EASL EUROPEAN ASSOCIATION FOR THE STUDY OF THE LIVER | JOURNAL OF HEPATOLOGY

Epigenetic modifications as new targets for liver disease therapies

Müjdat Zeybel, Derek A. Mann, Jelena Mann*

Institute of Cellular Medicine, Faculty of Medical Sciences, Newcastle University, Newcastle upon Tyne, UK

Summary

An important discovery from the human genome mapping project was that it is comprised of a surprisingly low number of genes, with recent estimates suggesting they are as few as 25,000 [1]. This supported an alternative hypothesis that our complexity in comparison with lower order species is likely to be determined by regulatory mechanisms operating at levels above the fundamental DNA sequences of the genome [2]. One set of mechanisms that dictate tissue and cellular complexity can be described by the overarching term "epigenetics". In the 1940s, Conrad Waddington described epigenetics as "the branch of biology which studies the causal interactions between genes and their products which bring the phenotype into being". Today we understand epigenetics as a gene regulatory system comprised of 3 major mechanisms including DNA modifications (e.g., methylation), use of histone variants and post-translational modifications of the amino acid tails of histones and non-coding RNAs of which microRNAs are the best characterized [3,4]. Together, these mechanisms orchestrate numerous sets of chemical reactions that switch parts of the genome on and off at specific times and locations.

Epigenetic marks, or the epigenome, exhibit a high degree of cellular-specificity and developmental or environmentally driven dynamic plasticity. Due to being at the interface between genome and the environment, the epigenome evolves at a very high rate compared to genetic mutations. Indeed, the differences in the epigenome account for most of the phenotypic uniqueness between closely related species, especially primates. More interestingly, the epigenetic changes, or epimutations, within an individual are not only maintained over cellular generations, but may also be transmitted between generations, such that adaptive epimutations generated in response to a particular environmental cue can influence phenotypes in our children and grandchildren [5].

Chromatin structure and non-coding RNAs

Genomic DNA in the nucleus is not present in a naked form, but rather it is associated with small proteins called histones to make a structure termed chromatin. Both DNA and histones are subject to epigenetic regulation; DNA by attachment of methyl group to cytosines (DNA methylation) within a cytosine-phospho-guanine dinucleotide (CpG) and histones by attachment of a number of chemical groups to specific amino acid residues (histone modifications) [4,6]. These two major epigenetic mechanisms will be considered in further detail in the latter sections.

Non-coding RNAs play an increasingly appreciated and crucial role in epigenetic regulation. Broadly, non-coding RNAs can be divided into long and short species, both of which play prominent but very separate roles in gene regulation. It is now known that only 1% of the mammalian genome codes for proteins, however 70–90% of the entire genome is transcribed at some point during the lifetime to produce a large transcriptome of long non-coding RNAs (lncRNAs) [7,8]. They are a class of messenger RNA-like transcripts that lack any discernible open reading frame [7]. Long ncRNAs are often poorly conserved, characterized by low copy number, fast turnover rates as well as very long length, ranging from 200 nucleotides up to 100 kilobases, sometimes encompassing a whole chromosome [9]. Due to their length, these RNA species offer the possibility of allele-specific action and targeting of a single location within the genome. This is in contrast to transcription factors or small non-coding RNAs that bind a short DNA sequence which may be repeated numerous times throughout the genome [7]. Although function of many lncRNAs is yet to be illuminated, it is now clear that at least some lncRNAs play a role in processes that lead both to gene activation and repression. This is at least in part achieved by the ability of lncRNAs to guide the catalytic function of chromatin-modifying enzymes to specific sites in the genome, and these complexes then act on chromatin modifications and structure [7,10]. Currently, there are no lncRNAs with assigned role in liver disease; however, there have been excellent examples within other disease contexts, such as MALAT1 (metastasis-associated lung adenocarcinoma transcript 1), which is a highly conserved lncRNA that regulates metastasis development in lung cancer by activating expression of metastasis-associated genes [11]. Latest studies have shown that antisense oligonucleotides can be used to silence *MALAT1* and thus prevent metastatic disease [11,12]. It will be exciting to learn the potential roles of lncRNAs in liver disease and to find out whether similar antisense silencing regimes may have a therapeutic impact.

MicroRNAs (miR) are a class of short (20–24 nucleotide) non-coding RNAs that regulate gene expression at a post-transcriptional level, through binding to the 3′-untranslated region of targeted messenger RNA and causing its degradation or destabilization; alternatively miRs can also effect translational

Keywords: Epigenetic modifications; Liver disease; Chromatin; microRNA.
Received 8 April 2013; received in revised form 23 May 2013; accepted 28 May 2013
* Corresponding author. Address: Institute of Cellular Medicine, Faculty of Medical Sciences, 4th Floor, William Leech Building, Newcastle University, Framlington Place, Newcastle upon Tyne, NE2 4HH, UK. Tel.: +44 191 222 5902; fax: +44 191 222 0723.
E-mail address: Jelena.Mann@ncl.ac.uk (J. Mann).

Open access under CC BY-NC-ND license.

repression [13]. The observed outcome is a modest change in gene expression, widely defined as a "fine-tuning effect" [14].

microRNAs and liver disease: intriguing transformation from basic science into clinics

In humans, over 1500 miRs have been identified which in turn target hundreds of genes within numerous biological pathways including liver development, regeneration, and fibrosis. miRNA profiling of distinctive liver diseases including chronic hepatitis C and B, NAFLD (nonalcoholic fatty liver disease), ALD (alcoholic liver disease), and drug-induced liver injury have revealed miRs that are now becoming novel therapeutic targets in various clinical conditions [15,16]. One of the best examples of such new developments is the discovery of miR-122 role in hepatitis C [17]. miR-122 was shown to facilitate HCV replication by binding to multiple sites in the 5′ untranslated region of HCV RNA genome and enhances translation of the viral proteins and protect HCV RNA against nucleolytic degradation. *In vivo* targeting of miR-122 was initially tested in chimpanzees where 12-week intravenous delivery of locked-nucleic-acid-[LNA]-modified complementary oligonucleotide led to depletion of miR-122 and durable reduction in HCV RNA levels as well as lower serum cholesterol levels [19]. Phase I and IIa human trials with Miravirsen (LNA-modified-antimiR-122) have also revealed good safety profiles and tolerability with prolonged reduction in HCV RNA following 18 weeks of treatment [20]. In addition, miR-122 is a mediator of insulin, lipid metabolism, and iron homeostasis. Antagonism of miR-122 reduces cholesterol and LDL levels through affecting hepatic cholesterol and fatty acid biosynthesis genes while inducing β oxidation of fatty acids as well as causing iron deficiency with lower levels of plasma and liver iron [18]. As such, antagonising miR-122 may have multiple beneficial outcomes.

While antagonism of miR-122 shows very encouraging results, two remarkable papers, using germline and liver-specific knockout mice, highlighted some unexpected roles of miR-122 in the liver. Mice with miR-122 deletions had reduced serum cholesterol levels similar to the pharmacological inhibition of miR-122, however, they developed microsteatosis with triglyceride accumulation, steatohepatitis with inflammatory cell recruitment, and elevated pro-inflammatory mediators (IL-6, TNF-α) [21]. Liver fibrosis, via the activation of KLF6 axis, and hepatocellular carcinoma-like tumours were also evident in this phenotype, indicating tumour suppressor properties of miR-122 [22]. With our current knowledge, the reason for the biological distinction between the temporary pharmacological inhibition and the global deletion of miR-122 is not yet known. However, it is likely that our current understanding of the mechanisms by which microRNAs operate in these systems is rather one-dimensional. With further research and improved insight into the ways in which various epigenetic regulatory aspects integrate with one another, we might discover means of manipulating homeostasis as well as disease states.

Histone modifications and liver disease: still on the bench

As already stated, DNA inside the nuclei is complexed with histones, forming a structure termed chromatin. The main histone proteins are called histone H2A, histone H2B, histone H3, and histone H4 [4,6]. The smallest unit of chromatin is a mononucleosome, which consists of 146 base pairs of genomic DNA

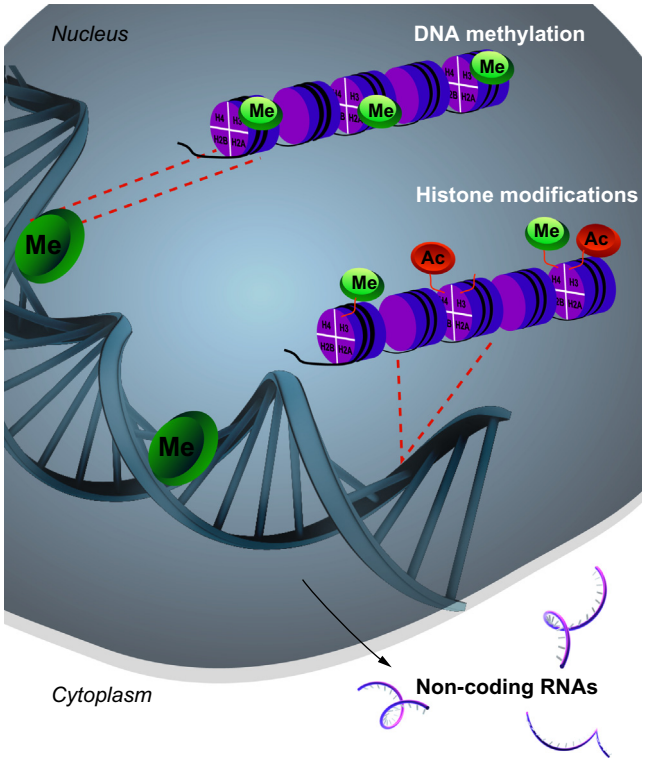

Fig. 1. Epigenetic mechanisms that regulate gene expression. The mammalian genome is packaged in the nucleus via wrapping of DNA around histones, forming a structure known as chromatin. The on and off state of gene expression is regulated by DNA methylation, post-translational modifications of various residues within histone tails, and non-coding RNAs.

sequence which is wrapped around an octamer of histones- two copies of each of the histones H2A, H2B, H3, and H4 (Fig. 1). Histones are small, globular proteins with long 'tails' on one end of the amino acid structure, this being the location of post-translational modifications carried out by numerous enzymes. There are in excess of 60 sites within each octamer of histones that are subject to chemical modification. Modifications include methylation, citrullination, acetylation, phosphorylation, SUMOylation, ubiquitination, and ADP-ribosylation. Combinations of various chemical groups are attached to the histone tails which then form binding sites for protein complexes that are able to affect transcriptional activity of a particular gene. Histone modifications are also able to induce conformational changes in chromatin such to promote transcription or silencing of gene expression in varying degrees. Both of these processes depend on the site, type, and number of chemical groups attached to the histone tails [4,6,23]. As there are strings of histone octamers at every 146 base pairs of DNA sequence along the gene, and each one can have a particular set of modifications at one of over 60 sites, the possible combinations that can be achieved are huge [4]. Each combination of histone marks potentially provides a specific recognition site for a protein complex that can alter chromatin structure. In that sense, the histone modifications provide a complex blueprint that regulates transcriptional activity, which is also referred to as the histone code [4].

In terms of a functional outcome, histone acetylation almost invariably underpins induction of gene transcription; however, methylation of lysine residues within the histone tail can either stimulate (histone 3 lysine 4) or inhibit gene expression (histone

Download English Version:

https://daneshyari.com/en/article/6104032

Download Persian Version:

https://daneshyari.com/article/6104032

Daneshyari.com