

## Contents

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### INVESTIGATIONS

- 553–560 **Biparental Resequencing Coupled With SNP Genotyping of a Segregating Population Offers Insights Into the Landscape of Recombination and Fixed Genomic Regions in Elite Soybean**  
*Ying-hui Li, Yu-lin Liu, Jochen C. Reif, Zhang-xiong Liu, Bo Liu, Michael F. Mette, Ru-zhen Chang, and Li-juan Qiu*
- To study the landscape of fixed genomic regions and recombination in soybean, we re-sequenced two parental elite soybean lines and developed a customized single-nucleotide polymorphism (SNP) array for fingerprinting of the corresponding segregating bi-parental population. The results suggested that this technology could efficiently bridge the genotyping gap and provide deep insights into the landscape of recombination and fixed genomic regions in bi-parental segregating populations of soybean with implications for fine mapping of QTL.
- 561–578 **Comparative Genomic and Transcriptomic Analysis of *Wangiella dermatitidis*, A Major Cause of Phaeohyphomycosis and a Model Black Yeast Human Pathogen**  
*Zehua Chen, Diego A. Martinez, Sharvari Gujja, Sean M. Sykes, Qiandong Zeng, Paul J. Szaniszló, Zheng Wang, and Christina A. Cuomo*
- This report presents an analysis of the genome and transcriptional response to pH stress of the phaeohyphomycosis agent *Wangiella dermatitidis*, the most studied black fungal pathogen of humans. We find the genes responsible for producing a major fungal cell wall component are absent, yet other types of cell wall genes are highly induced by pH stress. We characterize the genomic basis of melanin production, revealing striking differences in genome organization and coordinated response to stress and light. We identify a novel horizontal transfer event of a two-gene cluster involved in sugar metabolism between algal viruses and fungi including *W. dermatitidis*.
- 579–593 **Genome-Wide Analysis of Functional and Evolutionary Features of *Tele-Enhancers***  
*Di Huang and Ivan Ovcharenko*
- Enhancers are gene regulatory elements that elevate the level of gene expression. Some distant enhancers can act over either a part or full body of an unrelated, intermediate gene. We dubbed these tele-enhancers and studied their distribution in the human genome. We show that around 60% tissue-specific enhancers belong to the class of tele-enhancers. Tele-enhancers regulate basic biological processes and are more conserved than their proximal counterparts. A sequence composition study of heart enhancers revealed that proximal and tele-enhancers employ different transcription factors for their activation, with NKX2-5 preferentially activating tele- but not proximal heart enhancers, for example.

- 595–604 **Identification of Major and Minor QTL for Ecologically Important Morphological Traits in Three-Spined Sticklebacks (*Gasterosteus aculeatus*)**  
*Jun Liu, Takahito Shikano, Tuomas Leinonen, José Manuel Cano, Meng-Hua Li, and Juha Merilä*  
 Morphological traits such as defensive armor, body shape, and body size are ecologically important in the three-spined sticklebacks (*Gasterosteus aculeatus*). Several major quantitative trait loci (QTL) have been identified for these morphological traits in Pacific three-spined sticklebacks. However, little is known about the genetic architecture of variation in these morphological traits of three-spined sticklebacks of Atlantic origin. By using a QTL mapping approach, the authors identified several major QTL and multiple minor QTL for three-spined sticklebacks of the Atlantic Ocean, which add new dimensions to the understanding of the genetic architecture of phenotypic traits in the three-spined stickleback.
- 605–612 **Horizontal Transfer and Gene Conversion as an Important Driving Force in Shaping the Landscape of Mitochondrial Introns**  
*Baojun Wu and Weilong Hao*  
 This study demonstrates unexpectedly fast turnover of the intron-encoded endonuclease within a mobile group I intron, and extensively mosaic sequences in both the intron and the endonuclease. Our results suggest that recurrent gene conversion following horizontal transfer can introduce not only intron sequence diversity but also endonuclease content variation, and ultimately promote intron mobility. As a consequence, horizontal transfer and gene conversion can play an important role in shaping the intron landscape in mitochondrial genome architecture. Our findings add another mechanical explanation on the dynamics and mobility of group I introns and intron-encoded endonucleases.
- 613–622 **Edc3 Function in Yeast and Mammals Is Modulated by Interaction with NAD-Related Compounds**  
*Robert W. Walters, Igor A. Shumilin, Je-Hyun Yoon, Wladek Minor, and Roy Parker*  
 In this work, we demonstrate that Edc3, a conserved activator of decapping and P-body formation is controlled by an apparent NADH binding pocket. As decapping and P-bodies are integral components of mRNA degradation, this links mRNA decay to the energy status of the cell in an unexpected and surprising manner. It also provides a novel line of evidence to support the emerging hypothesis that metabolic enzymes, small molecules, and RNA are interconnected in regulatory networks.
- 623–631 **Accuracy of Estimation of Genomic Breeding Values in Pigs Using Low-Density Genotypes and Imputation**  
*Yvonne M. Badke, Ronald O. Bates, Catherine W. Ernst, Justin Fix, and Juan P. Steibel*  
 This article studies genomic selection using imputed genotypes in pigs by comparing genomic predictions under two scenarios: 1) high accuracy-high cost, where training animals are genotyped in high density and prediction animals are imputed from low density genotypes with high accuracy ( $R^2=0.95$ ) and 2) low cost-low accuracy scenario with all animals genotyped in low density and imputed with lower accuracy ( $R^2=0.88$ ). Genomic evaluation using genotypes imputed with  $R^2=0.95$  was as accurate as evaluation with all animals genotyped in high density. In contrast, we observed minimal (3%), albeit significant decrease in genomic evaluation accuracy in the low imputation accuracy scenario. We also report that when all animals (training and prediction) are imputed with a small reference panel, relationships are overestimated, resulting in inflation of individual GEBV accuracies.
- 633–642 **Centromeric Barrier Disruption Leads to Mitotic Defects in *Schizosaccharomyces pombe***  
*Terilyn L. Gaither, Stephanie L. Merrett, Matthew J. Pun, and Kristin C. Scott*  
 How the cell identifies, delimits and maintains centromeric chromatin subdomains and how the unique chromatin landscape of the centromere directs accurate cell division remain critically unanswered questions. Here, we take advantage of an *S. pombe* strain bearing insertions of exogenous DNA into chromatin barriers that flank centromere 1. Using this mutant strain, we demonstrate a role for centromeric chromatin barriers in maintaining the structural integrity of centromeric chromatin domains. Mutant strains display reduced fitness, chromosome segregation defects and an increase in centromeric recombination. Our results have important implications for understanding the mechanisms that underpin centromere identity and inheritance.

- 643–656 **Disruption of the *protein kinase N* gene of *Drosophila melanogaster* Results in the Recessive *delorean* Allele (*pkn<sup>dn</sup>*) With a Negative Impact on Wing Morphogenesis**  
*Georgette L. Sass and Bruce D. Ostrow*
- We describe the *delorean* allele of the *Drosophila melanogaster* protein kinase N gene (*pkn<sup>dn</sup>*) that causes defects in wing morphology. We have determined the molecular lesion that causes the *delorean* mutation, a P-element insertion in the first intron of the gene, as well as ascertained the specific transcripts associated with the *pkn<sup>dn</sup>* allele. Genetic analysis of the wing and eye phenotypes indicates the *delorean* allele is sensitive to pairing, and provides novel information about the function of *Drosophila* PKN. The *pkn<sup>dn</sup>* allele is valuable because it is adult viable whereas all other known *pkn* alleles are embryonic or larval lethal.
- 657–667 **EMS Mutagenesis in the Pea Aphid *Acyrtosiphon pisum***  
*Denis Tagu, Gaël Le Trionnaire, Sylvie Tanguy, Jean-Pierre Gauthier, and Jean-René Huynh*
- Genes are key regulators of the behavior of cells, tissues and even whole organisms. One strategy to discover which gene controls which behavior is to impair the function of a gene and test whether the process of interest is then disrupted in the absence of this gene. Geneticists have used this strategy to interrogate the entire genome by randomly mutating genes on different chromosomes. This method played a big part in the success of genetic model organisms such as the fly *Drosophila melanogaster* and the worm *C. elegans*. However, these organisms cannot model all the diversity of behaviors that exist, and many biological questions cannot be addressed with these model organisms. For example, aphids, which are small sap-sucking insects, can switch from parthenogenesis to sexual reproduction depending on day/night cycles. Aphids with strictly identical genomes can also develop very different morphologies (a phenomenon called polyphenism). These behaviors cannot be studied using *Drosophila* or *C. elegans*. To uncover the genes regulating these processes in aphids, we developed a protocol to mutagenize the aphid genome using EMS, which induces random mutations on different chromosomes. Here, we report the efficiency of such a strategy to induce lethal mutations on the X chromosome. We also isolated the first aphid mutant line with defects in male development, and showed that this mutation is heritable from one generation to the next. Our work shows that mechanistic analysis of gene function can be performed in aphids.
- 669–679 **Sequencing, Assembling, and Correcting Draft Genomes Using Recombinant Populations**  
*Matthew W. Hahn, Simo V. Zhang, and Leonie C. Moyle*
- Current methods for genome assembly produce thousands of assembled pieces, none of which are assigned to chromosomes and many of which have errors. We show how combining genome assembly with a recombinant population can solve both of these problems, even in non-model organisms.
- 681–692 **Evidence that Natural Selection on Codon Usage in *Drosophila pseudoobscura* Varies Across Codons**  
*Richard M. Kliman*
- The genomes of most organisms show some degree of codon bias, the uneven use of synonymous codons. In many organisms, including *Drosophila*, codon bias appears to be influenced by natural selection on translation. Ongoing natural selection should predictably shift the site frequency spectrum of derived states at polymorphic sites, allowing for statistical tests of selection. The current study uses a very large data set from *Drosophila pseudoobscura* to assess the differential impact of natural selection on subsets of codons. Shifts in the spectra tend to be more pronounced when codon bias and/or codon preference is more pronounced.
- 693–706 ***unfulfilled* Interacting Genes Display Branch-Specific Roles in the Development of Mushroom Body Axons in *Drosophila melanogaster***  
*Karen E. Bates, Carl Sung, Liam Hilson, and Steven Robinow*
- The mushroom body of *Drosophila* is composed of interneurons that are required for a range of complex behaviors. The differentiation of each interneuron involves the formation of axonal branches that result in the formation of five adult specific lobes from three neuronal subtypes. The *unfulfilled* gene is required for the differentiation of all mushroom body neurons. To identify *unfulfilled* downstream genes, we developed a genetic screen to identify *unfulfilled*-interacting genes. We identified seven *unfulfilled* interacting genes. Each gene impacts only a subset of axonal branches suggesting that the differentiation of each branch is controlled by a distinct genetic network.

707–715 **Sequence Profiling of the *Saccharomyces cerevisiae* Genome Permits Deconvolution of Unique and Multialigned Reads for Variant Detection**

Claire Jubin, Alexandre Serero, Sophie Loeillet, Emmanuel Barillot, and Alain Nicolas

Genomic repetitive regions represent a challenge to the analysis of short reads produced by High Throughput Sequencing (HTS). In the mapping step, repetitions are a source of multi-alignments that prevent robust identification of the original genomic locations of sequenced reads, and finally results in these regions are spoiled by false positives. This is annoying because repetitions contain important features like multi-copy genes. Using the *S. cerevisiae* genome, we performed a comprehensive study of multi-alignments along the reference sequence and we set up an efficient multi-alignment filter. Using this filter we identified SNPs in both unique and repetitive regions.

717–731 **Extensive Cotransformation of Natural Variation into Chromosomes of Naturally Competent *Haemophilus influenzae***

Joshua Chang Mell, Jae Yun Lee, Marlo Firme, Sunita Sinha, and Rosemary J. Redfield

Recent studies have made it abundantly clear that many bacteria regularly exchange genes between lineages using homologous recombination of DNA from their environment, a situation with parallels to sexual reproduction. We have been using natural genetic variation to characterize the limits and extent of transformational recombination, a common mechanism of bacterial gene transfer, in the human pathogen *Haemophilus influenzae*. Here, we report results of sequencing 96 experimentally transformed genomes, providing estimates of the extent of natural transformation to single genomes. We also isolated an interval carrying variation affecting transformability, opening the door to quantitative trait locus studies in transformable bacteria.

733–747 **Use of an Activated Beta-Catenin to Identify Wnt Pathway Target Genes in *Caenorhabditis elegans*, Including a Subset of Collagen Genes Expressed in Late Larval Development**

Belinda M. Jackson, Patricia Abete-Luzi, Michael W. Krause, and David M. Eisenmann

In *C. elegans*, canonical Wnt signaling regulates several processes during larval development; however, few target genes are known. We conditionally activated Wnt signaling during larval life by overexpressing an activated beta-catenin protein and identified 166 Wnt regulated genes by microarray analysis. Most were upregulated, including a subset of cuticular collagen genes normally expressed in the L4 stage. Reduction of function for several genes caused phenotypes suggestive of defects in adult cuticle integrity. Therefore we have identified a large set of potential Wnt pathway targets in *C. elegans*, including a group of genes that may function in synthesis of the adult cuticle.

## MUTANT SCREEN REPORTS

749–760 **A Genetic Screen Based on *in Vivo* RNA Imaging Reveals Centrosome-Independent Mechanisms for Localizing *gurken* Transcripts in *Drosophila***

Rippe Hayashi, S. Mark Wainwright, Sophie J. Liddell, Sheena M. Pinchin, Stuart Horswell, and David Ish-Horowicz

This paper presents a novel genetic screen for maternal mutations affecting *Drosophila* oocyte patterning based on directly visualizing fluorescent *gurken* RNA localization during oogenesis. It describes a rapid method for mapping the causative mutations, and presents detailed analysis of mutants that are defective in dorsoventral axis formation, especially those affecting microtubule organization and motor activity which provide evidence of both centrosome-mediated and -independent modes of localizing *gurken*. We discuss how the breakage of radial symmetry to establish unique dorsoventral asymmetry might emerge as a consequence of microtubule self-organization.

761–767 ***Saccharomyces cerevisiae* Essential Genes with an Opi<sup>-</sup> Phenotype**

Bryan Salas-Santiago and John M. Lopes

Yeast is a model for the study of membrane synthesis. Regulation of this process has provided significant insights into the regulation of transcription of eukaryotic genes. This study was the first to use a genome-wide screen to identify essential gene mutants that may affect regulation of membrane synthesis.