of these rearrangements appear to be intra-chromosomal suggesting an overall karyotypic stability, which is consistent with that of that of modern birds. Our results support the hypothesis that the characteristically avian genome was present in the saurian ancestor and that it has remained remarkably stable in the 280 million years since. It is credible therefore to suggest that this 'avian-style' genome may be one of the key factors in the success of this extraordinarily diverse animal group.

P1: The analysis of transcriptional regulation by cohesin and its loader with semi-in vitro reconstitution methods

## Akiyama K, Bando M, Shirahige K

Research Center for Epigenetic Disease, Institute for Molecular and Cellular Biosciences, The University of Tokyo, Tokyo, Japan

Sister chromatid cohesion (SCC) is crucial to ensure accurate chromosome segregation during mitosis. The cohesin complex mediates SCC, and recent studies show cohesin and NIPBL/Mau2 complex, a loader protein required for the loading of cohesin onto chromatin, as important player in transcriptional regulation and chromatin architecture. Discoveries of mutations in subunits of cohesin and NIPBL in human developmental disorders, so-called cohesinopathies, reveal crucial roles for cohesin in development, cellular growth, and differentiation. However, it is still unclear how cohesin and its loader work in the transcriptional regulation. To reveal the complicated mechanisms played by cohesin and its loader in transcriptional regulation, we applied in vitro Pre-initiation complex (PIC) and Early Elongation Complex (EEC) assembly systems. In this system, we used the biotin-labeled DNA template, which contained 5xGAL4 DNA binding motifs, adenovirus late promoter sequence and a part of luciferase gene. After binding of activator protein, GAL4-VP16 recombinant protein, to this DNA, PIC and EEC assembly were induced by addition of the nuclear extract from HeLa cells. Each component of protein complex formed on template DNA was monitored by Western blotting. We showed that PIC factors, mediators, general transcriptional factors and RNA polII, were recruited to the template, which depended on the activator-binding. Further, we observed cohesin- and NIPBL/Mau2-binding to the template, and their recruitments also depend on the activator binding. Interestingly, cohesin seemed to get more stably bound after addition of activator. Furthermore, when we treated lysate with 5,6-dichloro-1- $\beta$ -D-ribofuranosyl-benzimidazole (DRB), a cyclin-dependent kinase (CDK) inhibitor, we found that DNA binding of NIPBL and Mau2 is dramatically enhanced. And we performed immunoprecipitation by NIPBL antibody in PIC and EEC assembly condition, and analized the interacting proteins by LC-MS/MS. As a result, NIPBL interacted with mediators only under the activator-binding condition. Taken together, we propose that cohesin-loader and cohesin together regulates step that controls activation of mediators and paused RNA polII nearby promoter.

P2: Crossing experiments reveal gamete contribution into appearance of di- and triploid hybrid frogs in *Pelophylax esculentus* population systems

Dedukh DV<sup>1</sup>, Litvinchuk SN<sup>2</sup>, Rosanov JM<sup>2</sup>, Shabanov DA<sup>3</sup>, Krasikova AK<sup>1</sup>

<sup>1</sup>Department of Biology, Saint-Petersburg State University, Saint-Petersburg, Russia; <sup>2</sup>Institute of Cytology, Russian Academy of Sciences, Saint-Petersburg, Russia; <sup>3</sup>Department of Biology, V.N. Karazin Kharkiv National University, Kharkiv, Ukraine

Speciation through hybridization is connected with appearance of interspecies hybrids which can survive and reproduce owing to changes in their gametogenesis. In animals, these changes lead to appearance of clonal animals, which for successful reproduction usually depend on parental species and lack of recombination during gamete formation. Polyploidization can resolve these problems and may lead to emergence of new species. Pelophylax esculentus complex (complex of European water frogs) represents one of the appropriate models for studying interspecies hybridization and processes of polyploidization. Hybrid nature of the P. esculentus (RL genotype, 2n=26) was confirmed after crossings of two parental species P. ridibundus (RR genotype, 2n=26) and P. lessonae (LL genotype, 2n=26). Nevertheless absence of one parental species (P. lessonae) and abundance of triploid hybrid frogs (RRL and LLR genotypes, 3n=39) in population systems at the East of Ukraine challenged us to understand how di- and triploid hybrids can appear and prosper in population systems where hybrids exist only with P. ridibundus (R-E type population system). To answer this question we performed cytogenetic analysis of tadpoles appeared after artificial crossing experiments of



diploid and triploid hybrids. Moreover, we identified karyotypes transmitted in growing oocytes of females participated in the crossings. Genome composition of mature frogs and tadpoles was established using FISH revealing interstitial (TTAGGG)<sub>n</sub> repeat sites that differed in two parental species. After crossings of six triploid hybrid females with RRL genotype and one female with LLR genotype with diploid hybrid males and triploid hybrid males with RRL genotype, tadpoles with karyotypes corresponding to P. ridibundus karyotype appeared. Lampbrush chromosomes obtained from oocytes of all triploid females participated in the crossings were represented by 13 bivalents corresponding to P. ridibundus chromosomes. Analysis of lampbrush chromosomes from oocytes of additional 11 hybrid females with RRL genome composition also revealed oocytes with 13 bivalents corresponding to P. ridibundus chromosomes. We suppose that such oocytes can overcome meiosis and form haploid gametes with P. ridibundus genome. After crossings of two pairs of diploid hybrids we obtained triploid tadpoles with RRL and LLR karyotypes. Oocytes from diploid hybrid females participated in the crossing and four additional diploid hybrid females contained 26 bivalents corresponding to P. ridibundus and P. lessonae chromosomes. Such oocytes presumably can form diploid gametes after meiotic division. One diploid female after crossing with P. ridibundus male produced both P. ridibundus and diploid P. esculentus tadpoles and had oocytes with 26 bivalents corresponding to P. ridibundus and P. lessonae chromosomes. Other six diploid hybrid females had oocytes with 13 bivalents corresponding to *P. ridibundus* chromosomes. Crossings of seven diploid males with P. ridibundus females or triploid females with RRL genome composition resulted in appearance of tadpoles with karyotypes corresponding to *P. ridibundus* karyotype. Thus diploid males most probably produced haploid gametes with P. ridibundus genome. We suggest that triploid hybrid frogs cannot reproduce independently from diploid hybrids. In studied population systems, diploid hybrid females are likely to be responsible for appearance of triploid hybrids as well as new diploid hybrids.

P3: Recombination dynamics in Bovidae

<u>Fernández J<sup>1</sup></u>, Vozdova M<sup>2</sup>, Capilla L<sup>1</sup>, Sanchéz-Guillén RA<sup>1</sup>, Sebestova H<sup>2</sup>, Frohlich J<sup>2</sup>, Robinson TJ<sup>3</sup>, Rubes J<sup>2</sup>, Ruiz-Herrera A<sup>1,4</sup>

<sup>1</sup>Genome Integrity and Instability Group, Institut de Biotecnologia i Biomedicina (IBB), Universitat Autònoma de Barcelona, Barcelona, Spain; <sup>2</sup>Central European Institute of Technology, Veterinary Research Institute, Brno, Czech Republic; <sup>3</sup>Evolutionary Genomics Group, Department of Botany and Zoology, University of Stellenbosch, Stellenbosch, South Africa; <sup>4</sup>Departament de Biologia Cel·lular, Fisiologia i Immunologia, Universitat Autònoma de Barcelona, Barcelona, Spain

Homologous chromosomes exchange genetic information through recombination during meiosis, a process that increases genetic diversity, and is fundamental to sexual reproduction. Meiotic studies in mammalian species are scarce and mainly focused on human and mouse. Therefore, our understanding of the mechanisms underlying recombination variation among mammalian species is skewed and poorly understood. In an attempt to shed light on the dynamics of mammalian meiosis and implication that these hold for genome evolution, we have established recombination maps of different bovid species by analyzing the distribution of RAD51 and MLH1 foci at the different stages of prophase I (leptonema, zygonema and pachynema). These species include representatives of Tribe Bovini (Bos taurus, Syncerus caffer caffer), Tragelaphini (Taurotragus oryx), Aepycerotini (Aepyceros melampus), Antilopini (Antidorcas marsupialis), Reduncini (Kobus leche kafuensis), Hipotragini (Oryx gazella), Alcelaphini (Dammaliscus pygargus phillipsi and Connochaetes taurinus taurinus) and Caprini (Ovis aries). Our data show that although the number of meiotic double-strand breaks per cell and recombination rates might vary between individuals, both variables were dependent on the diploid number. These results suggest that both the number of meiotic double-strand breaks initiated in the early stages of meiosis, and the final crossover events, are influenced by how the genome is organized into chromosomes.

P4: Porcine IVP: The development of a rapid sexing technique using fluorescent PCR in preimplantation porcine embryos

Gould RL<sup>1</sup>, Fowler KE<sup>1</sup>, Karamtzioti P<sup>1</sup>, Waters D<sup>1</sup>, Sadraie MS<sup>1</sup>, Summers M<sup>2</sup>, Walling GA<sup>3</sup>, Griffin DK<sup>1</sup> School of Biosciences, University of Kent, Canterbury, Kent, UK; <sup>2</sup>The Bridge Centre, London, UK; <sup>3</sup>JSR Genetics Ltd, Southburn, Driffield, East Yorkshire, UK



## 20th International Chromosome Conference (ICCXX)

50th Anniversary, University of Kent, Canterbury, 1st-4th September 2014

Darren K. Griffin • Katie E. Fowler • Peter J. I. Ellis • Dean A. Jackson

© Springer Science+Business Media Dordrecht 2015



## Dear Colleagues—Welcome to ICCXX

On behalf of the International Chromosome and Genome Society (ICGS), in September 2014 we welcomed several hundred delegates to the beautiful city of Canterbury. The programme was distinguished as always by its high scientific interest and contained ample opportunity for social interaction.

It is 50 years since Cyril Darlington first initiated the (then "Oxford") Chromosome Conferences and this

D. K. Griffin · K. E. Fowler · P. J. I. Ellis School of Biosciences, University of Kent, Canterbury CT2 7NJ, UK

Published online: 23 January 2015

D. A. Jackson Life Sciences, The University of Manchester, Carys Bannister Building, Dover St., Manchester M13 9PL, meeting was the 20th to be held. Dubbed by his biographer (Oren Solomon Harman) as "the man who invented the chromosome", Cyril Dean Darlington was born in Chorley, Lancashire in 1903. He was educated at Mercer's School, Holborn, 1912-17, St. Paul's School, 1917-20 and then, in 1920-23, came to Kent to study at Wye College, Ashford—just down the road from the conference venue. In 1923 he began an association of more than 30 years with the John Innes Institute, starting as a volunteer but later becoming head of Cytology in 1937 and Director in 1939. It was at the John Innes that he did much of his groundbreaking work on chromosomes, augmented by expeditions overseas and through collaboration with many distinguished British, American and Russian colleagues. He resigned in 1953 and accepted the Sherardian Professorship of Botany at Oxford where he took a keen interest in the Botanic Garden, creating "the Genetic Garden." He vigorously promoted the cause of teaching genetics in the University, retiring in 1971 and remaining in Oxford where he continued to study and publish prolifically until his death in 1981.

Darlington's legacy is that he was the world's leading expert on chromosomes of his time and one of the leading biological thinkers of the twentieth century. He sought to answer nature's biggest biological questions such as how species arise and how variation occurs. Often suffering rebuke, isolation, and obscurity along the way, he lived through Nazi atrocities, the Cold War, the molecular revolution, eugenics, the Lysenko controversy, the Civil Rights movement, the formation of the welfare state and the differing social views of man's place in the natural world. Darlington's work provoked him to ask questions

