DISCOVER MAGAZINE

The Loom

« The Slow, Slow Road to De-Discovery

The Birth of the New, The Rewiring of the Old

In 1988, Richard Lenski, an evolutionary biologist now at Michigan State University, launched the longest running experiment on natural selection. It started with a single microbe–*E. coli*–which Lenski used to seed twelve genetically identical lines of bacteria. He placed each line in a separate flask, which he provisioned with a scant supply of glucose. The bacteria ate up the sugar in a few hours. The next day, he took a droplet of microbial broth from each flask and let it tumble into a new one, complete with a fresh supply of food. The bacteria boomed again, then starved again, and then were transferred again to a new home. Lenski and his colleagues have repeated this procedure every day for the past 24 years, rearing over 55,000 generations of bacteria.

I first reported on Lenski's experiment 12 years ago, and since then I've revisited it every few years. The bacteria have been evolving in all sorts of interesting ways, and Lenski has been able to reconstruct the history of that evolution in great detail, thanks to a frozen fossil record. Every 500 generations Lenski and his students sock away some bacteria from each flask in a freezer. They can thaw out these ancestors whenever they wish and compare them to their youngest descendants. Biotechnology has improved drastically since 1988, giving Lenski an increasingly powerful evolutionary microscope. When he started out, it could take months to identify just one of the many mutations that arose in each lineage. These days, he and his colleagues can sequence an entire *E. coli* genome for a few hundred dollars and find every single new mutation in its DNA.

Four years ago, I wrote here about one particularly fascinating episode in the evolution going on in Lenski's lab. It started in 2003, when the scientists there noticed something odd in one of the 12 flasks. It had become much more cloudy than the others. In a microbiology lab, that's a sure-fire sign that the bacteria in a flask have experienced a population explosion.

At first the team suspected that some other species of bacteria had slipped into the flask and was breeding quickly. But they found that the flask was packed with *E. coli*—descendants of the original ancestor that Lenski had used to start the entire experiment.

Somehow the bacteria in this one flask had evolved a way to grow much faster than the other bacteria.

The scientists determined that the bacteria had made a drastic switch: from feeding on the glucose to another compound, called citrate. Citrate is an ingredient in the broth where Lenski's *E. coli* grows. It's not food; instead, it helps keep the minerals in the broth in the right balance for *E. coli* to grow.

To a microbiologist, the emergence of *E. coli* that can eat citrate in a lab is deeply weird. *E. coli* typically can't feed on citrate in the presence of oxygen. Some strains of *E. coli* can draw in citrate, but only if there's no oxygen around. To make the reaction work, they have to pump out another compound called succinate at the same time. The ability of E. coli to feed on citrate in the presence of oxygen is extremely rare; it occurs when E. coli picks up the necessary genes from other species. In its normal environment (inside us), natural selection must not favor these mutants. Scientists have been studying *E. coli* in labs for over a century, making it the most intensely studied species, on Earth (as I explain in my book *Microcosm*). But in all that time, there has been only a single report of a citrate-feeding *E. coli* in a lab, back in 1982.

The inability of *E. coli* to grow on citrate is so stark, in fact, that microbiologists use it as a way to tell whether bacteria they come across are *E. coli* or not. It was thus a surprise to Lenski and his students to find a flask of *E. coli* suddenly feeding on citrate. The bacteria had not picked up the genes from another species. Instead, their new ability must have evolved after Lenski started his experiment with a single *E. coli*. This was not simply a case of natural selection enabling a species to do something better. This was a case of doing something new.

Zachary Blount–then a graduate student in Lenski's lab and now a post-doctoral researcher there–led the investigation into this strange new development. Blount and his colleagues took away the glucose and found that the *E. coli* could thrive on citrate alone. They then defrosted the bacteria's ancestors and fed them citrate to figure out when they acquired the ability. They found that a tiny fraction of the bacteria around generation 31,000 were able to grow very slowly on the citrate. Over a couple thousand generations, they got better at growing on citrate–so good, eventually, that they took over the flask and turned it cloudy.

Blount and Lenski first reported the evolution of the citrate-eaters in 2008. Now, after another four years of painstaking research, they're back with a new paper that details what happened down at the molecular level. It's a fascinating look at how new traits evolve by duplicating and recycling old parts.

The scientists found that the evolution took place in three chapters. In the first chapter, the stage was set for the transformation. In the second chapter, the bacteria became citrate feeders. And in the third chapter, they became much better citrate feeders.

It usually makes sense to start a story with Chapter One. But in this case, it's better to start with Chapter Two: The Birth of the Citrate Feeders.

When *E. coli* finds itself in the absence of oxygen, it switches on a gene called *citT*. Like other species (including us), *E. coli* turns genes on and off by attaching proteins to short stretches of DNA nearby. When *E. coli* senses a lack of oxygen, proteins clamp onto one of these genetic switches near *citT*. Once they turn the gene on, it produces proteins that gets delivered to the surface of the cell. There they poke one end out into the environment and pull in citrate, while also pumping out succinate. After the citrate gets inside the microbe, the bacteria can chop it up to harvest its energy.

When *E. coli* is growing in oxygen, however, no proteins land on the genetic switch near the *citT* gene, and so it remains silent. The microbe wastes no energy making a protein that won't help it grow.

Evolution has rewritten this little algorithm in the citrate eaters. As one cell in Lenski's flask divided, it duplicated its DNA with one fateful mistake. It accidentally copied the *citT* twice. The new copy ended up near a different genetic switch–a switch that turns on neighboring genes in the *presence* of oxygen, not the absence.

While *citT* remained silent in the other bacteria in the flask, in the mutant cell, it switched on. The microbe began sticking citrate transporters on its surface and started drawing in the molecule. This mutation must have occurred by generation 31,500, when Blount found the earliest citrate eaters. The mutation was a crude hack; it produced a microbe that could draw in a little citrate, but not a lot. It still had to feed on glucose to get by.

Thus endeth Chapter Two.

In Chapter Three, life got better for the feeble citrate eaters. They copied the *citT* gene, along with its oxygen-switch promoter. Now the bacteria could make even more CitT channels, and thus pull in even more citrate. The bacteria made a third copy, and could pull in even more. Blount and his co-authors proved that the extra copies helped the bacteria this way by defrosting bacteria from Chapter Two and inserting copies of *citT* into them. Those early citrate eaters immediately got much better at feeding.

The scientists also found other mutations that arose during Chapter Three. While they have yet to figure out what those mutations did, the evidence they've gathered so far suggests the mutations allowed the bacteria to break down citrate more efficiently so they could get more energy from their food.

The most intriguing part of the story, however, is the first– Chapter One: Setting the Stage.

When Lenski and Blount first began to study the citrate eaters, they wondered what would happen if they wound back the evolutionary tape and let the bacteria re-evolve. Would the citrate feeding evolve again?

Blount thawed out ancestors from various moments in the history of the bacteria and started putting them through the same evolutionary experiment again. In some trials, the bacteria did indeed evolve into citrate eaters–but only if they came from after generation 20,000. This discovery suggested only after 20,000 generations were the bacteria prepared to evolve into citrate eaters. They must have already acquired other mutations that set the stage.

To test this idea, Blount and his colleagues thawed out some of the "prepared" bacteria: late-generation E. coli that had not yet gained mutations to *citT*. They created a miniature ring of DNA loaded with many copies of CitT and the oxygen-sensitive switch, and inserted it into the prepared bacteria. As they predicted, the bacteria now could suddenly feast magnificently on citrate.

But if Blount and his colleagues inserted the DNA ring into the original ancestor of the line, it grew poorly on citrate. That failure suggested that the early-generation bacteria were not ready to receive this evolutionary gift.

And thus a history takes shape:

Chapter One (from generaton zeo to at least generation 20,000): Our hero, *E. coli*, picks up mutations that don't seem to have anything to do with feeding on citrate. They might have helped the bacteria grow better on their stingy rations of glucose. At least one of those mutations set the stage for feeding on citrate.

Chapter Two (around generation 31,500): The bacteria accidentally rewire their genome, so that a new copy of *citT* switches on in the presence of oxygen. Thanks to the mutations of Chapter One, this rewiring yields a modest but important improvement. Now the bacteria can feed a little on citrate, as well as on glucose.

Chapter Three (from about generation 31,500 to 33,000–and beyond): The bacteria make extra copies of the new and improved *citT*. They can pull in more citrate; new mutations fine-tune their metabolism to grow quickly on the molecule. World domination soon follows.

It's remarkable how this experiment contains many elements of evolution that scientists have noted in other species. It's common for genes to get duplicated, and for the new copy to be rewired for a new job. Snake venom, to pick one example, also evolved when genes were accidentally copied and then rewired. A gene that originally produced a digestive enzyme in the pancreas, for instance, now started making that enzyme in a snake's mouth. It turned out to be a crude but effective venom. Later mutations fine-tuned the new venom gene until it became wickedly good.

The only important difference is that it took millions of years for snakes to evolve their arsenal of venoms, and scientists can only reconstruct their evolution by comparing living species. But in the case of *E. coli*, the transition unfolded fast enough for someone to track it from start to finish–and restart it when necessary.

Reference: Blount et al, "Genomic analysis of a key innovation in an experimental Escherichia coli population." *Nature*, September 19 2012.

September 19th, 2012 1:00 PM by Carl Zimmer in Evolution, Microcosm: The Book, The Tangled Bank, Top posts | 16 comments | RSS feed | Trackback >

16 Responses to "The Birth of the New, The Rewiring of the Old"

1. 1. *Kevin T. Keith* Says:
   September 19th, 2012 at 1:25 pm

   Good example of pre-adaptation at the molecular level. Also good evidence for molecular vestigialism – they abandon the glucose mechanism after gaining the citrate mechanism, but presumably there is DNA residue of that pathway.

   Beautiful piece of science.

2. 2. *Amos Zeeberg (Discover Web Editor)* Says:
   September 19th, 2012 at 1:57 pm

   I'm curious about how the other 11 strains developed—whether they went through some of the same chapters as the one that evolved to eat citrate, or whether that one went off in an entirely different direction in evolution space.

   [CZ: None of the other 11 lines evolved citrate metabolism.]

3. 3. *neuroecology* Says:
   September 19th, 2012 at 2:29 pm

Do they have any idea what the mutations are that ‘set the stage’ for citT? That seems equally interesting (possibly coming in a future paper!).

[CZ: The mutations are catalogued in the supplementary material, but the scientists can't say yet which one (or ones) set the stage, or how.]

4. 4. *Christopher Moore* Says:
   September 19th, 2012 at 3:02 pm

   Dr. Lenski’s research has been extremely valuable for the field of evolutionary biology. Evolutionary and ecological biology need more resources and incentives to conduct long-term research.

5. 5. *Mark* Says:
   September 19th, 2012 at 3:29 pm

   Fascinating. I try to remain sceptical of experiments with amazing results, but no reason for scepticism here (none that I see, anyway). I can’t wait to watch this experiment over the next few decades. I’d really love to see what adaptations come up (in all 12 samples)

6. 6. *Avi Burstein* Says:
   September 19th, 2012 at 6:06 pm

   Wow. Amazing story. Thanks!

7. 7. *Paul* Says:
   September 19th, 2012 at 9:12 pm

   So, what evolved were changes in the number of genes and control of the expression of the genes, but not alterations to the proteins themselves?

   [CZ: All of the above.]

8. 8. *D. Kim* Says:
   September 19th, 2012 at 10:20 pm

   It would be even more interesting to continue to fund this research for an even longer period — dare I say hundreds of years more? — and see what new developments occur.

   Could a new species present itself? Perhaps even the rise of a multicellular form? Now THAT would be a giant leap for evolutionary biology.

9. 9. *Scott* Says:
September 20th, 2012 at 12:16 am

Cool experiment! So much hard work must have gone into it.

I have a couple questions:
1) Would the generation 1 E coli outcompete the citrate E coli in a glucose rich environment?

2)In some ways, shouldn't we expect more? I mean, after 55, 000 generations of a huge number of E coli in a very controlled environment perfectly suited for evolution and really all we get is a change in promoters and some duplications of that. Nothing really new is added; all the pieces of citrate metabolism were there beforehand.

Maybe you should try putting E coli in an environment that they truly are not able to utilize at and see if they will eventually evolve to create a completely novel digestion system?

10. 10. *sparc* Says:
September 20th, 2012 at 1:07 am

Creationsts will not accept this research. They will state that adding citrate to the media was an intervention in the frst place that influenced the outcome of the experiments. "Methinks it is like a weasel", Dawkins, latching, in 3 … 2 … 1 …

11. 11. *Ramesh* Says:
September 20th, 2012 at 1:08 am

If such a wonderful evolution occurred and was so rigorously scrutinised,the genesis of life itself starts looking lesser surprising!

12. 12. *Paul Craze (TREE editor)* Says:
September 20th, 2012 at 5:46 am

A really nice piece on one of the iconic studies in experimental evolution. I totally agree with Christopher Moore – there has to be security of funding for long-term studies. The most exciting thing here is Chapter One (because we don't understand it and it could only be studied with an experiment like this), but you could imagine someone with a business-minded attitude to science saying "20,000 generations and no results. That's not cost-effective so we'll stop the funding". Every long-term study I know of has produced important results, almost as if running a study for a long time is a sure way to make the findings important. Probably a low risk strategy in funding terms!

BTW, at the risk of being called self-promoting, this very result features on the cover of October's Trends in Ecology and Evolution along with a major review on experimental evolution for anyone who wants to read more about the potential of this approach.

13. 13. *Joe G* Says:
September 20th, 2012 at 7:16 am

What is all the hype about? Even YEC's baraminology is OK with the type of "evolution" Lenski is observing and studying.

Also there is no way you can determine that the bacteria "accidentally" rewired" their genome.

14. 14. *John Kwok* Says:
September 20th, 2012 at 7:55 am

Sorry Joe G, I think you missed the importance of Lenski's work, especially with the likelihood that he and his team witnessed a "speciation event" in the strain that began metabolizing citrate. I agree with Paul Craze that this ongoing experiment by the Lenski team should be viewed as one of the "iconic studies of experimental evolution".

15. 15. *Joe G* Says:
September 20th, 2012 at 8:41 am

John Kwok,

Baraminology is OK with speciation. The problem is you and your ilk still believe darwin's strawman that your opposition preaches the fixity of species.

16. 16. *Joe G* Says:
September 20th, 2012 at 8:44 am

sparc sez:
Creationsts will not accept this research.

Why is that? That research does absolutely nothing to undermine anything Creationists have said.

Leave a Reply

Name (required)

Mail (will not be published) (required)

Website

Submit Comment

-

- DISCOVER blogs

  80beats
  Discoblog
  Bad Astronomy
  Gene Expression
  Cosmic Variance
  The Loom
  The Crux
  Not Exactly Rocket Science

- # About The Loom

  "Celebrated curiosity monger"

  --Brain Pickings

  Carl Zimmer writes about science regularly for the New York Times and magazines such as Discover, where he is a contributing editor and columnist.

He is the author of twelve books, the most recent of which is *Science Ink: Tattoos of the Science Obsessed*. His website is carlzimmer.com and his address is blog at carlzimmer dot com .

- ## Google Profile

Google+ Statistics My profile→

Carl Zimmer
Followers: 913,235
Following: 125

Statistics by SocialStatistics.com

- ## Facebook

Subscribe 6.2k

- Search

- ## Recent Posts

  - The Birth of the New, The Rewiring of the Old
  - The Slow, Slow Road to De-Discovery
  - Another Science Hurricane: A Massive Survey of the Genome
  - Why Debunking ESP Can Be Interesting
  - Hello, Norway! Two talks in Oslo next week

- ## Science Tattoo Emporium

I once wondered aloud if scientists had tattoos of their science. The answer was yes, and this ever-growing collection is the evidence. I've turned them into a book about art and science called *Science Ink: Tattoos of Science Obsessed*.

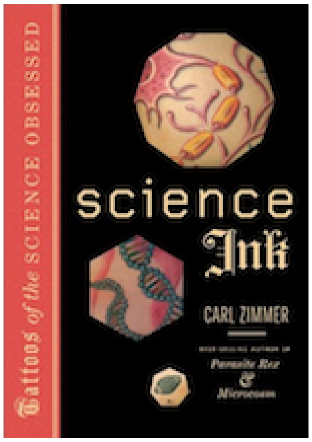
- # Loom Junior

My Tumblr home for scattershot

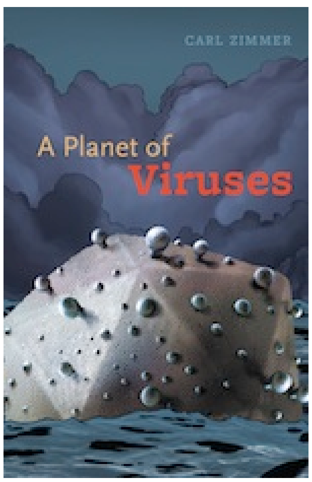
- # Books

Carl Zimmer is the author of twelve books and counting.

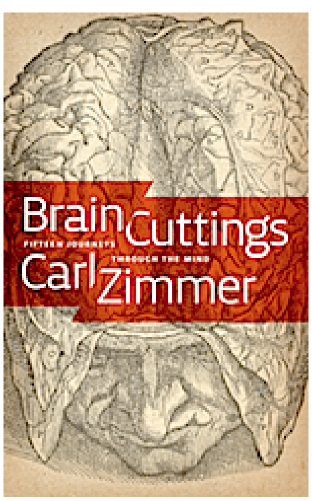
Evolution: Making Sense of Life
"Stunning"-*John Thompson, UC Santa Cruz*
Order a copy



"Beautiful. Packed with fascinating stories"-*Nature*
[Order a copy]

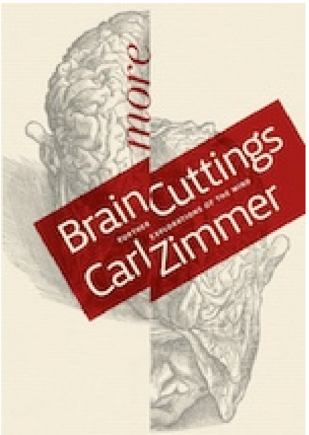


"Fascinating and enlightening"--The Guardian
Available in paperback, hardback or Kindle

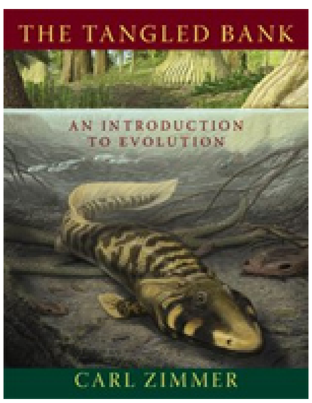


"Carl Zimmer takes us behind the scenes in our own heads. He has ferreted out all the most wondrous, bizarre stories and studies and served them up in

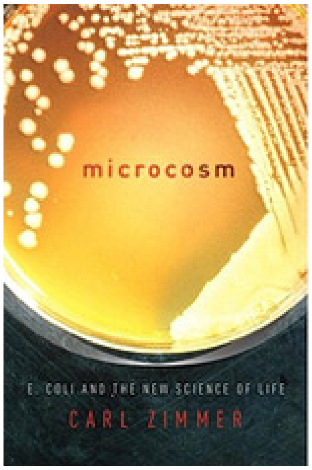
this delicious, sizzling, easy-to-digest platter of neuro-goodness." —Mary Roach, author of Packing for Mars and Stiff
An ebook exclusive: [Amazon](), [Barnes and Noble](), [carlzimmer.com]()



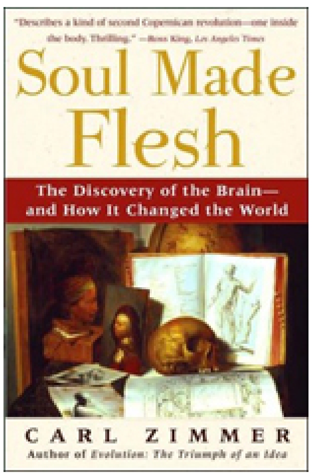
New! More Brain Cuttings:
Further Explorations of the Mind
Order from Amazon and Barnes & Noble and Apple



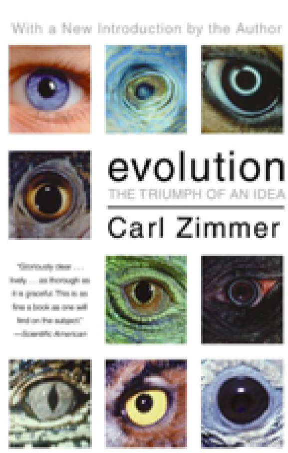
"The Tangled Bank is the best written and best illustrated introduction to evolution of the Darwin centennial decade, and also the most conversant with ongoing research."--Edward O. Wilson, Harvard University
Order a copy



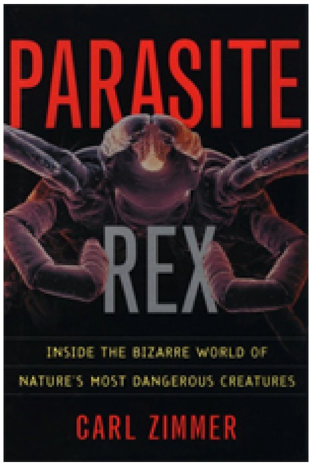
"Superb...quietly revolutionary"--Boston Globe
[Order a copy]()



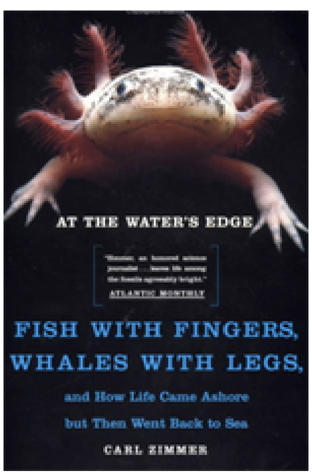
"Fascinating...thrilling... Zimmer has produced a top-notch work of popular science."--Los Angeles Times
[Order a copy]()



"As thorough as it is graceful...This is as fine a book as one will find on the subject."--Scientific American
[Order a copy]



"A book capable of changing how we see the world."--The Los Angeles Times
Reissued with a new epilogue by the author.
[Order a copy]



"A fascinating story, which Zimmer unfolds as a tale of high-stakes scientific sleuthing."--Booklist
[Order a copy]

- # Twitter Updates

  - In case you missed them, 2 new Loom posts: chronic fatigue syndrome http://t.co/HRVysvJF and real-time evolution http://t.co/ui0D5QRb 13 minutes

ago
  - Two more windows for registering for #scio13 Next one: today at 2 pm ET. Details here: http://t.co/1e7ZYS02 25 minutes ago
  - Female guppies that mate with lots of males end up with lots more grandchildren. http://t.co/Hp1mwovi 54 minutes ago
  - This week the Arctic sea ice reached a record-setting low for the year. In this picture, the yellow line mark… http://t.co/N6Ry0WaL about 12 hours ago
  - @PMOrwin @phylogenomics @edyong209 No problem. Ed & I thumb-wrestle for these sorts of stories. about 12 hours ago

follow me on Twitter

- # Comment Policy

Light but firm. Details here.

- # Recent comments

  - Joe G on The Birth of the New, The Rewiring of the Old
  - Joe G on The Birth of the New, The Rewiring of the Old
  - John Kwok on The Birth of the New, The Rewiring of the Old

- # Categories

Select Category

- # Blogroll

  - A Blog Around the Clock
  - Aetiology
  - Babel's Dawn
  - Bad Science
  - Creature Cast
  - Culture Dish (Rebecca Skloot)
  - Dan Ariely
  - David Dobbs
  - dechronization
  - Developing Intelligence
  - Evolution & Medicine Review
  - Gene Expression

- Genome Boy
- Genomicron (Ryan Gregory)
- io9
- john hawks
- John Rennie
- Jonah Lehrer
- Knight Science Journalism Tracker
- Laelaps (Brian Switek)
- Language Log
- Mind Hacks
- Mind Matters (David Berreby)
- Mixing Memory
- Mystery Rays From Outer Space
- Nobel Intent
- Not Exactly Rocket Science
- Oscillator
- Pharyngula
- Prerogative of Harlots
- RealClimate
- Robert Krulwich
- Sandwalk
- Science Cheerleader
- Science Made Cool
- Skeptical Science
- Small Things Considered
- Speakeasy Science (Deborah Blum)
- Steve Silberman
- Steven Johnson's blog
- Superbug
- synthesis
- Tetrapod Zoology
- The Intersection
- The Inverse Square Blog
- The Last Word On Nothing
- The Panda's Thumb
- The Tree of Life
- This Week in Evolution
- Why Evolution Is True
- Word Routes (Ben Zimmer)
- Zooillogix

- **My stuff**
  - CarlZimmer.com
  - Facebook
  - microcosm: E. coli and the New Science of Life
  - My article archive

- **Archives**

Select Month

- **Nifty Fifty**

- **Why “The Loom”?**

"...among the joyous, heartless, ever-juvenile eternities, Pip saw the multitudinous, God-omnipresent, coral insects, that out of the firmament of waters, heaved the colossal orbs. He saw God's foot upon the treadle of the loom, and spoke it; and therefore his shipmates called him mad." --Moby Dick