Thumbs up -- a tiny ancestral remnant lends developmental edge to humans

September 4 2008

Subtle genetic changes that confer an evolutionary advantage upon a species, such as the dexterity characteristic of the human hand, while difficult to detect and even harder to reproduce in a model system, have nevertheless generated keen interest amongst evolutionary biologists. In findings published online in the September 5 edition of the journal *Science*, researchers from the U.S. Department of Energy's Lawrence Berkeley National Laboratory and their collaborators, have uncovered a specifically human 13-nucelotide change concealed in the vast three-billion-letter landscape of the human genome.

Their experiments reveal this stretch of DNA to be a recently evolved regulator of gee expression that, when introduced into a mouse embryo system, influence the molecular machinery to yield human limb and thumb development patterns.

The study reinforces the conclusion that certain regions of genomes—those which are conserved across many species over evolutionary time and do not encode genes—can have a powerful regulatory influence on gene expression or the production of proteins.

"The study points to how human nucleotide substitutions can alter the regulation of genes in humans distinct from that of non-human primates, such as chimps," said one of the study's corresponding/senior authors Eddy Rubin, Director of Berkeley Lab's Genomics Division and the U.S. Department of Energy Joint Genome Institute. "This highlights a strategy that could be applied across the genome to understand at a molecular

level what leads to differences between humans and non-human primates."

The goal of the experiment was not to produce mice with human fingers. Rather, an indirect assay was employed to test the expression readout of a single human genetic fragment and compare it with the chimp version. The strategy, which links a color-inducing reporter gene when expression is activated, turns the targeted tissue blue, as it did in the published case. This led to the conclusion that the human-versus-chimp pattern of activation was different, consistent with the fast evolution of the human sequence, which may also give rise to a new function.

Previously published work from the Rubin lab by co-authors Shyam Prabhakar (now at the Genome Institute of Singapore), James Noonan (now at the Yale University School of Medicine), and postdoctoral fellows describes a global survey they conducted of genomes—human, chimpanzee, rhesus macaque, mouse, rat, and dog. They screened across these species to find the most conserved regions, but where humans had many more changes relative to the others. By comparing the occurrence of these features, they were seeking to home in on evidence of positive selection—sequence changes that evolve more rapidly since the human and chimp paths split six million years ago.

When queried about the extent of variation between humans and chimpanzees on a DNA level, the prevailing consensus amongst researchers is that humans are only about one percent different from our furrier friends—meaning one changed letter of code in one hundred nucleotides. It has long been postulated that the differences are not a consequence of protein-encoding changes, but must be attributed to a more cryptic mechanism because there are not enough sequence changes to account for the obvious differences.

Building on pioneering work in the Rubin lab on the identification and

characterization of these conserved noncoding sequences and the changes they induce in the genome the researchers generated a long list of such conserved elements. Using what is known as the "surprisal" test, a statistical tool for determining whether a particular event occurs at a rate greater than chance, they uncovered such a feature, dubbed human-accelerated conserved noncoding sequence 1 (HACNS1). Following the hypothesis that this was a gene regulatory sequence, they took this 546-base pair element, one that they determined has changed the most in humans relative to the other species over time, and plugged it into their mouse transgenic assay. They discovered that the human sequence enhances gene expression in limb and brain—a gain of function.

The next question that they asked was if the human and chimp elements have sequence differences, why do they still function similarly. They took the chimp version, placed it in an identical construct in mice and found that the human and chimp sequences have very different expression properties.

"Our results led us to believe that HACNS1 has contributed to uniquely human aspects of digit and limb patterning," Rubin said. "We suspect the gain of function in HACNS1 may have influenced the evolution of these or other human limb features by altering the expression of nearby genes during limb development."

The researchers have yet to define the precise molecular mechanism by which the substitutions in HACNS1 confer the human expression patterns. "To get a more complete picture of our HACNS1 factors into human morphological evolution will require additional studies," Rubin said.

"If this really is deterministic, you should see some type of change. But it will be a long, hard path forward," according to Rubin.

Source: Joint Genome Institute