The three-dimensional architecture of chromatin: zigzag-shaped band of DNA is disposed between two layers of nucleosomes forming a chromosome

Kurchii B.A.

Institute of Plant Physiology and Genetics, 31/17 Vasylkivska St., 03022 Kyiv, Ukraine
Email: kurchii@mail.ru

Abstract
The three-layered structure of individual nucleosomes and the packaging of DNA in the form of zigzag-shaped band are presented. The nucleosomal histones are grouped into three layers: L1 (H3-H4), L2 (H2A-H2B), L3 (H1 or H5). Each this layer possesses in different functions. L1 layer is conjugated with DNA ribbon by hydrogen bonds between amino acids of nucleosomes and phosphoric acid of DNA. L2 layer glues all nucleosomes into the nucleosomal layer of the chromosome. L3 layer functioning as a protective shell for L1 and L2 histone layers. It is proposed that the eukaryotic chromosome is composed from two layers of nucleosomes, and DNA is disposed between them in the “zigzag folding” band. Three nucleosomes are arranged into one row along the long axis of the chromosome forming a nucleosomal triplet which is a basic structural subunit of the chromosome. Parental nucleosomes do not undergo disruption processes during replication and transcription.

Key words: nucleosome, chromatin, snaky model of DNA packaging, histones, nucleosomal triplet, “zigzag folding” ribbon of DNA

Introduction
It is suggested [11] that individual nucleosome is a particle of a flattened cylinder 10.5 nm in diameter and 5.7 nm in height (i.e., the nucleosome possesses in the disk-shaped form that has sizes approximately 6 nm × 11 nm × 11 nm). Because of the eukaryotic histone octamer contains five different histones H2A, H2B, H3, H4 and H1 (or H5), it makes it difficult to interpret such nucleosome positioning sequences in terms of individual histone-DNA interactions. The function of the four different core histones H2A, H2B, H3, and H4 is to package DNA into the cell nucleus but little is known about the functional role of each of them in the eukaryotic histone octamer. Packaging of DNA into the nucleosomes have negative effects on essentially all DNA transactions, including replication, recombination, repair, and transcription [9, 14, 15]. Although there is no understanding of why DNA should wrap around the nucleosome, a large number of models to explain DNA disposition on the nucleosome were proposed [see ref. 4, 5, 9, 17]. Unfortunately, we do not know the reason for the packaging of DNA by the way to wrap the nucleosome. Why each individual nucleosome is composed from different (mostly 5) proteins? There is no any logic for canonical nucleosomes to disassemble and then reassemble during chromatin functioning from a tremendous energetic cost as well. If DNA is disposed on the surface of the nucleosomes, in what way they inhibit the transcription being grabbed by DNA? Evidence suggests that the eukaryotic nucleosome is composed from the H3-H4 tetramer, two H2A-H2B dimers, and histone H1 (mostly frequently presented in eukaryal nucleosomes). Why is found H1 with the nucleosomal core, but not with DNA? By what mechanism is DNA protected from the oxidative agents if H1 forms the stronger bonds with the nucleosomal core than with DNA? Because we could not find satisfactory explanations on these and many another questions in the literature we would like to propose our model for the chromatin packaging and functioning in the eukaryotic cells.

Results and discussion
We proposed [10] that each individual nucleosome is a parallelepiped (6 nm × 11 nm × 11 nm) that consists from three layers of histones (stacked one on another): L1 (is composed from H3-H4-histones), L2 (is composed from H2A-H2B-H2A-H2B-histones), and L3 (is composed from H1 or H5 histones) (Fig. 1A) linked among themselves by hydrogen bonds (Fig. 1B). Because histone proteins of L1 layer are rich in positi-
vely charged basic amino acids they form the bonds with negatively charged phosphate groups of DNA.

Fig. 1. Proposed structure of the eukaryotic nucleosome. Three layers of a nucleosome are composed from different histones: the first layer L1 is formed from H3 and H4 histones, the second layer L2 contains H2A and H2B histones and the third layer L3 is presented by one H1 (or H5) histone and serves to safeguard L1-L2 layers and DNA (A). A general view for the complete structure of the nucleosome (B).

There are two possibilities in the formation of linkages between nucleosomes: the nucleosomes can be bonded through H2A and H2B (Fig. 2A) or H4 and H3 histones (Fig. 2B). We believe that position in Fig. 2A is more preferable. Each L1 layer in the nucleosomal triplet and each triplet are also bonded between themselves.

Because the histone H1 hinders the access of transcriptional co-activators to DNA [3, 7], acts as a general repressor of transcription [18], and at last stabilizes the chromatin structure [14] we believe that it occupies the external position in the chromosome, i.e. controversial to H3-H4 histones, which contact with DNA. Thus, both the core histone tail domains and linker histones (e.g. H1, H5) are required to form strongly condensed chromatin states.

Fig. 2. The formation of linkages between nucleosomes: the individual nucleosomes can be bonded through H3 and H4 (A) or H2A and H2B histones (B).
The milestone of this model is a notion that DNA possessing in a "zigzag folding" ribbon is disposed between two layers of nucleosomes that compose the chromosome. Naked DNA does not exist if chromosomes or nucleosomes are undamaged and can be considered as the disruption of histone-DNA interactions (e.g. during in vitro physical manipulations). We proposed that the eukaryotic chromosome is composed from two layers of nucleosomes, and the DNA is disposed between them in the "zigzag-shaped" band (Fig.3A). Three nucleosomes by the formation of bonds between L1 layers are grouped into a row (Fig.3B). Each three nucleosomes forming one row are bonded among themselves and form a column crosswise to the chromosome. We believe that linear sizes of three nucleosomes correspond to the size of DNA and RNA polymerases (especially to their length). Thus, nucleosomes perform their dual function in the eukaryotic cell, both to safeguard DNA from reactive agents and to provide regulated access to the information contained therein. In this connection we believe that bilaterality of alive essences is caused by function of "left" or "right" genes in the different chromosomes.

Fig. 3. Model for the packaging of DNA and nucleosomes into the chromosome: a general view of the chromosome (A), that are composed from two layers of nucleosomes and DNA between them, a view from en face (B). DNA is disposed onto the nucleosomal triplet (the upper row of nucleosomes is absent) in "zigzag folding" ribbon, a view of top (C).

Two fundamental chemical processes such as oxidation and reduction reactions can determine many biological processes including functioning of nucleic acids. In this connection DNA and RNA should have a reliable defense systems do not be damaged by oxidative agents and enzymes (another than DNA or RNA polymerases). Thus, nucleosomes are not only architectural unit, they protect DNA from damage, i.e. they impede to access of reactive substances (i.e. free radicals) to DNA. A side of the nucleosome, which contacts with the DNA ribbon does not contain any reactive groups. Such substances as purine, adenine, guanine, pyrimidine and cytosine having unsaturated bonds that are less reactive in comparison to 2-deoxyribose, ribose, uracil and
Thymine which possesses high reactivity because they have non-hindered functional reactive groups such as \( =\text{NH} \) or \( =\text{OH} \). DNA can be linked with NH\(_2\)-groups of amino acids, which form the nucleosome and thus the DNA-nucleosome complex can be formed by hydrogen bonds. To preserve the chaotic moving into the nucleus the chromosomes can be fastened to nucleus membranes for example through the telomer and should be liberated in the period of DNA replication.

**Fig. 4.** A model to explain the synthesis of novel chromosomes. The nucleosomal triplet of the chromosome (view in front, i.e. from the side of a promoter). Mechanism of the chromosome duplication: DNA replication machinery displaces all parental nucleosomes from the external sides of the sister chromosomes, and newly synthesized nucleosomes are displaced into “the cavity” formed by parental layers of nucleosomes.

It is noteworthy that remodeling occurs without large changes in either the conformation or the configuration of the core histone octamer [2]. Also, RSC (remodels the structure of chromatin) chromatin-remodeling complex exposes nucleosomal DNA to attack by nucleases, and this exposure occurs without loss of histones, which is paradoxical, because the DNA behaves as if it was free and bound at the same time [1]. From our model one could explain this central paradox of RSC action: the DNA in the activated state of the nucleosome is exposed along its entire length, and yet the nucleosome remains intact. As you can see from Fig. 4, 5, 6 replication and transcription processes occur without disruption of bonds between one strand of DNA and nucleosomes.

Evidence suggest that chromatin are packaged into compact 30-nm fibers which can be formed by three neighboring nucleosomes (called as a nucleosomal triplet). The nucleosome triplet being a parallelepiped-like structure and having the size 33×11×6-nm can present a basic subunit of the chromosome. The rows of these nucleosomes are assembled into a column the length of which is equal to the width of the chromosome. Each such column begins with the codes of the chromosome, the gene (cluster) and the promoter and ends by the terminator. All codes, promoters and terminators can be double strands of DNA or RNA. DNA is disposed in the “zigzag folding” band on the square of such nucleosome triplet. The ribbon of DNA (it does not bend the individual nucleosome) is situated on the plane of the nucleosome and can vary from the value of 146 bp to 260 bp in length. Hence, on the square of "such triplet of nucleosomes" can be disposed from 876 to 1560 bp of DNA. The DNA of 33-nm in length is equal approximately 97 bp or 88 bp linear molecules for the 30-nm string of bead (nucleosomes). Consequently 876 bp and 1560 bp of DNA can be arranged into 9 or 16 rows. Each row of 88 bp or 97 bp in length (the ladder DNA) may correspond to the Okazaki fragments. Than the ribbon of DNA turns back to the beginning of the nucleosomal triplet forming two rows of DNA ribbon. By the next turn on 180° starts the new cycle of the formation of two next rows (Fig.1C). The “zygzag-like” ribbon of DNA proceeds to the next edge of the chromosome and is ended by the terminator. Genes that are shorter of width of the chromosome can be completed by the intron. Also two or more genes into the column can be divided by the intron.

Thus, our model supposes that DNA remains straight in the condensed fiber as is proposed earlier for linker DNA [19], and is supported in another review [16], but this is in contradiction to the well-known solenoid model in which the linker DNA is bent [4, 6]. Nevertheless, above mentioned works are in accordance to the sterical configuration of naked DNA in the salt solutions. As follows from our model during the S-phase, both DNA and nucleosomes are duplicated concomitantly and the parental nucleosomes do not transiently disrupt during passage of the replication fork and subsequently reassemble onto the two DNA daughter strands.

The synthesis of DNA can occur by the next way. DNA is surrounded by nucleosomes from top and bottom. If there are some impediments to approach of DNA polymerases...
from button side (for example the chromosome is linked to the nuclear membrane) the top strand of DNA will be removed from native chromosome. Only DNA polymerases have contacts with DNA within the chromosome. When synthesis of DNA is finished the newly formed nucleosomes are bonded to the bottom strand of DNA from top and to the top strand of DNA from bottom. These newly synthesized structures are disposed within the chromosome (Fig.3). The parental layers of nucleosomes are completed by two identical daughter layers, each of which have one parental strand and one newly synthesized strand of DNA. Hence, mode of nucleosome formation de novo is semiconservative resembled the synthesis of DNA. In such structures, DNA is constantly safeguarded of accidental attacks by diverse reactive (i.e. free radicals) agents, including several enzymes. Also, as follows from this scenario, during DNA replication both parental strands of DNA are constantly adjacent to parental nucleosomes.

It is believed that the DNA mediated processes can function in the presence of at least two groups of chromatin remodeling enzymes. One group comprises enzymes that change the structure of the nucleosomal histones [13] leading to destabilize the folding of nucleosomal arrays and promote DNA replication or RNA transcription. Other group of enzymes disrupt histone-DNA interaction [8]. As you can see from our model histones are not disrupted during transcription or translation. Last processes also do not require any enzymes to separate DNA from histones core.

We believe that during replication, transcription and repair reactions only DNA-polymerases (their sizes are equal to the one row of DNA consisting, for example, of 97 bp of DNA) have contacts with matrix DNA within the chromosome. The formation of all RNAs occurs into two steps: firstly is synthesized DNA (a single gene surrounded by two layers of nucleosomes) which than is transformed into RNAs by RNA-polymerases within the nucleus, i.e. outside of the chromosome. Such synthesis excludes the accidental including of ribonucleotides into the matrix DNA strand (gene). Thus, the full replication fork can include about 200 bp of DNA in which leading and lagging strand consists of 100 bp of DNA.

It is therefore likely that replication do not require any codes in contrast to transcription. If transcription takes place on many genes simultaneously that are disposed within one cluster the individual code of genes can absence but individual codes of the chromosome and the cluster should be presented. So called an enhancer can present the code of the cluster.

---

**Fig. 5.** The scenario for the putative mechanism of DNA synthesis (i.e. replication process). A general view of the first row of two strands of DNA from the side of a promoter (A). A replicative fork: the upper and lower DNA (that corresponds to Okazaki fragments with a total number about 100 bp of DNA) strands of DNA are complemented by novel DNA strands (B). The new cycle of DNA synthesis in the second band continues in the way back (C).
Because the DNA wraps around the histone octamer, it forms a large number of noncovalent interactions. During the transcription only DNA-polymerases have contacts with matrix DNA within the chromosome. The formation of all RNAs occurs into two steps: firstly is synthesized DNA (a single gene/s surrounded by two layers of nucleosomes) which than is transformed into RNAs by RNA-polymerases outside of the chromosome (Fig. 6).

**Fig. 6.** Schematic representation of the possible mechanism of DNA synthesis for RNA formation (i.e. transcription). As in the case of replication only one of DNA-polymerase contacts with matrix DNA. Synthesized DNA (gene/s) for RNA formation undergoes to RNA-polymerase action outside the chromosome.

Hence, as follows from our model there is no any necessity in the sliding of the histone octamer in cis along DNA by the hSWI/SNF complex and other chromatin remodeling complexes. Moreover, recent data suggest that hSWI/SNF causes both sliding and disruption of nucleosome structure on nucleosome arrays [12].

**Conclusions**

In conclusion, proposed by us the simplified model of chromatin remodeling do not assumes the DNA wrapping around the nucleosome, condensation and decondensation of nucleosomes during replication, transcription and repair processes. DNA is stored between two layers of nucleosomes in a “zigzag folding” band. As mentioned above, nucleosomes are not structurally inert entities, but play important role in the function of chromatin in vivo. Firstly they safeguard DNA from oxidative action of diverse reactive agents and enzymes. Secondly it is important to point out that only DNA-polymerases have contacts with the native DNA within the chromosome. The formation of RNA occurs outside the chromosome where separate blocks of DNA are transformed into all type of RNA by RNA-polymerases. Such synthesis excludes the accidental including of ribonucleotides into the matrix DNA strand (gene).

Also we would like to note, however, that this is a working model which does not encompass all the known phenomena of chromatin packaging and function into the eukaryal cells. We understand that this model requires experimental verifications and hence cannot be considered as the final true, but nevertheless we hope that it could be useful for many specialists in the field of molecular biology.

**References**


Трьохвимірна архітектура хроматину: зигзагопетляюча стрічка ДНК розміщена між двома шарами нуклеосом, які утворюють хромосому

Курчій Б. О.

Інститут фізіології рослин і генетики, вул. Васильківська 31/17, 03022 Київ, Україна

Резюме Трьохшарова структура індивідуальних хромосом і упаковка ДНК в формі зигзагопетляючої стрічки описана в статті. Гістони нуклеосом згруповані в три шари: L1 (H3-H4), L2 (H2A-H2B), L3 (H1 or H5). Кожний із цих шарів має різні функції. L1 шар з’єднаний із стрічкою ДНК за допомогою водневих зв’язків між амінокислотами нуклеосом і фосфорною кислотою ДНК. L2 шар служить для з’єднування всіх нуклеосом в один шар хромосоми. L3 шар служить захисною оболонкою для L1 і L2 гістонових шарів. Запропоновано, що хромосома еукаріот складається із двох шарів нуклеосом і ДНК розміщена між ними у вигляді зигзагоподібної стрічки. Три нуклеосоми згруповані вздовж повздовжньої осі хромосоми у ряди (триплет нуклеосом), формуючи базову (елементарну) структуру хромосоми. Батьківські хромосоми не руйнуються під час реплікації і транскрипції.

Ключові слова: нуклеосома, хроматин, змійовидна модель упаковки ДНК, гістони, нуклеосомальний триплет, зигзагоподібна стрічка ДНК