SMP: Proteomics

Project: Functional Analysis of Human and Ape Proteomes

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Introduction

The origin of human-specific phenotypic traits, particularly those associated with complex cognitive abilities, is a question that has fascinated people for centuries. Recent determination of chimpanzee and other primate genome sequences provides a new means to study structural and functional aspects of human evolution and brings us one step closer to understanding the molecular mechanisms underlying human-specific abilities. Some human behavioral and cognitive traits have changed dramatically in the 5 to 7 million years since our evolutionary divergence from a common ancestor shared with chimpanzees. It is reasonable to assume that, on a phenotypic level, a number of these changes spread among humans due to positive Darwinian selection [1]. On the DNA sequence level, approximately 1.2% of the human genome differs from that of our closest living relatives, chimpanzees [2]. Although this difference may appear small, it accounts for more than 35,000,000 nucleotide substitutions, not including such differences as insertions, deletions and duplications. The majority of these differences; however, even those affecting the amino acid composition of proteins, are likely to be selectively neutral and thus have no detectable effect on phenotypic traits [3]. It has long been argued that, in addition to gene sequence differences, changes in RNA and protein expression may provide additional and crucial perspective on the evolutionary differences between humans and chimpanzees [4]. However, similar to DNA sequence differences, the majority of gene expression differences seen between species are likely to be selectively neutral [5]. Thus, the identification of genes affected by positive selection on either DNA sequence or RNA and protein expression levels during human evolution represents a challenge. Nevertheless, this is a challenge worth perusing—identification of positively selected genetic changes in humans can shed light on the molecular mechanisms underlying human-specific abilities.

Project Status

Genome and transcriptome evolution in different tissues

The chimpanzee genome sequence provides the first opportunity to study, on a genome-wide scale, the evolution of both gene expression and gene sequences in two closely-related mammals. In our current work, we compared humans and chimpanzees with respect to differences in expression levels and protein-coding sequences for genes expressed in brain, heart, liver, kidney and testis [6]. We analyzed the expression of approximately 21,000 human genes with Affymetrix U133plus2 and restricted the analysis to include only the 51,460 probe sets with identical sequences between the human and the chimpanzee genomes. We found that the patterns of differences in gene expression and in gene sequences are remarkably similar between the two species and consistent with a model of neutral evolution with negative selection. Thus, most observed differences in evolutionary patterns among the five tissues, on both gene expression and gene sequences levels, are due to differences in the amount of negative selection. More specifically, there is a gradation of selective constraints among the tissues such that brain shows the fewest differences between the species while liver shows the most. Furthermore, expression levels and amino acid sequences of genes that are expressed in many tissues show less divergence less between the species than genes that are expressed in only a few tissues. Interestingly, X-chromosomal genes expressed in testis show patterns suggestive of positive selection for both sequence changes and expression changes. In addition, despite the comparatively few amino acid sequence and expression differences seen between the species in brain, a larger proportion of these changes might have occurred on the human lineage than on the chimpanzee lineage.

These results suggest that evolutionary changes at both the level of gene regulation and the level of protein sequence have played crucial roles in the evolution of certain organ systems, such as those involved in cognition and male reproduction. Consequently, the modest number of human-chimpanzee gene sequence differences cannot be taken as evidence that regulatory changes would necessarily be more important than protein structural changes during human evolution. Rather, it is likely that both types of changes have acted in concert to give rise to the human phenotype.

Fig 1: *Our results provide the first clue why humans and chimpanzees age differently.*

Signatures of positive selection in the human brain

Our studies, as well as those of other research groups, have shown an excess of expression and protein sequence changes on the human evolutionary lineage as compared to the chimpanzee lineage in the brain but not in other tissues [6-9]. These findings are compatible with a relative increase in positive selection acting on brain-expressed genes during human evolution. Alternatively, they are also compatible with a relaxation of selective constraint on such genes leading to a smaller influence of negative selection. It is essential to distinguish between these two possibilities in order to identify positively selected changes. Positive selection events can be detected by the signature they leave on the patterns of nucleotide polymorphism within a species. One such signature is an extended non-random association among neighboring allelic variants known as linkage disequilibrium (LD).

In our current work, we combined publicly available LD data from three human populations with gene expression data from brain, heart, kidney and liver in humans, chimpanzees and orangutans. We found indications of recent positive selection acting on gene expression in human brain but not in the other three tissues or other species examined. Functional analysis based on Gene Ontology annotation shows that these positive selection events have lead to increased expression of genes involved in certain aspects of metabolism, transcriptional regulation and DNA repair. This finding is compatible with the hypothesis that changes in brain energy metabolism have been positively selected during the evolution of anthropoid primates (i.e. New World monkeys, Old World monkeys, apes and humans) due to an increased energy demand connected with changes in brain size and in maximal lifespan [10]. The latter possibility is particularly appealing since maximal lifespan in humans is believed to have increased relatively recently. In addition, gene expression changes associated with aging differ drastically between the human and chimpanzee cerebral cortex [11].

Outlook

Our results show that the combination of different types of biological data, such as gene expression, genome sequence and genetic polymorphism, can shed light on the molecular mechanisms underlying human-specific phenotypic features. It will be essential to include protein expression data in future efforts to understand the molecular basis of human-specific phenotypic features. Thus, future work will concentrate on the collection of protein expression data from humans and closely related primates.

*Lit.: **1.** Darwin, C., The origin of species by means of natural selection; or, the preservation of favored races in the struggle for life. 1859, London. xxi, 538 p. **2.** Ebersberger, I., et al., Genomewide Comparison of DNA Sequences between Humans and Chimpanzees. Am J Hum Genet, 2002. 70(6): p. 1490-7. **3.** Kimura, M., The neutral theory of molecular evolution. 1983, Cambridge [Cambridgeshire]; New York: Cambridge University Press. xv, 367. **4.** King, M.C. and A.C. Wilson, Evolution at two levels in humans and chimpanzees. Science, 1975. 188(4184): p. 107-16. **5.** Khaitovich, P., et al., A neutral model of transcriptome evolution. PLoS Biol, 2004. 2(5): p. E132. **6.** Khaitovich, P., et al., Parallel Patterns of Evolution in the Genomes and Transcriptomes of Humans and Chimpanzees. Science, 2005(in press). **7.** Enard, W., et al., Intra- and interspecific variation in primate gene expression patterns. Science, 2002. 296(5566): p. 340-3. **8.** Caceres, M., et al., Elevated gene expression levels distinguish human from non-human primate brains. Proc Natl Acad Sci U S A, 2003. 100(22): p. 13030-5. **9.** Dorus, S., et al., Accelerated evolution of nervous system genes in the origin of Homo sapiens. Cell, 2004. 119(7): p. 1027-40. **10.** Grossman, L.I., et al., Accelerated evolution of the electron transport chain in anthropoid primates. Trends Genet, 2004. 20(11): p. 578-85. **11.** Fraser, H.B., et al., Aging and Gene Expression in the Primate Brain. PLoS Biol, 2005. 3(9): p. e274.*