Reading questions
As you are reading, answer the following either through annotation or on a separate sheet of paper. Some questions will require a written response.

1. How does the size of the human genome compare to that of 'less complex' organisms, such as onions?
2. Describe the method Gregory uses to compare the size of the human genome with that of an onion.
3. How many basepairs does the human genome have? How many genes? What percentage of the human genome codes for proteins directly?
4. What is meant by the term 'junk' DNA?
5. Why does Collins reject the use of the term 'junk DNA'? Why does Gregory disagree with that rejection?
6. How could evolution by natural selection have led to a 'messy' genome?
7. Describe the evidence collected by Vogel that supports the idea that much of the human genome is 'junk.' How does mutation rate support this claim?
8. How can DNA become 'noncoding'?
9. Explain what is meant by the 'Panglossian paradigm' in terms of noncoding DNA and evolution.
10. How are transposable elements 'playing Darwin's game but at the wrong level,' according to Gould?
11. Why is hotair's function in terms of embryo development? How does hotair relate to polycomb?
12. Based on Rinn's experiments, is hotair 'junk' DNA?
13. How is the genome like 'origami'?
14. How does the presence of noncoding DNA support Darwin's theory of evolution?

Is Most of Our DNA Garbage?
New York Times Magazine, March 5, 2015, Carl Zimmer
Carl Zimmer writes the Matter column for The New York Times. He is the author of 13 books, including "Parasite Rex" and "Evolution: The Triumph of an Idea."

T. Ryan Gregory's lab at the University of Guelph in Ontario is a sort of genomic menagerie, stocked with creatures, living and dead, waiting to have their DNA laid bare. Scorpions lurk in their terrariums. Tarantulas doze under bowls. Flash-frozen spiders and crustaceans — collected by Gregory, an evolutionary biologist, and his students on expeditions to the Arctic — lie piled in beige metal tanks of liquid nitrogen. A bank of standing freezers holds samples of mollusks, moths and beetles. The cabinets are crammed with slides splashed with the fuchsia-stained genomes of fruit bats, Siamese fighting fish and ostriches.

Gregory's investigations into all these genomes has taught him a big lesson about life: At its most fundamental level, it's a mess. His favorite way to demonstrate this is through what he calls the "onion test," which involves comparing the size of an onion's genome to that of a human. To run the test, Gregory's graduate student Nick Jeffery brought a young onion plant to the lab from the university greenhouse. He handed me a single-edged safety razor, and then the two of us chopped up onion stems in petri dishes. An emerald ooze, weirdly

luminous, filled my dish. I was so distracted by the color that I slashed my ring finger with the razor blade, but that saved me the trouble of poking myself with a syringe — I was to supply the human genome. Jeffery raised a vial, and I wiped my bleeding finger across its rim. We poured the onion juice into the vial as well and watched as the green and red combined to produce a fluid with both the tint and viscosity of maple syrup.

After adding a fluorescent dye that attaches to DNA, Jeffrey loaded the vial into a boxy device called a flow cytometer, which sprayed the onion juice and blood through a laser beam. Each time a cell was hit, its DNA gave off a bluish glow; bigger genomes glowed more brightly. On a monitor, we watched the data accumulate on a graph. The cells produced two distinct glows, one dim, one bright, which registered on the graph as a pair of peaks.

One peak represented my genome – the entirety of my DNA. Genomes are like biological books, written in genetic letters known as bases; the human genome contains about 3.2 billion bases. Print them out as letters on a page, and they would fill a book a thousand times longer than "War and Peace." Gregory leaned toward the screen. At 39, with a chestnut-colored goatee and an intense gaze, he somewhat resembles a pre-Heisenberg Walter White. He pointed out the onion's peak. It showed that the onion's genome was five times bigger than mine. "The onion wins," Gregory said. The onion always does.

But why? Why does an onion carry around so much more genetic material than a human? Or why, for that matter, do the broad-footed salamander (65.5 billion bases), the African lungfish (132 billion) and the Paris japonica flower (149 billion)? These organisms don't appear to be more complex than we are, so Gregory rejects the idea that they're accomplishing more with all their extra DNA. Instead, he champions an idea first developed in the 1970s but still startling today: that the size of an animal's or plant's genome has essentially no relationship to its complexity, because a vast majority of its DNA is — to put it bluntly — junk.

The human genome contains around 20,000 genes, that is, the stretches of DNA that encode proteins. But these genes account for only about 1.2 percent of the total genome. The other 98.8 percent is known as noncoding DNA. Gregory believes that while some noncoding DNA is essential, most probably does nothing for us at all, and until recently, most biologists agreed with him. Surveying the genome with the best tools at their disposal, they believed that only a small portion of noncoding DNA showed any evidence of having any function.

But in the past few years, the tide has shifted within the field. Recent studies have revealed a wealth of new pieces of noncoding DNA that do seem to be as important to our survival as our more familiar genes. Many of them may encode molecules that help guide our development from a fertilized egg to a healthy adult, for example. If these pieces of noncoding DNA become damaged, we may suffer devastating consequences like brain damage or cancer, depending on what pieces are affected. Large-scale surveys of the genome have led a number of researchers to expect that the human genome will turn out to be even more full of activity than previously thought.

In January, Francis Collins, the director of the National Institutes of Health, made a comment that revealed just how far the consensus has moved. At a health care conference in San Francisco, an audience member asked him about junk DNA. “We don’t use that term anymore,” Collins replied. “It was pretty much a case of hubris to imagine that we could dispense with any part of the genome — as if we knew enough to say it wasn’t functional.” Most of the DNA that scientists once thought was just taking up space in the genome, Collins said, “turns out to be doing stuff.”

For Gregory and a group of like-minded biologists, this idea is not just preposterous but also perilous, something that could yield bad science. The turn against the notion of junk DNA, they argue, is based on over-interpretations of wispy evidence and a willful ignorance of years of solid research on the genome. They’ve challenged their opponents face to face at scientific meetings. They’ve written detailed critiques in biology journals. They’ve commented on social media. When the N.I.H.’s official Twitter account relayed Collins’s claim about not using the term “junk DNA” anymore, Michael Eisen, a professor at the University of California, Berkeley, tweeted back with a profanity.

The junk DNA wars are being waged at the frontiers of biology, but they’re really just the latest skirmish in an intellectual struggle that has played out over the past 200 years. Before Charles Darwin articulated his theory of evolution, most naturalists saw phenomena in nature, from an orchid’s petal to the hook of a vulture’s beak, as things literally designed by God. After Darwin, they began to see them as designs produced, instead, by natural selection. But some of our greatest biologists pushed back against the idea that everything we discover in an organism had to be an exquisite adaptation. To these biologists, a fully efficient genome would be inconsistent with the arbitrariness of our genesis, with the fact that every species emerged through pure happenstance, over eons of false starts. Where some look at all those billions of bases and see a finely tuned machine, others, like Gregory, see a disorganized, glorious mess.

In 1953, Francis Crick and James Watson published a short paper in the journal Nature setting out the double-helix structure of DNA. That brief note sent biologists into a frenzy of discovery, leading eventually to multiple Nobel Prizes and to an unprecedented depth of understanding about how living things grow and reproduce. To make a protein from DNA, they learned, a cell makes a single-stranded copy of the relevant gene, using a molecule called RNA. It then builds a corresponding protein using the RNA as a guide. This research led scientists to assume that the genome was mostly made up of protein-coding DNA.

But eventually scientists found this assumption hard to square with reality. In 1964, the German biologist Friedrich Vogel did a rough calculation of how many genes a typical human must carry. Scientists had already discovered how big the human genome was by staining the DNA in cells, looking at the cells through microscopes and measuring its size. If the human genome was made of nothing but genes, Vogel found, it would need to have an awful lot of them — 6.7 million genes by his estimate, a number that, when he published it in Nature, he admitted was “disturbingly high.” There was no evidence that our cells made 6.7 million proteins or anything close to that figure.

Vogel speculated that a lot of the genome was made up of essential noncoding DNA — possibly operating as something like switches, for example, to turn genes on and off. But other scientists recognized that even this idea couldn't make sense mathematically. On average, each baby is born with roughly 100 new mutations. If every piece of the genome were essential, then many of those mutations would lead to significant birth defects, with the defects only multiplying over the course of generations; in less than a century, the species would become extinct. Faced with this paradox, Crick and other scientists developed a new vision of the genome during the 1970s. Instead of being overwhelmingly packed with coding DNA, the genome was made up mostly of noncoding DNA. And, what's more, most of that noncoding DNA was junk — that is, pieces of DNA that do nothing for us. These biologists argued that some pieces of junk started out as genes, but were later disabled by mutations. Other pieces, called transposable elements, were like parasites, simply making new copies of themselves that were usually inserted harmlessly back in the genome.

Junk DNA's recognition was part of a bigger trend in biology at the time. A number of scientists were questioning the assumption that biological systems are invariably "well designed" by evolution. In a 1979 paper in The Proceedings of the Royal Society of London, Stephen Jay Gould and Richard Lewontin, both of Harvard, groused that too many scientists indulged in breezy storytelling to explain every trait, from antlers to jealousy, as an adaptation honed by natural selection for some essential function. Gould and Lewontin refer to this habit as the Panglossian paradigm, a reference to Voltaire's "Candide," in which the foolish Professor Pangloss keeps insisting, in the face of death and disaster, that we live in "the best of all possible worlds." Gould and Lewontin did not deny that natural selection was a powerful force, but they stressed that it was not the only explanation for why species are the way they are. Male nipples are not adaptations, for example; they're just along for the ride.

Gould and Lewontin called instead for a broader vision of evolution, with room for other forces, for flukes and historical contingencies, for processes unfolding at different levels of life — what Gould often called "pluralism." At the time, geneticists were getting their first glimpses of the molecular secrets of the human genome, and Gould and Lewontin saw more evidence for pluralism and against the Panglosses. Any two people may have millions of differences in their genomes. Most of those differences aren't a result of natural selection's guiding force; they just arise through random mutations, without any effect for good or ill.

When Crick and others began to argue for junk DNA, they were guided by a similar vision of nature as slipshod. Just as male nipples are a useless vestige of evolution, so, in their theory, is a majority of our genome. Far from the height of machine-like perfection, the genome is largely a palimpsest (collection of repeatedly erased and rewritten instructions) of worthless instructions, a den of harmless parasites. Crick and his colleagues argued that transposable elements were common in our genome not because they did something essential for us, but because they could exploit us for their own replication. Gould delighted at this good intellectual company, arguing that transposable elements behaved like miniature organisms, evolving to become better at adding new copies to their host

genomes. Our genomes were their ocean, their savanna. “They are merely playing Darwin’s game, but at the ‘wrong level,’ ” Gould wrote in 1981.

Soon after Gould wrote those words, scientists set out to decipher the precise sequence of the entire human genome. It wasn’t until 2001, shortly before Gould’s death, that they published their first draft. They identified thousands of segments that had the hallmarks of dead genes. They found transposable elements by the millions. The Human Genome Project team declared that our DNA consisted of isolated oases of protein-coding genes surrounded by “vast expanses of unpopulated desert where only noncoding ‘junk’ DNA can be found.” Junk DNA had started out as a theoretical argument, but now the messiness of our evolution was laid bare for all to see.

If you want to see the genome in a fundamentally different way, the best place to go is the third floor of Harvard’s Department of Stem Cell and Regenerative Biology, in a maze of cluttered benches, sequencing machines and microscopes. This is the lab of John Rinn, a 38-year-old former competitive snowboarder who likes to ponder biological questions on top of a skateboard, which he rides from one wall of his office to the other and back. Rinn is overseeing more than a dozen research projects looking for pieces of noncoding DNA that might once have been classified as junk but actually are essential for life.

Rinn studies RNA, but not the RNA that our cells use as a template for making proteins. Scientists have long known that the human genome contains some genes for other types of RNA: strands of bases that carry out other jobs in the cell, like helping to weld together the building blocks of proteins. In the early 2000s, Rinn and other scientists discovered that human cells were reading thousands of segments of their DNA, not just the coding parts, and producing RNA molecules in the process. They wondered whether these RNA molecules could be serving some vital function.

As a postdoctoral fellow at Stanford University, Rinn decided he would try to show that one of these new RNA molecules had some important role. After a couple years of searching, he and a professor there, Howard Chang, settled on an RNA molecule that, somewhat bizarrely, was produced widely by skin cells below the waist but not above. Rinn and Chang were well aware that this pattern might be meaningless, but they set out to investigate it nevertheless. They had to give their enigmatic molecule a name, so they picked one that was a joke at their own expense: hotair. (“If it ends up being hot air, at least we tried,” Rinn said.)

Rinn ran a series of experiments on skin cells to figure out what, if anything, hotair was doing. He carefully pulled hotair molecules out of the cells and examined them to see if they had attached to any other molecules. They had, in fact: they were stuck to a protein called Polycomb.

Polycomb belongs to a group of proteins that are essential to the development of animals from a fertilized egg. They turn genes on and off in different patterns, so that a uniform clump of cells can give rise to bone, muscle and brain. Polycomb latches onto a number of genes and muzzles them, preventing them from making proteins. Rinn’s research revealed

that hotair acts as a kind of guide for Polycomb, attaching to it and escorting it through the jungle of the cell to the precise spots on our DNA where it needs to silence genes.

When Rinn announced this result in 2007, other geneticists were stunned. Cell, the journal that released it, hailed it as a breakthrough, calling Rinn's paper one of the most important they had ever published. In the years since, Chang and other researchers have continued to examine hotair, using even more sophisticated tools. They bred engineered mice that lack the hotair gene, for example, and found that the mice developed a constellation of deformities, like stunted wrists and jumbled vertebrae. It appears very likely that hotair performs important jobs throughout the body, not just in the skin but in the skeleton and in other tissues too.

In 2008, having been lured to Harvard, Rinn set up his new lab entirely in hopes of finding more hotair-like molecules. The first day I visited, a research associate named Diana Sanchez was dissecting mouse embryos the size of pinto beans. In a bowl of ice next to her were tubes for the parts she delicately removed — liver, leg, kidney, lung — that would be searched for cells making RNA molecules. After Rinn and I left Sanchez to her dissections, we ran into Martin Sauvageau, a blue-eyed Quebecer carrying a case of slides, each affixed with a slice of a mouse's brain, with stains revealing cells making different RNA molecules. I tagged along with Sauvageau as he headed to a darkened microscope room to look at the slides with a pink-haired grad student named Abbie Groff. On one slide, a mouse's brain looked as if it wore a cerulean (bright blue) mustache. To Groff, every pattern comes as a surprise. She once discovered an RNA molecule that created thousands of tiny rings on a mouse's body, each encircling a hair follicle. "You come in in the morning, and it's like Christmas," she said.

In December 2013, Rinn and his colleagues published the first results of their search: three potential new genes for RNA that appear to be essential for a mouse's survival. To investigate each potential gene, the scientists removed one of the two copies in mice. When the mice mated, some of their embryos ended up with two copies of the gene, some with one and some with none. If these mice lacked any of these three pieces of DNA, they died in utero or shortly after birth. "You take away a piece of junk DNA, and the mouse dies," Rinn said. "If you can come up with a criticism of that, go ahead. But I'm pretty satisfied. I've found a new piece of the genome that's required for life."

As the scientists find new RNA molecules that look to be important, they are picking out a few to examine in close molecular detail. "I'm totally in love with this one," Rinn said, standing at a whiteboard wall and drawing a looping line to illustrate yet another RNA molecule, one that he calls "firre." The experiments that Rinn's team has run on firre suggest that it performs a spectacular lasso act, grabbing onto three different chromosomes at once and drawing them together. Rinn suspects that there are thousands of RNA molecules encoded in our genomes that perform similar feats: bending DNA, unspooling it, bringing it in contact with certain proteins and otherwise endowing it with a versatility it would lack on its own.

“It’s genomic origami,” Rinn said about this theory. “In every cell, you have the same piece of paper. Stem cell, brain cell, liver cell, it’s all made from the same piece of paper. How you fold that paper determines if you get a paper airplane or a duck. It’s the shape that you fold it into that matters. This has to be the 3-D code of biology.”

To some biologists, discoveries like Rinn’s hint at a hidden treasure house in our genome. Because a few of these RNA molecules have turned out to be so crucial, they think, the rest of the noncoding genome must be crammed with riches. But to Gregory and others, that is a blinkered optimism worthy of Dr. Pangloss. They, by contrast, are deeply pessimistic about where this research will lead. Most of the RNA molecules that our cells make will probably not turn out to perform the sort of essential functions that hotair and firre do. Instead, they are nothing more than what happens when RNA-making proteins bump into junk DNA from time to time.

“You say, ‘I found it — America!’ ” says Alex Palazzo, a biochemist at the University of Toronto who co-wrote a spirited defense of junk DNA with Gregory last year in the journal PLOS Genetics. “But probably what you found is a little bit of noise.”

Palazzo and his colleagues also roll their eyes at the triumphant declarations being made about recent large-scale surveys of the human genome. One news release from an N.I.H. project declared, “Much of what has been called ‘junk DNA’ in the human genome is actually a massive control panel with millions of switches regulating the activity of our genes.” Researchers like Gregory consider this sort of rhetoric to be leaping far beyond the actual evidence. Gregory likens the search for useful pieces of noncoding DNA to using a metal detector to find gold buried at the beach. “The idea of combing the beach is a great idea,” he says. But you have to make sure your metal detector doesn’t go off when it responds to any metal. “You’re going to find bottle caps and nails,” Gregory says.

He expects that as we examine the genome more closely, we’ll find many bottle caps and nails. It’s a prediction based, he and others argue, on the deep evolutionary history of our genome. Over millions of years, essential genes haven’t changed very much, while junk DNA has picked up many harmless mutations. Scientists at the University of Oxford have measured evolutionary change over the past 100 million years at every spot in the human genome. “I can today say, hand on my heart, that 8 percent, plus or minus 1 percent, is what I would consider functional,” Chris Ponting, an author of the study, says. And the other 92 percent? “It doesn’t seem to matter that much,” he says.

It’s no coincidence, researchers like Gregory argue, that bona fide creationists have used recent changes in the thinking about junk DNA to try to turn back the clock to the days before Darwin. (The recent studies on noncoding DNA “clearly demonstrate we are ‘fearfully and wonderfully made’ by our Creator God,” declared the Institute for Creation Research.) In a sense, this debate stretches back to Darwin himself, whose 1859 book, “On the Origin of Species,” set the course for our understanding natural selection as a natural “designer.” Later in his life, Darwin took pains to stress that there was more to evolution than natural selection. He was frustrated to see how many of his readers thought he was arguing that natural selection was the only force behind life’s diversity. “Great is the power

of steady misrepresentation," Darwin grumbled when he updated the book for its sixth edition in 1872. In fact, he wrote, he was quite open-minded about other forces that might drive evolution, like "variations that seem to us in our ignorance to arise spontaneously."

Darwin was certainly ignorant about genomes, as scientists would continue to be for decades after his death. But Gregory argues that genomes embody the very mix of adaptation and arbitrariness that Darwin had in mind. Over millions of years, the human genome has spontaneously gotten bigger, swelling with useless copies of genes and new transposable elements. Our ancestors tolerated all that extra baggage because it wasn't actually all that heavy. It didn't make them inordinately sick. Copying all that extra DNA didn't require them to draw off energy required for other tasks. They couldn't add an infinite amount of junk to the genome, but they could accept an awful lot. To subtract junk, meanwhile, would require swarms of proteins to chop out every single dead gene or transposable element — without chopping out an essential gene. A genome evolving away its junk would lose the race to sloppier genomes, which left more resources for fighting diseases or having children.

The blood-drenched slides that pack Gregory's lab with their giant genomes only make sense, he argues, if we give up thinking about life as always evolving to perfection. To him, junk DNA isn't a sign of evolution's failure. It is, instead, evidence of its slow and slovenly (messy) triumph.