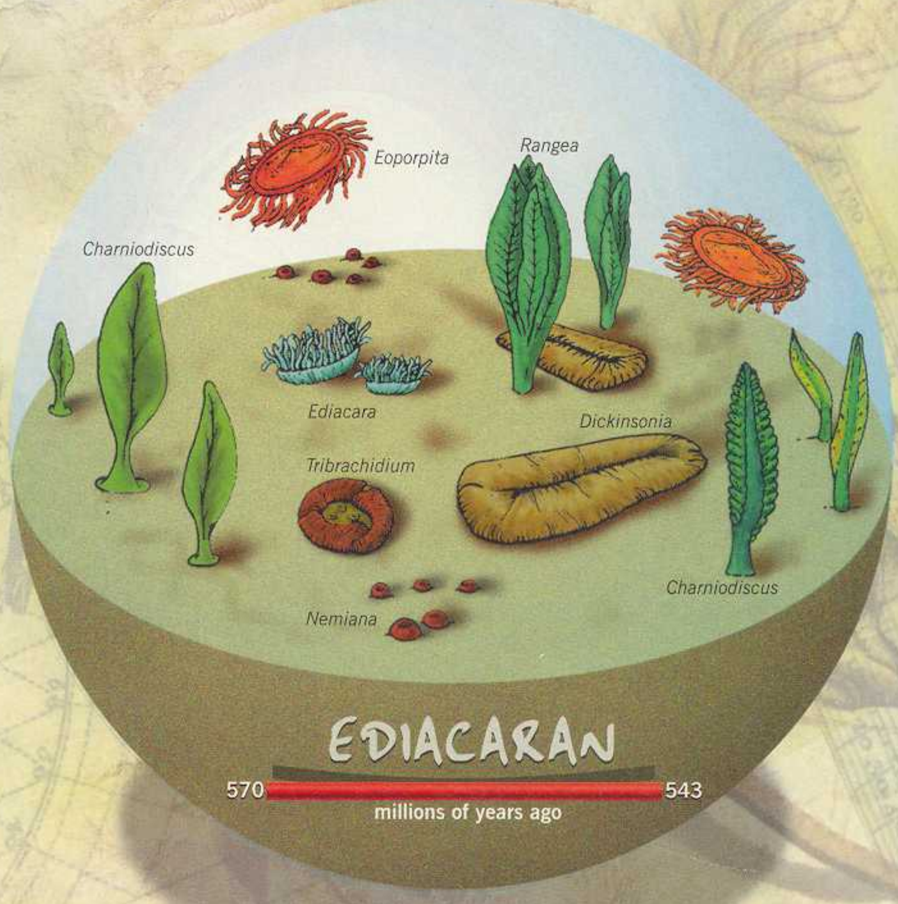
Yet over the past few years, developmental biologists have accumulated more and more evidence of astounding similarities between the DNA sequences in the mapmaking genes of these two groups. Flies and frogs, and by inference their common ancestor in the Cambrian or before, share six *hox* genes. Another gene

THE GARDEN OF EDIACARA

WHEN something goes kaboom in a good action film, there have usually been some mysterious characters hanging about beforehand. So, too, for the Cambrian explosion, that orgy of evolutionary activity that created the menagerie of animal body shapes around today. In this case, the skulkers are the Ediacarans, named after the Ediacara Hills in South Australia, where palaeontologists stumbled on the first big deposit of their fossils in the 1940s. These creatures dominated the Earth for tens of millions of years leading right up to the beginning of the Cambrian, according to some major fossil finds in Namibia over the past two years. That means that the Ediacarans were at the very least eye witnesses to the Cambrian explosion. Or they might have been at the explosion's ground zero—the simple forebears of the elaborate animals that it created.

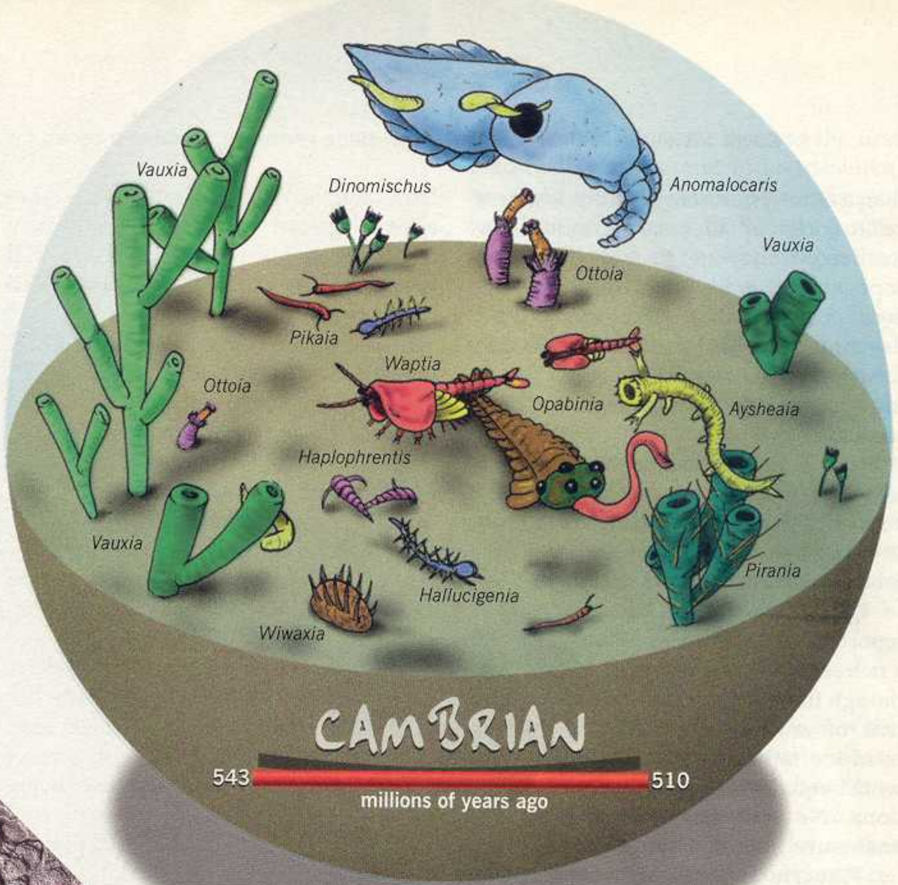
Palaeontologists who are trying to reconstruct the course of events that led up to evolution's big bang disagree wildly over where these strange creatures fit in. "The problem is that the same fossils are interpreted in completely different ways by different people," says Simon Conway Morris of Cambridge University.

To some people the long, graceful plumes, strange ribbed discs and amorphous, quilt-like creatures up to a metre long look like



an oozing blob of cells?'

called *decapentaplegic*, which marks the upper half of the fly embryo, is kin to *Bmp-4*, which marks the lower half of the frog embryo. Somewhere in the course of evolution, the developmental processes triggered by the genes did a half somersault in one of the two lineages. Apart from that, the two have diverged so little in more than half a billion years that scientists can snip the gene out of a fly, plug it into a frog, and it will work perfectly, triggering the development of the bottom half of a frog wherever you insert it in the embryo. You can do the same thing with a gene called *pax6*, which controls eye development. Even genes that dictate the development of such modern-seeming accoutrements as hearts, nervous systems,and body segments appear to have been present in the frog-fruit fly common ancestor, way back in the Cambrian or earlier.



This was just the evidence the three palaeontologists needed—and its implications are still sinking in. For a start, it questions the old assumption that the common ancestor of horses and horseflies, lobsters and Londoners—the giga-great-grandmother of the early Cambrian or before—was a "roundish flatworm", little more than an oozing blob of cells. After all, if the genetic evidence is to be believed, that ancestor could equally well have been a sophisticated, segmented worm with eyes, a heart, a nervous system, possibly even antennae or legs, says Eddy De Robertis, an embryologist at the University of California at Los Angeles.

Sensory bump

Another explanation, and the one that fits in best with Valentine & Co's hypothesis, is that those key developmental genes existed in that roundish flatworm, but didn't trigger the growth of eyes, limbs, hearts, and segments as they do in post-Cambrian animals. "The problem is that we see what these genes do now and we assume they were doing the same thing way back then," explains Nipam Patel, a developmental biologist at the University of Chicago. "But you have to be very careful." The Precambrian "eye" that *pax6* helped form may have been nothing more

nothing else on Earth. One, Ernietta, looks like a round-bottomed mug made out of drinking straws. These people argue that some Ediacarans—perhaps all—deserve their own kingdom, albeit an extinct one, on a par with today's plants, animals and fungi.

But other Ediacaran fossils show affinities to segmented worms and relatives of corals and jellyfish, says James Gehling of the University of South Australia in Adelaide. And one, Kimberella, has many hallmarks of a primitive mollusc, Russian and American researchers announced in August in *Nature* (vol 388, p 868). Evidence like that suggests that at least some Ediacarans are the forerunners of modern animals.

Absolutely not, says Mark McMenamin of Mount Holyoke College in South Hadley, Massachusetts. "I've figured out the Ediacaran problem. I'm convinced now that these things are not animals." His new analysis shows that during their embryonic development, Ediacarans never pass through a stage consisting of a hollow ball of cells known as a blastula, as all animals do. He plans to present his evidence at next week's meeting of the Geological Society of America in Salt Lake City, Utah.

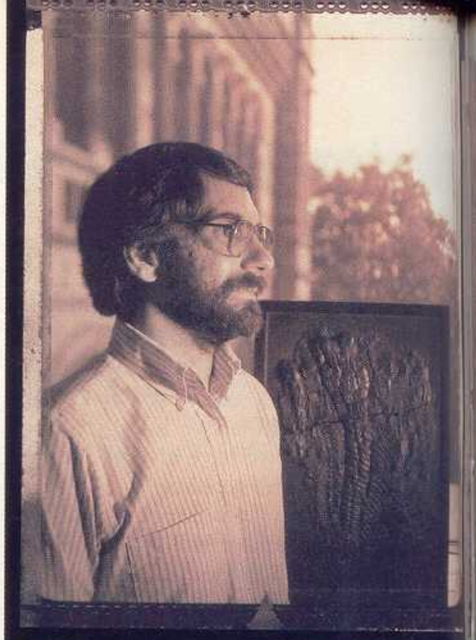
McMenamin's claim is sure to prove contentious, but even those who don't buy it think that he might be right on a second point: his assessment of Ediacarans' fate in the Cambrian. McMenamin imagines the late Precambrian as a "Garden of Ediacara", an idyllic time where large, soft-bodied creatures lolled about on the seabed in a peaceful coexistence, drawing nourishment from photosynthetic algae or microorganisms living within their tissues, much as many corals do today.

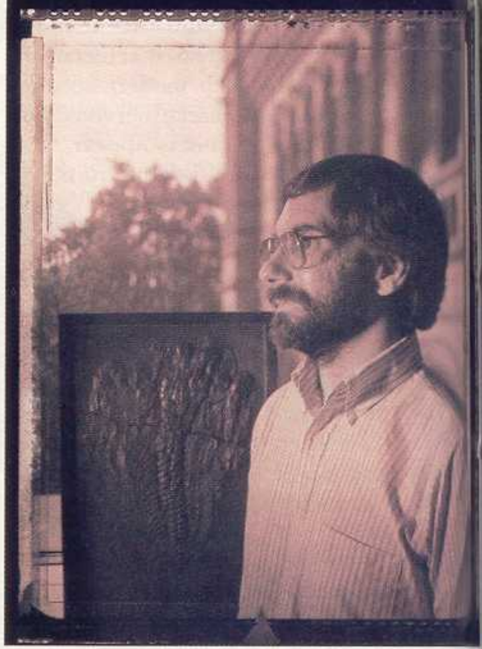
Their idyll came to a sticky end when the animal forebear (be it an Ediacaran or some other organism) evolved into the first large predator. For a creature with the right equipment—sense organs to find prey and mouthparts to bite it with—this peaceable kingdom was a well-stocked supermarket full of tasty treats.

The evolution of such predators around the beginning of the Cambrian would spark an evolutionary arms race in which the organisms that could get faster, fiercer, or more furtive survived to give rise to the animals we see in the Cambrian, says Bruce Runnegar of the University of California at Los Angeles. Those that couldn't adapt—including the biggest, blobbiest Ediacarans—made a quick exit.

This theory, appealing as it is, remains untested so far. But palaeontologists are already scouring rocks at the Cambrian boundary for signs of the sudden appearance of predation. "I'm sure we're going to have many discoveries in the next 12 months," says Gehling.

David Jablonski: without the key mapmaking genes we would still be worms





than a crude photosensor with a pigment cell to back it up, for instance. Indeed, the fossil record shows no trace of anything as sophisticated as the insect and vertebrate eyes in the earliest protostomes and deuterostomes. Likewise, the "appendage" for which the ancestor carried mapmaking genes may have been just a little sensory bump.

There is other evidence that Jablonksi, Valentine and Erwin's hypothesis could be correct. In a paper published in August in the *Biological Bulletin* (vol 193, p 62), John Finnerty and Mark Martindale of the University of Chicago report that sea anemones have a rich set of *hox* genes, even though these most primitive of animals have no head or tail—they face the world equally well in all directions. No one knows why sea anemones should have such genes, but Finnerty suspects they may map out the anemones' far simpler up-and-down axis instead. The best guess is that the Precambrian flatworm then commandeered these crude mapmaking genes for use in the crucial front-to-back axis of its more complex body plan—possibly their first big role as the "language of evolution".

"Creationists have often said that the one thing we can't explain is the extremely rapid appearance of body plans in the Cambrian," says Valentine. "And this is the answer. We haven't understood it until these development guys."

But not all the new discoveries in evolutionary molecular biology fit as well with the whole idea of a Cambrian boom. There have always been rumblings from certain quarters that perhaps the Cambrian explosion is nothing more than an illusion—a lovely, imaginative, but completely wrong reconstruction of evolutionary history. The main evidence for the explosion has been the total absence—despite a lot of diligent searching—of fossils showing modern-style body plans until the start or just before the start of the Cambrian. "If you go to the same kind of rocks and the animals are not there, and you look all over the world for tracks and trails and they're not there, the most parsimonious explanation is that complex large [animals] just were not there. It takes a lot of special pleading to argue they were there and just were not preserved. It would require a truly perverse fossil record," says Jablonski.

In fact, that's just what the anti-Cambrians believe—that somehow, through not knowing where to look, or simply through sheer bad luck, fossil hunters have missed the bodies, burrowings and footprints of complex animals' Precambrian history. And now they have some molecular ammunition to bolster their claim. In a study published one year ago in *Science* (vol 274, p 568), Gregory Wray and his colleagues at the State University of New York at Stony Brook measured the changes in the sequences of seven genes in dozens of living species, and concluded that the protostome-deuterostome split happened roughly a billion years ago, long before the alleged Cambrian explosion. More recent branches on the animal tree, such as the split between echinoderms (starfish and sea urchins) and chordates (the group to which vertebrates belong) also happened well before the beginning of the Cambrian. "If this guy is right," says Finnerty. "there's no explosion."

But the "molecular clock" technique Wray and his colleagues used is controversial. First, they assumed that the longer ago two lineages split, the more different their gene sequences should be. Then, they counted how many mutations had accumulated since several branchings that can be clearly dated from fossil records. For example, bony fishes split from sharks 415 million years ago and crocodiles from birds 235 million years ago. They used this to estimate the mutation rate, and so to calculate the age of unknown splits, such as protostomes from deuterostomes

'In the post Cambrian world, competition is severe. If you are no good

The big fizzle

The problem is that mutations may occur in spurts, throwing off estimates of branch dates, says Erwin. Moreover, all the known branch dates Wray used to calibrate his clock are relatively young, so he had to work backwards to date the branches of interest. Like the wavering tip of a long pole gripped at one end, such extrapolations are erratic. To Erwin and other sceptics, this uncertainty means that the "billion-year-old" branches could turn out to be just 600 million years old or less, compatible with the notion of a Cambrian explosion.

The proponents of molecular clocks admit that they are not very precise, but deny that they are way off mark. What's more, other analyses that attempt to take into account variations in mutation rates also put the key lineage splits much earlier than the Cambrian. Taken together, that evidence convinces at least some that the Cambrian explosion never happened. Instead, evolution burned slow and steady for at least 100 million years to create all the different animal forms. "The

Oxford scientific films The Natural History Museum

Photography: Matt Cooke

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Cambrian is a dull period in evolutionary history. It's not where the interesting things really happened," says Jeffrey Levinton, one of Wray's co-authors.

So what should we believe: the hard copy embedded in rocks around the world, or the high-tech virtual reality of the molecular clocks? Perhaps both, says Simon Conway Morris, a palaeobiologist at Cambridge University. Even if the molecular data tell us that the lineages—the echinoderms and the chordates, the protostomes and the deuterostomes—split early on, that doesn't mean they acquired their spiffy new body plans prior to the Cambrian. "You can have your animals ticking away for as long as you like, but if they're not doing anything morphologically, who cares?" he asks.

Which brings us back to the Cambrian. If the lineages had gone their separate ways long before, and if we know mapmaking genes were present before the split, what happened 535 million years ago to trigger the explosion in body plans? Before the evidence for the very early existence of the mapmaking genes, Valentine and his colleagues had toyed with the idea that the genes themselves might have been the trigger rather than part of the explosive mix. That theory has been laid to rest, but as always there are still plenty of others to choose from. Perhaps the first hard-bodied predator chanced to evolve, sparking off an evolutionary arms race in which everybody had to get larger, better armoured, or cleverer to stay alive? Or the oxygen content of the atmosphere increased, suddenly making larger, more energetic animals feasible? Or a burst of speedy continental drift scattered Earth's nearshore habitats, forcing animals to come up with different solutions to new adaptive challenges? "We have several plausible and a lot of nutty suggestions," says Valentine. Until some fresh evidence pops up—more genes, or perhaps, some major fossil find—the Cambrian trigger is destined to remain mere speculation.

Carefree worms

But whatever happened to kick-start the Precambrian worm into an evolutionary frenzy, it probably found it much easier to spin off new body plans than a creature would today. "You can do a lot to that animal in terms of its basic body arrangement for two reasons," says Raff. "One is that the world is empty ecologically, so there can be a lot of experimentation. Whatever you're making doesn't have to be very good, because there isn't much competition. The second thing is that because the body's relatively simple, changes that would now be horrendous—like changing dorsal and ventral—would have been nothing at all.

"In the post-Cambrian world, competition is severe, so if you're not good at making a living, you're dead meat. And body plans have become more elaborate, so changes that you make have more consequences. You've connected the genetic machinery together in a certain way, and it may be hard to unconnect it. That's left us with a world in which evolution is largely within body plans."

In short, animal life on Earth has grown up. The heady, experimental days of its carefree youth are past and now, saddled with a job, a mortgage, responsibilities, it has settled down into a steady, plodding respectability. Looking back now, we can shed a tear for those days so long ago when life was young and we were worms. □

Further reading: "Developmental evolution of metazoan bodyplans: the fossil evidence" by J.W. Valentine, D. H. Erwin, and D. Jablonski, *Developmental Biology*, vol 173, p 373 (1996)
The Shape of Life—Genes, Development, and the Evolution of the Animal Form by R. A. Raff (University of Chicago Press)