Chapter 11
Role of Epigenetics in Cancer Genomics

Amit Nagal
GVK Biosciences Private Limited, India

ABSTRACT

Epigenetics is the study of changes in organisms caused by modification of gene expression rather than alteration of the genetic code itself. ChIP-seq, is a method used to analyze protein interactions with DNA. It is a type of epigenetic analysis technique. Chromatin immunoprecipitation coupled with massive parallel sequencing (ChIP-seq) is gaining popularity day by day because of its clinical significance. It is a very effective tool in diagnosis of disease such as cancer. ChIP-seq is found to be very effective tool in understanding basic regulatory mechanism, cell differentiation study and studying disease processes with the decreasing cost of sequencing, ChIP-seq has become an indispensable tool for studying gene regulation and epigenetic mechanisms. The Present review explores epigenetic methods, pipeline and its role in cancer.

INTRODUCTION

Chromatin is the combination of DNA and proteins in eukaryotic cells. Genome-wide mapping of protein-DNA interactions and epigenetic marks and their modifications is essential for a full understanding of transcriptional regulation and cell differentiation. Chromatin states can influence transcription directly by altering the packing of the DNA

ChIP-seq is a technique to interpret protein interactions with DNA. Antibodies are used to select specific proteins or nucleosomes which enriches for DNA-fragments that are bound to these proteins or nucleosomes. These selected fragments can be either hybridized to a microarray (ChIP-ChIP) or sequenced on modern NGS platform (ChIP-seq).ChIP-seq combines chromatin immunoprecipitation (ChIP) with massively parallel DNA sequencing to identify the binding sites of DNA-associated protein. It was first introduced by David and Barbara (2007). However, NGS provides relatively high resolution, low noise, and high genomic coverage with compared to other technology like ChIP-ChIP assays (ChIP followed by microarray hybridization).

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BACKGROUND

In chromatin immunoprecipitation, cells are lysed and protein-DNA interactions are crosslinked to form covalent bonds by formaldehyde or other chemical reagents which explained by Solomon and Varshavsky (1998). Then the crosslinked DNA is sheared by sonication or DNA-cutting enzymes (e.g., micrococcal nuclease, often called MNase) into 150–500 bp-long fragments. Those DNA fragments crosslinked with the DNA-binding factor of interest are immunoprecipitated using an antibody specific to the factor. ChIP can be applied to a wide range of DNA binding factors, including TFs, transcription co-activators, co-repressors, chromatin regulators, and modified histones. After reverse cross-linking the protein-DNA complexes, the pulled-down DNA fragments are PCR amplified and then subjected to massively parallel sequencing it has been practiced that (Metzker, 2010, pp. 31-46) .Finally, when the resulting ChIP-seq reads are mapped back to the genome, the locations of the factor-DNA interactions can be identified.

MAIN FOCUS OF THE CHAPTER

The Role of Epigenetic in Clinical and Cancer Genomics

The importance and role of ChIP-seq was well described by various scientists. It has been found that STAT1 has DNA association. In this study ChIP-seq was used to map the signal transducers and activators of transcription 1 (STAT1) targets in interferon γ (IFNγ)-stimulated and unstimulated human cervical cancer HeLa S3 cells. By using ChIP-seq, scientists identified 41,582 and 11,004 putative STAT1-binding regions in stimulated and unstimulated cells, respectively. Out of that 34 loci known to contain STAT1 interferon-responsive binding sites, ChIP-seq found 24 (71%). ChIP-seq targets were enriched in sequences similar to known STAT1 binding motifs. This report demonstrated the high coverage and accuracy of the ChIP-seq approach. The performance of ChIP-seq was then compared to the alternative protein–DNA interaction methods of ChIP-PCR and ChIP-ChIP. The other study suggested that Yeast genes seem to have a minimal nucleosome-free promoter region of 150bp in which RNA polymerase can initiate transcription. However, ChIP-seq was used to compare conservation of TFs in the forebrain and heart tissue in embryonic mice. In this study scientist identified and validated the heart functionality of transcription enhancers, and determined that transcription enhancers for the heart are less conserved than those for the forebrain during the same developmental stage.

Several scientist recently discovered global binding maps of androgen receptor (AR) and commonly over-expressed transcriptional corepressors including histone deacetylase 1 (HDAC1), HDAC2, HDAC3, etc., in prostate cancer cells. Their results surprisingly revealed that HDACs are directly involved in androgen-regulated transcription and wired into an AR-centric transcriptional network via a spectrum of distal enhancers and/or proximal promoters. Moreover, they show that these corepressors function to mediate repression of AR-induced gene transcription that promotes epithelial differentiation and inhibits metastasis.

ChIP-seq analysis of SOX2 revealed a consensus sequence of wwTGywTT. An integrated expression profiling and ChIP-seq analysis show that SOX2 is involved in the BMP signaling pathway, steroid metabolic process, histone modifications, and many receptor-mediated signaling pathways such as IGF1R and ITPR2 (Inositol 1,4,5-triphosphate receptor, type 2).