MEETING REVIEW

piRNAtes of the Caribbean

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Sun, sand and science was provided in surplus during the 3rd Abcam Chromatin Structure and Function Meeting (5-8 December 2006) in Punta Cana; a primarily resort-based region on the Eastern-most tip of the Dominican Republic. Over a packed four-day schedule, the primary focus ce tred on elucidating and characterising epigenetic mechanisms in controlling eukaryotic development and gene expression. Recent developments in the emerging field of RNA silencing in relation to other epigenetic phenomena were presented, and the significance of small RNA and their targets in the regulation of modern eukaryotic genomes was discussed.

Small non-coding RNAs have been proposed to play a role in targeting of epigenetic regulatory complexes to specific genes to enable transcriptional silencing. One class of small RNAs, piRNAs, are enriched in meiotic cells and associate with Piwi proteins to form complexes (piRCs) linked to transcriptional gene silencing of genetic elements such as retrotransposons in germ line cells. Bob Kingston (Massachusetts General Hospital, Boston) outlined the possible role of Piwi-interacting RNA (piRNA) in mammalian gene silencing. By using 454 "deep" sequencing (a parallel sequencing-by-synthesis approach), oligonucleotides analysed from piRCs from rat testes extracts were found to have a number of similar characteristics; 29-30 nucleotides long, distinct in size and protein complex association to miRNAs, and found in distinct (in many cases, syntenic) genomic clusters. Although piRNAs recently were found to be generated from longer, single stranded precursor, their mechanism of action (especially in directing slicing activity of the piRC) remains elusive. However, data presented on the direct interaction of Piwi family proteins with recombinant RecQ (rRecQ; homologue of the Neurospora qde-gene implicated in silencing pathways) and linkage to H3K27 methylation strongly suggests involvement of piRNAs in specifying epigenetic inheritance.

Dicer has been proposed to play a central role in maintaining centromeric silencing in a number of organisms, including mammals. RNAi-mediated heterochromatin assembly in fission yeast requires the RNA-induced transcriptional silencing (RITS) complex; in which Chp1, Ago1 and Tas3 proteins couple with centrometrically-derived siRNA to target specific chromosome regions for assembly into heterochromatin. Analysis of Tas3 protein defective in Ago1 binding presented by Janet Partridge (Noffsinger Lab; St Jude's Research Hospital, Memphis) illustrated that processes involving RITS-dependent establishment of centromeric heterochromatin are distinct from their maintainence. Although Ago1 is able to maintained at centromeres through binding siRNA, establishment must involve binding Tas3, supporting a model where the RNAi pathway is crucial for RITS maintainence at the centromere.

Recent advances in connecting aberrant small RNA expression with cancer development suggest that miRNAs can be potential therapeutic targets. Roberta Benetti (Spanish National Cancer Center; CNIO, Madrid) with her collegue María Blasco, have made major contributions in the field of telomerases and their role in tumorogenesis and aging. Dr Benetti outlined recent advances on elucidating a role for small RNAs as an essential component for specifying maintenance of proper telometric gene expression and length. Studies on mouse ES cells and skin keratinocytes conditionally deleted for Dicer show abnormally elongated telomeres, which are subject to increased recombination. Importantly, this phenomena was found to be independent of telomere repeat factors 1 and 2 (TERF1/2) which have crtical roles in telomere length control and capping. The elongated telometric phenotype exhibited by Dicer-null cells is accompanied by concomitant increases in histone methylation and decreases in acetylation density. This change toward epigenetic chromatin silencing is supported by noticeable decreases in telomeric RNA transcripts, and strongly supports a rather unexpected role of Dicer-mediated regulation of mammalian telometric chromatin.

Regional silencing of telomeres in the absence of Dicer thus suggest that miRNAs play a role (directly or indirectly) in telomere regulation and function, and are therefore implicated in being contributive in telomeric–related disease (such as age-related cancer, liver and heart disease). Stefan Schoeftner (CNIO, Madrid) is extending this observation by screening for miRNAs regulating mammal-

ian telomeres. miRNA-induced changes in telomeric or subtelometric function are currently being assessed by transfecting miRNA library vectors (consisting of 150 predicted miRNAs) into HeLa cell lines carrying a luciferase-reporter gene in close telometric proximity, where changes in expression of the reporter gene is resultant due to a phenomena known as "telomere position effect" (TPE). Additional studies on the effect of transfected miRNAs on telemetric length and global chromatin structure are also underway. These collaborative studies are making great inroads into providing greater insight into the telomeric function of miRNAs and the dissection of their role promising for therapeutics.

The 2007 Chromatin Structure and Function meeting is scheduled for 27th-30th December in Antigua. As evidence of the biological relevance of RNA silencing mechanisms (by either posttranscriptional, transcriptional, RNAi or quelling means) in controlling gene regulation, chromosomal structure and genome defence mounts; we eagerly anticipate greater representation and discussion of the role small RNAs play in these epigenetic processes.