Science and Medicine

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Aussies out of Africa

An Aboriginal Australian genome reveals separate human dispersals into Asia

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Background and Methods: This study reports an Aboriginal Australian genomic sequence obtained from a 100-year-old lock of hair donated by an Aboriginal man from southern Western Australia in the early 20th century.

Results: The authors detected no evidence of European admixture and estimated contamination levels to be below 0.5%. They found that Aboriginal Australians are descendants of an early human dispersal into eastern Asia, possibly 62,000–75,000 years ago. This dispersal is separate from the one that gave rise to modern Asians 25,000–38,000 years ago. They also found evidence of gene flow between populations of the two dispersal waves prior to the divergence of Native Americans from modern Asian ancestors.

Conclusions: The findings support the hypothesis that present-day Aboriginal Australians descended from the earliest humans to occupy Australia, likely representing one of the oldest continuous populations outside of Africa.

In *Yearbook 2011* we highlighted the sequencing of the Neanderthal genome, which suggested that Europeans and Asians have inherited 2–6% of their genomes from Neanderthals. The current paper is an interesting report of genomic sequencing based on DNA extracted from a lock of hair collected 100 years ago on an Australian train, from what the collector described as a pure-blood native Aboriginal Australian. The findings show that people living in Southeast Asia and Australian Aboriginals inherited about 5% of their DNA from the Denisovans (linked only to a cave in southern Siberia), as well as 4–6% from Neanderthals, but that they mostly descend from the first humans who ventured out of Africa more than 60,000 years ago. It suggests at least two waves of human migration into Asia: early migrants that included the ancestors of contemporary Aboriginal Australians, New Guineans and some other Oceanians, followed by a second wave that gave rise to the present residents of mainland Asia. Some members of the two waves exchanged genes before the Denisovans vanished.

Discovery of sexual dimorphisms in metabolic and genetic biomarkers

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PLoS Genet 2011;7:e1002215

Background: Metabolomic profiling and the integration of genome-wide association study (GWAS) data is a powerful tool to comprehensively explore gene regulatory networks and to investigate the effects of genetic variation at the molecular level. Serum metabolite concentrations allow a direct readout of biological processes, and specific metabolomic signatures have been associated with complex diseases, such as Alzheimer's disease and cardiovascular and metabolic disorders. There are well-known differences between the sexes in the incidence, prevalence, age of onset, symptoms, and severity of many diseases, as well as drug responses. However, most of the studies published so far did not analyze their data stratified by gender.

Methods: This study investigated sex-specific differences in serum metabolite concentrations and their underlying genetic determination. Discovery and replication stages were based on over 3,300 independent individuals from KORA F3 and F4 with measurements of 131 metabolites, including amino acids, phosphatidylcholines, sphingomyelins, acylcarnitines, and C6 sugars.

Results: Significant differences in concentrations between males and females were found for 102/131 metabolites (p values $<3.8 \times 10^{-4}$; Bonferroni-corrected threshold). Sex-specific GWAS showed genome-wide significant differences in β estimates between SNPs in the CPS1 locus (carbamoyl-phosphate synthase 1, $p <3.8 \times 10^{-10}$) and glycine concentrations. The metabolite profiles of males and females were differed significantly and specific genetic variants in metabolism-related genes depicted sexual dimorphism.

Conclusions: This study provides new important insights into sex-specific differences in cell regulatory processes and underscores that studies should consider sex-specific effects in design and interpretation.

The new science of metabolomics addresses the entire metabolic tree, or at least large groups of metabolites, rather than each metabolite on its own. Thus, each individual has his/her own fingerprint metabolomic profile. Here is a no-surprise for a pediatric endocrinologist: the serum metabolomic profile shows significant differences between men and women, for 101 of the 131 metabolites, mostly in lipid and amino acid species. The same is true for people with different body composition patterns. Therefore, any analysis of metabolomic profiles must be normalized with respect to gender and body composition. It also means that gender-specific therapies may be required for diseases with an important metabolic component.

Bile away your fat

A nuclear-receptor-dependent phosphatidylcholine pathway with antidiabetic effects

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Nature 2011;474:506–510

Background: Nuclear hormone receptors regulate diverse metabolic pathways and the orphan nuclear receptor LRH-1 (also known as NR5A2) regulates bile acid biosynthesis. Structural studies have identified phospholipids as potential LRH-1 ligands, but their functional relevance is unclear. Here, the authors show that an unusual phosphatidylcholine species with two saturated 12 carbon fatty acid acyl side chains (dilauroyl phosphatidylcholine (DLPC)) is an LRH-1 agonist ligand in vitro.

Methods and Results: DLPC treatment induced bile acid biosynthetic enzymes in mouse liver, increased bile acid levels, and lowered hepatic triglycerides and serum glucose. DLPC treatment also decreased hepatic steatosis and improved glucose homeostasis in two mouse models of insulin resistance. Both the antidiabetic and lipotropic effects were lost in liver-specific *Lrh-1* knockouts.

Conclusions: These findings identify an LRH-1-dependent phosphatidylcholine signaling pathway that regulates bile acid metabolism and glucose homeostasis.

It is known that bile acids ward off fatty liver disease and maintain stable blood sugar levels. Therefore, it would be helpful if we could enhance the production of bile acids. To explore this possibility, Lee et al. investigated the liver receptor homolog-1 (LRH-1) protein, which maintains bile acid excretion. They found that a dilauroyl phosphatidylcholine (DLPC), a trace component of lecithin, activated both the mouse and human forms of the protein and mildly increased bile acid levels in the blood, which in turn reduced fat generation by the liver, reduced fatty liver disease, and improved glucose tolerance and insulin sensitivity. It is a fairly small compound and hopefully it will not be long before it is tested in obese patients with fatty liver disease.

Light relief

UVA phototransduction drives early melanin synthesis in human melanocytes

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Curr Biol 2011;21:1906–1911

Background: Exposure of human skin to solar ultraviolet radiation (UVR), a powerful carcinogen comprising ~95% ultraviolet A (UVA) and ~5% ultraviolet B (UVB) at the Earth's surface, promotes melanin synthesis in epidermal melanocytes, which protects skin from DNA damage. UVB causes DNA lesions that lead to transcriptional activation of melanin-producing enzymes, resulting in delayed skin pigmentation within days. In contrast, UVA causes primarily oxidative damage and leads to immediate pigment darkening (IPD) within minutes, via an unknown mechanism. No receptor protein directly mediating phototransduction in skin has been identified.

Methods and Results: The authors demonstrate that exposure of primary human epidermal melanocytes (HEMs) to UVA causes calcium mobilization and early melanin synthesis. Calcium responses were abolished by treatment with G protein or phospholipase C (PLC) inhibitors or by depletion of intracellular calcium stores. They show that the visual photopigment rhodopsin is expressed in HEMs and contributes to UVR phototransduction. Upon UVR exposure, significant melanin production was measured within 1 h; cellular melanin continued to increase in a retinal- and calcium-dependent manner up to fivefold after 24 h.

Conclusions: The findings identify a novel UVA-sensitive signaling pathway in melanocytes that leads to calcium mobilization and melanin synthesis and may underlie the mechanism of IPD in human skin.

Surprisingly, the photosensitive receptor rhodopsin, previously thought to exist only in the eye, is also expressed in melanocyte skin cells in response to UV irradiation, where it triggers the generation of melanin (tanning) – the natural sunscreen that protects us from UVB-induced DNA damage. The fine balance between too little UVB exposure, that will cause vitamin D deficiency, and too much UVB, and the ensuing folic acid deficiency, turns out to use rhodopsin activation not by an enzymatic reaction but by a photosensitive mechanism – just like photosynthesis in plants.

A cardiac microRNA governs systemic energy homeostasis by regulation of MED13

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Cell 2012;149:671–683

Background: Obesity, type 2 diabetes, and heart failure are associated with aberrant cardiac metabolism.

Methods and Results: The paper shows that the heart regulates systemic energy homeostasis via MED13, a subunit of the Mediator complex, which controls transcription by thyroid hormone and other nuclear hormone receptors. MED13, in turn, is negatively regulated by a heart-specific microRNA, miR-208a. Cardiac-specific overexpression of MED13 or pharmacologic inhibition of miR-208a in mice confers resistance to high-fat diet-induced obesity and improves systemic insulin sensitivity and glucose tolerance. Conversely, genetic deletion of MED13 specifically in cardiomyocytes enhances obesity in response to high-fat diet and exacerbates metabolic syndrome. The metabolic actions of MED13 result from increased energy expenditure and regulation of numerous genes involved in energy balance in the heart.

Conclusions: These findings reveal a role of the heart in systemic metabolic control and point to MED13 and miR-208a as potential therapeutic targets for metabolic disorders.

The ultimate complication of obesity, the metabolic syndrome and type 2 diabetes is cardiac disease. Research has focused on metabolic aberrations and how they compromise the cardiovascular system. Grueter et al. suggest that the heart itself may regulate whole-body metabolism. Their work focuses on MED13, a regulatory subunit of Mediator (a protein complex bridging transcription factors and RNA polymerase). Cardiac-specific overexpression of MED13 results in an increase in energy expenditure and resistance to diet-induced obesity and metabolism by altering expression of genes controlled by the thyroid hormone receptor. Another important aspect of this paper is that cardiac expression of MED13 is negatively regulated by a microRNA that is encoded by an intron of a cardiac-specific myosin gene. Promising direction inhibition of this miRNA in wild-type mice on a high-fat diet slowed both weight gain and the development of glucose intolerance.

Expressed by training

Acute exercise remodels promoter methylation in human skeletal muscle

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Cell Metab 2012;15:405–411

Background: DNA methylation is a covalent biochemical modification controlling chromatin structure and gene expression. Exercise elicits gene expression changes that trigger structural and metabolic adaptations in skeletal muscle.

Methods and Results: This work aimed to determine whether DNA methylation plays a role in exercise-induced gene expression. Whole genome methylation was decreased in skeletal muscle biopsies obtained from healthy sedentary men and women after acute exercise. Exercise induced a dose-dependent expression of PGC-1 α , PDK4, and PPAR- δ , together with a marked hypomethylation on each respective promoter. Similarly, promoter methylation of PGC-1 α , PDK4, and PPAR- δ was markedly decreased in mouse soleus muscles 45 min after ex-vivo contraction. In L6 myotubes, caffeine exposure induced gene hypomethylation in parallel with an increase in the respective mRNA content.

Conclusions: These results provide evidence that acute gene activation is associated with a dynamic change in DNA methylation in skeletal muscle and suggest that DNA hypomethylation is an early event in contraction-induced gene activation.

Epigenetic changes are much faster than we hitherto believed; transient promoter methylation occurs after an acute exercise. The same effect was achieved by ex-vivo muscle contraction and caffeine-mediated DNA hypomethylation in myocytes, implicating a role for Ca^{2+} release. Thus, genes can be activated by contraction, and this is preceded by their demethylation.

Prolonging the life of mammals

The sirtuin SIRT6 regulates lifespan in male mice

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Nature 2012;483:218–221

Background: The significant increase in human lifespan during the past century confronts us with great medical challenges. To meet these challenges, the mechanisms that determine healthy aging must be understood. Sirtuins are highly conserved deacetylases that have been shown to regulate lifespan in yeast, nematodes and fruit flies. However, the role of sirtuins in regulating worm and fly lifespan has recently become controversial. Moreover, the role of the seven mammalian sirtuins, SIRT1 to SIRT7 (homologues of the yeast sirtuin Sir2), in regulating lifespan is unclear.

Methods and Results: This study shows that male, but not female, transgenic mice overexpressing Sirt6 have a significantly longer lifespan than wild-type mice. Gene expression analysis revealed significant differences between male Sirt6-transgenic mice and male wild-type mice: transgenic males displayed lower serum levels of insulin-like growth factor 1 (IGF1), higher levels of IGF-binding protein 1 and altered phosphorylation levels of major components of IGF1 signaling, a key pathway in the regulation of lifespan.

Conclusions: This study shows the regulation of mammalian lifespan by a sirtuin family member and has important therapeutic implications for age-related diseases.

Mammals have 7 deacetylases that are labeled sirtuins (Sir1–Sir7). These are highly conserved NAD^{+} -dependent deacetylases that regulate lifespan in several organisms. Increasing the sirtuin level through genetic manipulation extends the lifespan of yeast, nematodes and flies. Some mammalian sirtuins regulate age-related diseases, but mice that overexpress SIRT1 have the same lifespan as control WT mice. We now learn that it is Sirt6 that prolongs life in mice; transgenic males displayed lower serum IGF1 and higher IGFBP1. What comes to mind next are the therapeutic implications for age-related diseases. And why do females not respond? Mice with a fat-specific insulin receptor gene knockout have been shown to have an increased mean lifespan of similar magnitude to that of SIRT6e, demonstrating the central role of fat in regulating lifespan.

Clearance of p16Ink4a-positive senescent cells delays aging-associated disorders

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Nature 2011;479:232–236

Background: Advanced age is the main risk factor for most chronic diseases and functional deficits in humans, but the fundamental mechanisms that drive aging remain largely unknown, impeding the development of interventions that might delay or prevent age-related disorders and maximize healthy lifespan. Cellular senescence, which halts the proliferation of damaged or dysfunctional cells, is an important mechanism to constrain the malignant progression of tumor cells. Senescent cells accumulate in various tissues and organs with aging and have been hypothesized to disrupt tissue structure and function because of the components they secrete.

Methods: This study examined whether senescent cells are causally implicated in age-related dysfunction and whether their removal is beneficial. To address these fundamental questions, they made use of a biomarker for senescence, p16Ink4a, to design a novel transgene, INK-ATTAC, for inducible elimination of p16Ink4a-positive senescent cells upon administration of a drug.

Results: In the BubR1 progeroid mouse background, INK-ATTAC removed p16Ink4a-positive senescent cells upon drug treatment. In tissues, such as adipose tissue, skeletal muscle and eye, in which p16Ink4a contributes to the acquisition of age-related pathologies, life-long removal of p16Ink4a-expressing cells delayed onset of these phenotypes. Furthermore, late-life clearance attenuated progression of already established age-related disorders.

Conclusions: Cellular senescence is causally implicated in generating age-related phenotypes and removal of senescent cells can prevent or delay tissue dysfunction and extend health span.

After dividing a limited number of times, cells enter a senescent nonproliferative phase. Baker et al. used genetic engineering to eliminate ensuing senescent cells. Mice with no senescent cells did not live longer than normal, but they lived better; they showed delayed onset of cataracts, fat accumulation, and muscle weakness, they could run for a longer time on a treadmill and perform more strenuous workouts – we remain hopeful for drug that will eliminate our own senescent cells.

Ineffective short-term diets

Linking long-term dietary patterns with gut microbial enterotypes

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Science 2011;334:105–108

Background: Diet strongly affects human health, partly by modulating gut microbiome composition.

Methods: The authors used diet inventories and 16S rDNA sequencing to characterize fecal samples from 98 individuals.

Results: Fecal communities clustered into enterotypes distinguished primarily by levels of *Bacteroides* and *Prevotella*. Enterotypes were strongly associated with long-term diets, particularly protein and animal fat (*Bacteroides*) versus carbohydrates (*Prevotella*). A controlled-feeding study of 10 subjects showed that microbiome composition changed detectably within 24 h of initiating a high-fat/low-fiber or low-fat/high-fiber diet, but that enterotype identity remained stable during the 10-day study.

Conclusions: Alternative enterotype states are associated with long-term diet.

The microbiome has been the focus of much interest and hope, particularly in the field of obesity. More recently the practice of fecal (microbiome) transplantation has been revived and received scientific evidence in the cure of broad-spectrum antibiotic-induced enteropathy [1]. The multispecies microbiome was classified, to include three enterotypes, named *Bacteroides*, *Prevotella*, and *Ruminococcus*. These enterotypes correlate with long-term diet, but not with age, weight, sex, or country; they can differ even between twin siblings. *Bacteroides* grow on high-meat diets; *Prevotella* on vegetarian food. But changes in the microbiome do not occur fast; Wu et al. show that 10 days of a dietary change makes no difference to your enterotype, suggesting that they are more influenced by long-term eating trends.

In utero epigenomic programming

Epigenetic mechanism underlying the development of polycystic ovary syndrome-like phenotypes in prenatally androgenized rhesus monkeys

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Context: The pathogenesis of polycystic ovary syndrome (PCOS) is poorly understood. PCOS-like phenotypes are produced by prenatal androgenization (PA) of female rhesus monkeys.

Hypothesis: Perturbation of the epigenome, through altered DNA methylation, is one of the mechanisms whereby PA reprograms monkeys to develop PCOS.

Methods: Infant and adult visceral adipose tissues (VAT) harvested from 15 PA and 10 control monkeys were studied. Bisulfite-treated samples were subjected to genome-wide CpG methylation analysis, designed to simultaneously measure methylation levels at 27,578 CpG sites. Analysis was carried out using Bayesian Classification with Singular Value Decomposition (BCSVD), testing all probes simultaneously in a single test. Stringent criteria were then applied to filter out invalid probes due to sequence dissimilarities between human probes and monkey DNA, and then mapped to the rhesus genome.

Outcomes: This yielded differentially methylated loci between PA and control monkeys, 163 in infant VAT, and 325 in adult VAT (BCSVD $p < 0.05$). Among these two sets of genes, we identified several significant pathways, including the antiproliferative role of TOB in T-cell signaling and transforming growth factor- β (TGF- β) signaling.

Conclusions: PA may modify DNA methylation patterns in both infant and adult VAT. This pilot study suggests that excess fetal androgen exposure in female nonhuman primates may predispose to PCOS via alteration of the epigenome, providing a novel avenue to understand PCOS in humans.

PCOS-like phenotypes are produced by prenatal androgenization (PA) of rhesus monkeys, sheep, rats and mice, including hyperandrogenism, oligomenorrhea, polyfollicular ovaries, increased adiposity, insulin resistance and impaired insulin secretion. Does this indicate that the intrauterine environment plays a role in the etiology of PCOS? The current study performed a genome-wide site-specific methylation array of visceral adipose tissue (VAT) from PA female rhesus monkeys. They report differentially methylated loci between PA and control monkeys' VAT. Among these are the antiproliferative T-cell signaling and TGF- β signaling, a pathway currently implicated in the pathogenesis of PCOS in women by genetic epidemiologic evidence [3]. Once again, epigenomics may be more relevant to diseases of modern civilization than genetics.

Transgenerational epigenetic instability is a source of novel methylation variants

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Science 2011;334:369–373

Background: Epigenetic information, which may affect an organisms' phenotype, can be stored and stably inherited in the form of cytosine DNA methylation. Changes in DNA methylation can produce meiotically stable epialleles that affect transcription and morphology, but the rates of spontaneous gain or loss of DNA methylation are unknown.

Methods: The authors examined spontaneously occurring variation in DNA methylation in *Arabidopsis thaliana* plants propagated by single-seed descent for 30 generations.

Results: 114,287 CG single methylation polymorphisms (SMPs) and 2,485 CG differentially methylated regions (DMRs) were identified, both of which showed patterns of divergence compared to the ancestral state.

Conclusions: Transgenerational epigenetic variation in DNA methylation can generate new allelic states that alter transcription providing a mechanism for inheritance of phenotypic diversity in the absence of genetic mutation.

But how can epigenetic variation lead to stable epimutations and gene sequence variants? See below.

Methylation stabilizes the genome

Genomic hypomethylation in the human germline associates with selective structural mutability in the human genome

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PLoS Genet 2012;8:e1002692

Background: The hotspots of structural polymorphisms and structural mutability in the human genome remain to be explained mechanistically. The authors examined associations between structural mutability and germline DNA methylation and with nonallelic homologous recombination (NAHR) mediated by low-copy repeats (LCRs).

Methods and Results: Combined evidence from four human sperm methylome maps, human genome evolution, structural polymorphisms in the human population, and previous genomic and disease studies consistently points to a strong association between germline hypomethylation and genomic instability. Specifically, methylation deserts, the ~1% fraction of the human genome with the lowest methylation in the germline, show tenfold enrichment for structural rearrangements that occurred in the human genome since the branching of chimpanzee and are highly enriched for fast-evolving loci that regulate tissue-specific gene expression. Analysis of copy number variants (CNVs) from 400 human samples identified using a custom-designed array comparative genomic hybridization (aCGH) chip, combined with publicly available structural variation data, indicates that association between structural mutability and germline hypomethylation is comparable in magnitude to the association between structural mutability and LCR-mediated NAHR. Moreover, rare CNVs occurring in the genomes of individuals diagnosed with schizophrenia, bipolar disorder, and developmental delay and de novo CNVs occurring in those diagnosed with autism are significantly more concentrated within hypomethylated regions.

Conclusions: These findings suggest a new connection between the epigenome, selective mutability, evolution, and human disease.

Approximately 10% of the human genome is structurally polymorphic at the submicroscopic scale (<4 Mb), a much larger fraction than affected by single nucleotide polymorphisms (SNPs). Structural mutations that occur in a number of well-studied unstable loci cause disease. The discovery of these structurally mutable disease-associated loci gave rise to the concept of genomic disorders. This excessive mutability has also accelerated evolution. Lack of methylation (hypomethylation) of genomic DNA has been previously associated with high structural mutability in cancer cells, and in this paper Li et al. related hypomethylation to structural mutability in the human germline. They confirm the role of low-copy repeats (LCRs) in promoting structural mutability on the genome scale and also reveal a surprisingly strong association between genomic instability and hypomethylation; the ~1% fraction of the human genome with the lowest methylation in human sperm harbors a tenfold higher number of structural mutations than the genome-wide average. This suggests a new connection between adaptive plasticity responses to the environment and heritable changes in gene sequence, which provide long-term evolutionary variation.

Unsilencing father's methylation

Topoisomerase inhibitors unsilence the dormant allele of Ube3a in neurons

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Nature 2012;481:185–189

Background: Angelman syndrome is a severe neurodevelopmental disorder caused by deletion or mutation of the maternally-inherited ubiquitin protein ligase E3A gene (UBE3A). The paternal allele of UBE3A is intact but is epigenetically silenced, raising the possibility that Angelman syndrome could be treated if the silenced allele could be activated to restore functional UBE3A protein.

Methods and Results: The authors used an unbiased, high-content drug screen, in primary cortical neurons from mice, and identified twelve topoisomerase I inhibitors and four topoisomerase II inhibitors that unsilenced the paternal Ube3a allele. These drugs included topotecan, irinotecan, etoposide and dexrazoxane (ICRF-187). At nanomolar concentrations, topotecan upregulated catalytically active UBE3A in neurons from maternal Ube3a-null mice. Topotecan downregulated expression of the Ube3a antisense transcript that overlaps the paternal copy of Ube3a. When administered in vivo, topotecan unsilenced the paternal Ube3a allele in neurons in the hippocampus, neocortex, striatum, cerebellum and spinal cord. Paternal Ube3a expression remained elevated in a subset of spinal cord neurons for at least 12 weeks after cessation of topotecan treatment.

Conclusions: These results indicate that topotecan unsilences Ube3a in cis by reducing transcription of an imprinted antisense RNA, and suggest a therapeutic strategy for reactivating the dormant but functional allele of Ube3a in patients with Angelman syndrome.

Angelman syndrome (AS) is often widely taught as the exemplar of imprinted gene disorders, as it is caused by the specific loss of the maternally-inherited UBE3A gene. Conversely, defects in the corresponding paternally-inherited gene region cause Prader-Willi syndrome. The frequent outbreaks of laughter and jerky movements characteristic of AS gave it the eponym 'happy puppet syndrome'. However the condition is far from a happy one, being usually marked by seizures, sleep disturbance and severe learning difficulties and, like many inherited neurological disorders, no effective treatment strategies exist. The authors explored the intriguing possibility that the intact but dormant paternal UBE3A allele might be awakened. To do this, they screened 2,306 drug-like molecules using a primary mouse cortical neuron assay that linked paternal Ube3a to a yellow fluorescent protein. Their findings showed that transient topoisomerase inhibition, using drugs that are currently used in treating cancer, has lasting effects on paternal Ube3a gene expression and therefore may have potential benefits for patients with AS.

Accelerated recruitment of new brain development genes into the human genome

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PLoS Biol 2011;9:e1001179

Background: How the human brain evolved has attracted tremendous interest for decades.

Methods: Motivated by case studies of primate-specific genes implicated in brain function, the authors examined whether the young genes, those emerging genome-wide in the lineages specific to the primates or rodents, showed distinct spatial and temporal patterns of transcription compared to old genes, which had existed before primates and rodents split.

Results: The authors found consistent patterns across different sources of expression data: there were significantly larger proportions of young genes expressed in the fetal or infant brains of humans than of the mouse, and more young genes in humans have expression biased toward early developing brains than old genes. Most of these young genes are expressed in the evolutionarily newest part of human brain, the neocortex. Remarkably, they also identified a number of human-specific genes expressed in the prefrontal cortex, which is implicated in complex cognitive behaviors. The young genes upregulated in the early developing human brain have diverse functions, with a significant enrichment of transcription factors. Genes originating from different mechanisms show a similar expression bias in the developing brain. Moreover, the young genes upregulated in early brain development showed rapid protein evolution compared to old genes also expressed in the fetal brain. Strikingly, genes expressed in the neocortex arose soon after its morphological origin.

Conclusions: These four lines of evidence suggest that positive selection for brain function may have contributed to the origins of young genes expressed in the developing brain. These data demonstrate a striking recruitment of new genes into the early development of the human brain.

MicroRNA-driven developmental remodeling in the brain distinguishes humans from other primates

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PLoS Biol 2011;9:e1001214

Background: While many studies have reported the accelerated evolution of brain gene expression in the human lineage, the mechanisms underlying such changes are unknown.

Methods: The authors address this issue from a developmental perspective by analyzing mRNA and microRNA (miRNA) expression in two brain regions within macaques, chimpanzees, and humans throughout their lifespan.

Results: Constitutive gene expression divergence (differences between species independent of age) is comparable between humans and chimpanzees. However, humans display a 3–5 times faster evolutionary rate in divergence of developmental patterns, compared to chimpanzees. Such accelerated evolution of human brain developmental patterns (i) cannot be explained by life-history changes among species, (ii) is twice as pronounced in the prefrontal cortex than the cerebellum, (iii) preferentially affects neuron-related genes, and (iv) unlike constitutive divergence does not depend on *cis*-regulatory changes, but might be driven by human-specific changes in expression of *trans*-acting regulators. Developmental profiles of miRNAs, as well as their target genes, showed the fastest rates of human-specific evolutionary change. Using a combination of computational and experimental methods, the authors identified miR-92a, miR-454, and miR-320b as possible regulators of human-specific neural development.

Conclusions: Different mechanisms underlie adaptive and neutral transcriptome divergence. Changes in the expression of a few key regulators may have been a major driving force behind rapid evolution of the human brain.

Human evolution is dominated by investment in big brains with unique cognitive abilities. While there have been no major genome-wide changes in the coding regions of brain-related genes, changes in the regulation of these genes have played a key role. Zhang et al. examined the expression profile of genes in both fetal and adult brains in the human and mouse, and discovered that an excess of recently evolved genes are expressed in the early (fetal or infant) developing human brain compared with those in mouse brain. Apparently, evolutionary change in the development of the human brain happened at the protein level by gene origination and also via evolution of regulatory networks. Somel et al. compared gene-expression patterns in the brains of humans, chimpanzees and rhesus, and show that human brains showed many more differences in gene-expression patterns as newborns developed into adults, particularly in cognition-specific regions. Genes that encode miRNAs were among those whose expression varied. Somel et al. suggest that a small number of miRNAs and proteins controlling brain development could have driven the evolution of the human brain.

Electrifying embryology

Transmembrane voltage potential controls embryonic eye patterning in *Xenopus laevis*

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Development 2012;139:313–323

Background: Uncovering the molecular mechanisms of eye development is crucial for understanding the embryonic morphogenesis of complex structures, as well as for the establishment of novel biomedical approaches to address birth defects and injuries of the visual system. During normal embryogenesis, a striking hyperpolarization demarcates a specific cluster of cells in the anterior neural field.

Methods and Results: To characterize change in transmembrane voltage potential (V_{mem}) as a novel biophysical signal for eye induction in *Xenopus laevis*. Depolarizing the dorsal lineages in which these cells reside results in malformed eyes. Manipulating V_{mem} of non-eye cells induces well-formed ectopic eyes that are morphologically and histologically similar to endogenous eyes. Remarkably, such ectopic eyes can be induced far outside the anterior neural field. A Ca^{2+} channel-dependent pathway transduces the V_{mem} signal and regulates patterning of eye field transcription factors.

Conclusions: These data reveal a new, instructive role for membrane voltage during embryogenesis and demonstrate that V_{mem} is a crucial upstream signal in eye development. Learning to control bioelectric initiators of organogenesis offers significant insight into birth defects that affect the eye and might have significant implications for regenerative approaches to ocular diseases.

Complex body structures arise from simple early embryonic structure by timed action of a relatively small ‘tool box’ of two dozen transcription factors. Pai et al. show how bioelectrical communications among cells specify the type of new organ to be created at a particular location. Manipulation of membrane voltage in *Xenopus* embryos caused the frog to grow eyes in the back and tail. By RNA encoding of ion channels, the researchers changed the membrane potential of these cell, causing abnormalities to occur. It seems that for every structure in the body there is a specific membrane voltage range that drives organogenesis.

Ecological specialization in fossil mammals explains Cope's rule

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Background: Cope's rule is the trend toward increasing body size in a lineage over geological time. The rule has been explained either as passive diffusion away from a small initial body size or as an active trend upheld by the ecological and evolutionary advantages that large body size confers.

Methods: An explicit and phylogenetically informed analysis of body size evolution in Cenozoic mammals.

Results: Results show that body size increases significantly in most inclusive clades. This increase occurs through temporal substitution of incumbent species by larger-sized close relatives within the clades. These late-appearing species have smaller spatial and temporal ranges and are rarer than the incumbents they replace, traits that are typical of ecological specialists. Overlain on a net trend toward average size increase, significant pulses in origination of large-sized species are concentrated in periods of global cooling.

Conclusions: Cope's rule appears to derive mainly from increasing ecological specialization and clade-level niche expansion rather than from active selection for larger size. Cooling pulses plausibly record direct selection for larger body size according to Bergmann's rule, which thus appears to be independent of but concomitant with Cope's.

As much as hominids grew strategically from Lucy's 105 cm to present-day females' 165 cm, increasing body size in a lineage over geological time periods is common: members of most lineages get bigger over time deep time. The authors of this paper composed phylogenetic trees of 554 mammal species and confirm the rule: most have become bigger: small and unspecialized species are 'replaced' by larger species. Most of the peaks in body size increase occur alongside changes in the climate; drop in temperature resulted in an increase in size.

Evolution of the earliest horses driven by climate change in the Paleocene-Eocene thermal maximum

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Background: Body size plays a critical role in mammalian ecology and physiology. Previous research has shown that many mammals became smaller during the Paleocene-Eocene thermal maximum (PETM), the period of profound global warming, but the timing and magnitude of that change relative to climate change are unclear.

Methods and Results: A high-resolution record of continental climate and equid body size change shows a directional decrease in size of ~30% over the first ~130,000 years of the PETM, followed by a ~76% increase during the recovery phase of the PETM. These size changes are negatively correlated with temperature, as inferred from oxygen isotopes in mammal teeth, and were probably driven by shifts in temperature and possibly high atmospheric CO₂ concentrations.

Conclusions: These findings could be important for understanding mammalian evolutionary responses to future global warming.

Body size sets the energetic demands of organisms; big animals consume more energy than small animals. Children in underprivileged nutritional environments are not 'stunted', but are adequately adapted to low energy cues. Yet, the survival and fitness advantage of being big make animals, including humans, grow bigger over evolutionary time for as long as energy is available. Thus, the body sizes of mammal lineages have varied greatly over evolutionary history. The extinction of dinosaurs ~65.5 million years ago eliminated big animals, but at the same time enhanced ecological (and therefore

phenotypic) diversification, which ultimately led to size increases to current magnitudes. Secord et al. illustrate the well-known 'Bergmann's rule' – the critical role of temperature in driving body size evolution; larger sizes are found in colder environments, and smaller sizes in warmer areas. Body size decreased by ~30% at the start of the PETM, and increased abruptly by more than 75% at its end. Recent global warming has already led to changes in the distribution and morphology of species.

Change and variability in Plio-Pleistocene climates: modeling the hominin response

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Background: Research into the links between climatic change and hominin evolution has generated numerous hypotheses. In recent years, methodological refinement of, and increased research effort directed towards, reliable proxies for paleoclimatic change have provided a growing body of data with which to test such hypotheses. Whilst many archaeologists are aware of these data, few are cognizant of the wealth of techniques developed by theoretical biologists over the last half century to explicitly address the evolutionary consequences of adaptation to temporally heterogeneous environments.

Methods: This study expands and adapts one such technique for use with empirical data, and applies it to a global paleoclimatic record spanning the last 5 million years, in order to discern the potential impact of environmental heterogeneity on hominin evolution during this period. Of particular interest are the contributions of climatic change, associated with directional selection, and climatic variability, associated with selection for phenotypic plasticity.

Results: At this macro-scale, the author finds an early peak in selection for plasticity at approximately 2–2.7 million years ago (mya), combined with three major shifts in directional selection at approximately 3.3–3.4, 1.4–1.5, and 0.5–0.6 mya.

Conclusions: These results link the fossil and archaeological records to a number of environmental hypotheses of human evolution. In particular, it is argued that the origins of the genus *Homo* and the spread of Oldowan technology are associated not with a major turnover pulse, but with a period of selection for phenotypic plasticity.

Climatic changes are among the most powerful forces directing evolutionary events. Among such events are (1) hominids' trend to become bigger in size over the last 4.5 million years, and (2) a very marked plasticity of hominids enabling us to adapt to various conflicting environmental cues. This article calls for a distinction between climatic change and climatic variability. Whereas climatic change results in directional selection, such as progressive growth when the earth cools, climatic variability selects for plasticity. The results suggest that selection for plasticity increased 2.3–2.5 million years ago. This date range coincides with the evolution of *Homo* and the spread of the Oldowan. Plasticity is a fundamental requirement for the unique human adaptability, enabling *Homo sapiens* to live in Greenland and the Sahara, in the Himalaya and the Dead Sea.

A small puzzle solved

LB1 and LB6 *Homo floresiensis* are not modern human (*Homo sapiens*) cretins

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Background: Excavations in the late Pleistocene deposits at Liang Bua cave, Flores, have uncovered the skeletal remains of several small-bodied and small-brained hominins in association with stone artifacts

and the bones of *Stegodon*. Due to their combination of plesiomorphic, unique and derived traits, they were ascribed to a new species, *Homo floresiensis*, which, along with *Stegodon*, appears to have become extinct ~17 ka (thousand years ago). However, recently it has been argued that several characteristics of *H. floresiensis* were consistent with dwarfism and evidence of delayed development in modern human (*Homo sapiens*) myxedematous endemic (ME) cretins.

Methods and Results: This research compared the skeletal and dental morphology in *H. floresiensis* with the clinical and osteological indicators of cretinism, and the traits that have been argued to be associated with ME cretinism in LB1 and LB6. Contrary to published claims, morphological and statistical comparisons did not identify the distinctive skeletal and dental indicators of cretinism in LB1 or LB6 *H. floresiensis*. Brain mass, skeletal proportions, epiphyseal union, orofacial morphology, dental development, size of the pituitary fossa and development of the paranasal sinuses, vault bone thickness and dimensions of the hands and feet all distinguish *H. floresiensis* from modern humans with ME cretinism.

Conclusions: The research team responsible for the diagnosis of ME cretinism had not examined the original *H. floresiensis* skeletal materials, and perhaps, as a result, their research confused taphonomic damage with evidence of disease, and thus contained critical errors of fact and interpretation. Behavioral scenarios attempting to explain the presence of cretinous *H. sapiens* in the Liang Bua Pleistocene deposits, but not unaffected *H. sapiens*, are both unnecessary and not supported by the available archaeological and geochronological evidence from Flores.

Yearbook has followed the saga of the hobbits from the Indonesian island Flores from its first publication (*Yearbook* 2005). With an average height of <1 m, a weight of 1 kg and brain weight of 380 g, it was debated whether these people who roamed the island 18,000 years ago were short-statured *H. sapiens*, or a new species. It evoked then the phenomenon of insular dwarfism, the phenomenon of plants and animals growing short when resources are limited and when there is no need to grow big for lack of predators. Others suggested that these were cretins or Laron dwarfs. Over the years the Flores hobbits were a matter of much debate and ugly politics of science. From available archaeological and geochronological evidence from Flores it is now clear that these were members of a new species, now labeled *H. floresiensis*. This is even more exciting, and the origin of this species is yet to be deciphered.

Last but not least

Phenotypic correlates of male reproductive success in western gorillas

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Background: Sexual selection is thought to drive the evolution of sexually dimorphic traits that increase male reproductive success. Despite a large degree of sexual dimorphism among haplorhine primates, phenotypic traits that may influence the reproductive success of males are largely unstudied due to their long lifespans and the difficulties in quantifying such traits noninvasively.

Methods: The authors employed digital photogrammetry of body length and crest size, as well as ranking of the gluteal muscle size, to test whether these sexually dimorphic traits are associated with long-term measures of male reproductive success in western gorillas.

Results: Among 19 adult male gorillas monitored for up to 12.5 years, all three physical traits were positively correlated with the average number of mates per male, but only crest size and gluteal muscle size were significantly correlated with offspring survival and the annual rate of siring offspring that survive to weaning age.

Conclusions: The authors discuss why such sexually dimorphic traits might be under ongoing selection in gorillas and other species.

Examination of morphological and behavioral features of great apes provides insights into the group structure, and social behavior of our ancestors. Gorilla males are twice as large in mass as females, and have markedly larger gluteus muscles. When it comes to sexual selection, longer males are preferred by females. Yet it is only the gluteus mass that predicts better survival of the offspring. Female gorillas look at the wrong trait: they find long males to be attractive, but crest size and gluteus musculature may be imperative; they correlate with the ability of the male to provide protection to offspring.

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