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Genes and chromosomes: control of development

OLEG SEROV[1,2] **and IRINA SEROVA**[1]

[1]Instituto de Biofísica Carlos Chagas Filho, Universidade Federal do Rio de Janeiro,
Ilha do Fundão, 21949-900 Rio de Janeiro, RJ, Brasil
[2]Institute of Cytology and Genetics, Academy of Sciences of Russia, Siberian Branch
Prospect Lavrentieva, 10 – 630090 Novosibirsk, Russia

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ABSTRACT

The past decade has witnessed immense progress in research into the molecular basis behind the developmental regulation of genes. Sets of genes functioning under hierarchical control have been identified, evolutionary conserved systems of genes effecting the cell-to-cell transmission of transmembrane signals and assigned a central role in morphogenesis have been intensively studied; the concept of genomic regulatory networks coordinating expression of many genes has been introduced, to mention some of the major breakthroughs. It should be noted that the temporal and tissue-specific parameters of gene expression are correctly regulated in development only in the context of the chromosome and that they are to a great extent dependent on the position of the gene on the chromosome or the interphase nucleus. Moreover epigenetic inheritance of the gene states through successive cell generations has been conducted exclusively at the chromosome level by virtue of cell or chromosome memory. The ontogenetic memory is an inherent property of the chromosome and cis-regulation has a crucial role in its maintenance.

Key words: epigenetic regulation, developmental genes, genetic networks, variegated position effect, chromosome territories, and organization of the interphase nucleus.

INTRODUCTION

With the advent of cloning of mammals, it became increasingly clear that the eukaryotic genome is not subject to irreversible changes during differentiation and that it can faithfully reprogram to a developmental potential resemble to the original zygotic (Kikyo and Wolffe 2000, Rideout et al. 2001, Surani 2001). Furthermore, it has been demonstrated that the nuclei of highly differentiated cells, such as B or T lymphocytes, are able to accomplish a complete reprogramming, i.e., to reiterate their original potency state, even though some of the genes undergo rearrangement in the course of their differentiation (Hochedlinger and Jaenisch 2002). The list of cell types capable of reprogramming is impressive enough. It includes the fibroblasts derived from both embryos or adult animals, cumulus cells, epithelial cells of the mammary gland and oviduct, embryonic stem cells, B and T lymphocytes, immature Sertoli cells, the neural cells of the cerebral cortex of mouse embryo (Ogura et al. 2000, Yamazaki et al. 2001, Wakayama and Yanagimachi 2001, Hochedlinger and Jaenisch 2002, Miyashita et al. 2002). It remains to be determined if each and every differentiated cell is able to reprogram its genome. It is pertinent to recall that the early

Correspondence to: Dr. Oleg Serov
E-mail: serovol@hotmail.com / serov@bionet.nsc.ru

transfer experiments of the nuclei of differentiated cells into enucleated amphibian eggs or oocytes also gave unequivocal support for the view that differentiation is often not associated with irreversible genomic changes (Gurdon et al. 1979, Gurdon 1986, 1999). Taken together, the body of evidence for cloning of amphibians and mammals is consistent with the idea that differential gene activity underlies embryonic and cell differentiation, while the phenotypic diversity of the cell types in the adult organism is maintained by the acquirement of distinct epigenetic states by the genome (Latham 1999, Wolffe and Matzke 1999). It should be emphasized that both plants and animals obey this rule governing the control of development (Meyerowitz 2002), although separated by long evolutionary distances and differing by developmental patterns. It is also important to remember that plant reproduction, which involves reprogramming of specific leaf, stem or root cells, followed by the establishment of the definitive forms with full-fledged reproductive organs, is widespread among plants in nature.

THE ROLE OF GENE INTERACTION IN DEVELOPMENTAL REGULATION

The genomes of multicellular eukaryotes harbor thousands of genes. Their number amounts to about 19,000 in the nematode *C. elegans* (The *C. elegans* Sequencing Consortium Genome Sequence 1998), 13,600 in the fruit fly *Drosophila* (Adams et al. 2000), 30,000-32,000 in human (International Human Genome Sequencing Consortium 2001), and roughly 25,500 in the flowering plant *Arabidopsis thalana* (The *Arabidopsis* Genome Initiative 2000). The function of all the genes provides the development and vital activities of the definitive organism composed of specialized cells of diverse types. Thus, in man, like in most mammals, more than 200 cell types have been identified. In turn, they can be subdivided (more often by using molecular markers) into a set of more functionally or morphologically specialized cell types (Volpert et al. 1998, Surani 2001). The current paradigm for differential gene activity in development suggests that the entire phenotypic diversity of specialized somatic cells obeys the rule that only the set of expressing genes unique to a particular cell type (Lewin 1994, Volpert et al. 1998).

From comparative analysis of the mammalian genomes it followed that their gene content is similar in most of the studied species despite their striking morphological differences (O'Brien et al. 1999a, b). The genes functionally important in development are often defined as the developmental genes thereby emphasizing their prominent role in this process. These include transcriptional factors, the homeobox-containing genes, the genes coding for the transmembrane signal proteins, those responsible for the cell-to-cell transmission of inductive regulatory signals and so on. As a rule, genes of this sort are conservative through evolution and present in the genomes of vertebrate and invertebrate animals, often performing similar functions in development. From comparative gene mapping it also follows that many gene associations are preserved among disparate species. For example, the gene content of the X chromosome is similar in all the mammalian species, and more than 10 large conserved associations of syntenic genes persisting partly or completely have been uncovered (O'Brien et al. 1999a, b). It may be inferred that the ontogeny of different mammalian species relies on function of similar sets of homologous (homeologous) genes that are similarly organized at both molecular and chromosomal levels, while the wide diversity of the morphological forms in mammals suggests that species-specificity is indispensable for ontogeny.

The species-specificity for ontogeny was explained by assuming that DNA changes involving either the coding portion or the *cis*-regulatory stretches neighboring it occur in the evolving genes controlling particular stages of development (Caroll 2000, Stern 2000). These DNA differences in the homologous (homeologous) genes are responsible for the temporal and/or the tissue-specific parameters of their expression. It was implicitly assumed that such changes in gene expression are ultimately manifest as changes in particular morphogenic pro-

cesses, giving rise to the diversity of the morphological forms of plants and animals.

Development may be well considered from the point of vantage of gene expression. In such a case, it is perceived as a multistep process with the continually changing patterns of expressed genes, depending on the stage of embryonic differentiation. It is also important to take into account that a plenitude (say, hundreds, even thousands) of genes, located on distinct chromosomes, even sites within a chromosome, are involved in substitution of one pattern with another. This implies a precise coordination of the expression of gene multitudes throughout the entire development and the life of the adult (Gilbert 1991). In such a case, the application of the term developmental program is reasonable provided that it means a coordinate expression of hundreds or thousands of genes ordered at the time and in the proper space.

What underlies the developmental program? There was a time when concepts appeared somewhat sketchy. This did not mean, however, that there were no powerful tools to resolve the issue. Progress in molecular biology made the concepts more meaningful, materialized them. It is now generally believed that the developmental process relies on gene interactions, with the products of gene of preceding developmental stage activating (the gene is turned on) new gene sets in the following stages and/or repressing (the gene is turned off) single genes in the preceding. Lewin (1994) has called this gene interaction type "cascade", to stress the succession in gene expression at the earlier and later stages. There are examples of this interaction in development: the protein product of the *bicoid* gene serves as a typical morphogen to form the anterior pole of the anterior-posterior axes. This very gene acts later as a positive regulator of the first zygotic genes, one is *hunchback*, by binding to the promoter. In turn, the *hunchback* protein is a regulator of the genes of the *gap* group, repressing the expression of some genes (*Krüppel* and *knirps*) and activating that of others (*giant*). The *even-skipped* gene contributes significantly to the setting of the boundaries of the segments in *Drosophila* embryo, and its expression is regulated by the *Krüppel* and *giant* proteins (repressors), *bicoid* and *hunchback* (activators) (Lewin 1994, Volpert et al. 1998). Other examples are the coordinate hierarchial interactions between the homeobox-containing genes, the members of the *C-ANT* and *C-BX* complexes in *Drosophila* or the *HOXA, HOXB, HOXC*, and *HOXD* genes in mammals (Lewin 1994, Volpert et al. 1998).

The portion of genes executing the function of transcription factors in the eukaryotic genome is small. In *Drosophila*, their number is 700, 5% of the total gene number, of which 279 participate directly in control of development (Adams et al. 2000). It is 500, or 2%, in the nematode *C. elegans* (The *C. elegans* Sequencing Consortium 1998), and it is also 500, or 2%, in the flowering plant *Arabidopsis thaliana* (The *Arabidopsis* Genome Initiative 2000). Therefore, there are 40-50 target genes for every regulator gene. If so, then the puzzle is: how can perfect coordination be possibly achieved despite the small number of regulator genes? A timely concept permeated the literature. Topics high on the agenda became unraveled by accepting the existence of the selector genes that bind in a straightforward manner to the *cis*-regulatory elements of the target genes, thereby assuring a coordinate gene expression that ends up in the finished complex morphological structures (Guss et al. 2001). Good examples are the *eyeless, Distal-less* and *scalloped* genes. They are the major elements in the sophisticated genetic regulatory network ensuring the formation of the wing in *Drosophila*. Guss et al. (2001) provided further support by demonstrating that the *scalloped* factor, when in complex with the *vestigial* and *spalt* transcription factors of the *Decapentaplegic* transmembrane system and *cut* of the *Notch* system, control the formation of all the parts of the wing, that is to say just one selector *scalloped*, exercises control over the formation of a complex structure. It is generally accepted that a common rule plausibly underlies gene control of morphogenesis in development.

It would be appropriate to examine a large group of genes providing gene regulation by transmitting inductive transmembrane signals from one cell to another where the target gene(s) reside. This genes include those assigned a major role in morphogenic processes enfolding in vertebrate and invertebrate animals. They make up several evolutionarily conserved groups: *FGF-FGFR* (the ligand, the fibroblast growth factor and its receptor), *Delta-Notch* (the ligand, the *Delta* protein and its receptor, the *Notch* morphogen), *Wnt-Frizzled* (a complex family of protein ligands, *wingless* in insects and *Wnts* in vertebrates, and their *Frizzled* receptor), *Hedgehog-Pached* (a complex of protein ligands, *Hedgehog* in insects and *Sonic hedgehog* in vertebrates and their receptor), the family of *BMP* proteins (the morphogen proteins of bone marrow, their serine-kinase receptors) and the related beta-*TGF* proteins (the fibroblast transforming factor), *Nodal* (in vertebrates) and *Decapentaplegic* (in insects) (Volpert et al. 1998, Hogan 1999). As a rule, a transmembrane signaling system involves dozens of genes or more. Their common rule of organization (scheme) is as follows: the inductive signal (the secretion factor, the ligand) of a single cell type binds to the receptor on the cell membrane surface of target cells activated by the ligand-receptor complex (by means of protein kinases, for example) and is transported directly either to the nucleus where it represses or activates the target gene(s), or interacts intermediately with the protein and non-protein components; then, the signal reaches the target genes and, as an ultimate result, the trans-signaling system regulates the expression of a number of target genes. Such systems may be adduced as examples of how genetic regulatory networks operate (Davidson et al. 2002).

CHROMOSOMAL CONTROL OF DEVELOPMENT

The foregoing concepts of gene control of development do not exclude its control at other levels, in particular the chromosomal. There are examples demonstrating that regulation of the tissue-specific genes cannot be correct outside the chromosomal context (for review, see Bonifer 2000). This is because the regulatory sequences often extend over dozens of kilobases away from the transcription start point, and therefore the mechanism needs for their spatial approximation that is only possible when the gene is a structural element of the chromosome. In fact, Jackson et al. (1996) have demonstrated that the hypersensitive sites for DNAase in the LCR (a locus controlling region) of the beta-globin gene cluster show enhancer activity only after integration of the transgene into the genome of the transformed cells. In contrast, the activity was either not observed or reduced in transient transformed cells. An important observation was that the synergism of the H2 and H3 hypersensitive sites was manifested only in the stable transformants but not in the transient. The LCR is at a distance of more than 20 kb away from the initiating codon. This suggested that an activating effect is feasible only when the LCR and a start point of transcription are brought close together. Direct evidence for their spatial proximity came from *in situ* hybridization allowing to directly visualizing the process (Dillon et al. 1998).

There are data indicating that transcription in the chromosomes of different eukaryotic species (yeast, *Drosophila*, and mammals) is under a spatial control (Cockell and Gasser 1999, Lyko and Paro 1999). In yeast, insertion of the transgenes into the telomere region is associated with a repression of their activity, a phenomenon resembling the gene position variegation effect in *Drosophila* (Grunstein 1998). Telomeric DNA in the nucleus of the yeast cell forms a compartment in the close vicinity to the nuclear membrane where the *Sir*-proteins ("*silent information regulator*") are concentrated. When the active gene is inserted close to the telomere, the *Sir*-proteins become complexed with DNA, thereby completely silencing it. However, when the perinuclear positioning of the telomere is disturbed under the effect of mutations (the *HDF1* or *HDF2* of the *Ku* family), the telomeres cease being repressive (the "telomeric position effect"). Thus, the action of telomeric heterochromatin can become repressive

under the only condition of perinuclear localization of the telomere, i.e., close by the nuclear membrane. In their elegant experiments with targeted "anchoring" of the transgene (fused with a reporter gene) to the nuclear periphery of the yeast cell, Andrulis et al. (1998) produced a complete silencing of a nearby reporter gene. The conclusion was made that the proximity of the nuclear membrane promotes gene silencing in yeast; however, this occurs only with the participation of the *Sir*-proteins that create centers of nucleation (Andrulis et al. 1998).

There is a considerable body of evidence that the three-dimensional organization of the interphase nucleus in eukaryotes affects gene expression. The organization relies on the different positioning of different chromosomes both relative to each other and the nuclear membrane (Cockell and Gasser 1999, Misteli 2001, Parada and Misteli 2002, Gasser 2002). A characteristic feature of the nuclear architecture is chromosome occupation of distinct territories corresponding to individual chromosomes (Zink and Cremer 1998, Zink et al. 1998, Edelman et al. 2001); in turn, chromosome territories are subdivided into subchromosomal domains of about 1 Mb in size (Zink and Cremer 1998, Zink et al. 1998). Certain aspects of the internal organization of chromosome territories have been clarified. The chromosome territories are polarized in such a way that the early-replicating regions of the genome are in one compartment (nearer to the center of the nucleus), whereas the late-replicating are in other compartments (localized at the nuclear periphery, or with a perinuclear localization) (Sadoni et al. 1999). The early- and late-replicating regions correspond to the R- and G/C- bands of the mitotic chromosomes dealt with in more detail below. Interestingly, the least gene-dense human chromosome 18 is preferentially positioned towards the periphery of the nucleus, whereas the most gene-dense human chromosome 19 is positioned in the interior of the interphase nucleus (Croft et al. 1999, Cremer et al. 2001). The gene density correlated preferential positioning of the chromosomes holds true for all human chromosomes, as Boyle et al. (2001) showed later. Also, evolutionarily conserved radial positioning of chromosomes 18 and 19 is observed in all higher primates of the Old World, as Tanabe et al. (2002) reported who believed that such an evolutionary conservation of the chromosome architecture in the interphase nucleus may have far-reaching implications for genome function. Sun et al. (2000) demonstrated that the positioning of the human chromosomes is size-dependent in the interphase nucleus: the telomeres of the larger chromosomes localized at its periphery, the smaller ones nearer its center. Thus, there was a good reason for believing that the chromosomes are non-randomly organized in the interphase nucleus. More than that, the chromosome territories themselves are stable and reproducible in the daughter cells after mitosis, and the chromosome compartments themselves are structurally immobilized by associations established with the different elements of the interphase nucleus (Chubb et al. 2002, Parada and Misteli 2002).

The spatial organization of the chromosomes in the interphase nucleus has been considered as a major factor in the regulation of both single genes and the entire genome (Cockell and Gasser 1999, Lyko and Paro 1999, Parada and Misteli 2002). Examples of the spatial control for gene regulation in yeast and the genes of the beta-globin cluster in mammals were given above. As this junction, here it should be highlighted that this control operates during cell differentiation, too. Thus, in the differentiating B lymphocytes, the *CD2, CD4, CD8alpha, CD19*, and *CD45lambda5* genes translocate to the heterochromatin-containing foci and their expression is repressed as a result (Brown et al. 1997). The association of silencing genes with heterochromatin is effected through the *Ikaros* protein that specifically binds to the gene promoters and recruits them into heterochromatin (Brown et al. 1997, 1999, Cobb et al. 2000). Thus, evidence was provided indicating that the chromosomal context (the proximity to heterochromatin) and transposition of particular chromosome regions in the interphase nucleus may be, indeed, consequential for the control of gene expression.

In light of this evidence, it would be appropriate to examine the consequences of changes in gene position within a chromosome or of gene translocation to the other chromosomes on their expression. Experimental evidence for the influence of chromosomal rearrangements on gene expression was long available. Analysis of the expression of the *Pgd* (*6-phosphogluconate dehydrogenase*) gene involved in 21 chromosome rearrangements in *Drosophila* demonstrated that gene activity was lost in 2 cases, markedly decreased in 10, increased in 3 while the rearrangements were without effect in 6 cases (Slobodyanyuk and Serov 1983). The most impressive example of the consequence(s) of chromosomal rearrangements is the phenomenon of the gene position effect in which heterochromatin juxtaposed to a gene moved to a new position causes its inactivation in some but not all somatic cells. Examples of this variegated or mosaic types of expression abound in the literature. For synopses of many years of research on the influence of heterochromatin on the expression of genes in its vicinity the reader is referred to Tarlof et al. (1984), Weiler and Wakimoto (1995), Zhimulev (1998).

It has been thought for a time that the variegating position effect is restricted to the *Drosophilae* family. However, achievements in transgenic technology have made it increasingly clear that the gene position effect occurs also in other animals and plants. Injection of recombinant DNA into the zygotes is the major strategy for generating transgenic animals. With this strategy, alien DNA is randomly integrated into a host genome with a consequence that every transgenic animal is unique in terms of the chromosomal localization of the transgene (Palmiter and Brinster 1986, Chan 1999). It was an early finding that the transgene expression varies greatly: from complete absence to an expression level comparable to the endogenous genes (Palmiter and Brisnter 1986). Moreover, the transcription level of the transgenes is often independent of transgene copy number in the genome of transgenic animals. Given the random pattern of transgene integration, there was a good reason to assume that the variability in transgene expression may be determined by the chromosomal context at the integration site of the transgene. It has been long thought that the variability is exclusively dependent on the transcription level of the transgene (Palmiter and Brinster 1986, Grosveld and Kollias 1992). However, it turned out that mosaicism lies at the basis of the variability and that the transgene expression depends on the relative proportion of cells having the transgene in an active or an inactive state (Porter and Meyer 1994, Robertson et al. 1995, Festenstein et al. 1996, Dobie et al. 1996). These data were obtained for transgenic animals and plants using the *E. coli* betagalactosidase reporter or the green fluorescent protein (GFP) reporter genes under the control of constitutive or tissue-specific promoters (comprehensively reviewed in Ramirez et al. (2001).

The mosaic expression in transgenic animals and the one caused by the gene position effect in *Drosophila* have important points in which they are similar. A reason is that both obey the all-or-none rule. Interestingly, the tissue mosaicism in transgenic and chimeric mice is similar and that may indicate similarity of their temporal parameters (Morley et al. 2002). Another common property is the induction of stable silencing by a transgene in the case its integration site is a next neighbor of heterochromatin (Dobie et al. 1996, Festenstein et al. 1996). There are, however, instances in which a transgene exhibits the variegated pattern expression at a long distance from centromeric heterochromatin (Ramirez et al. 2001). This was suggestive: What if the mosaic expression of a transgene may result from local heterochromatization induced by an increase in transgene copy number? (Dorer and Henikoff 1994, Garrick et al. 1998). Confounding were instances in which a transgene was expressed in a mosaic manner in animals having it in a single copy (Zhuma et al. 1999, Ramirez et al. 2001). It should be also noted that mosaicism in the transgene expression was overcome by the presence of LCR in it (Kioussis and Festenstein 1997, Ramirez et al. 2001).

With reference to the chromosomal context, it

appeared worthwhile to consider the ectopic expression often observed for transgenes under the control of the tissue-specific promoters (Palmiter and Brinster 1986, Chan 1999). An early explanation why transgenes can be ectopically expressed was that the promoters might not contain all the sequences required for their correct tissue-specific expression. Subsequently, evidence was obtained allowing to assess the role of the integration site of the transgene in the appearance of its ectopic expression. A case in point is a retinal-binding protein-*lacZ* fusion gene (the RBP-*lacZ* reporter gene) in transgenic mice: it provides correct expression of the transgene in liver cells in 3 founders, the transgene phenotype was not expressed in a fourth; however, it was seen in segmented embryonic structures, such as somites, hindbrain rhombomeres; moreover, it was also seen in facial musculature and neocortex in adult progeny (Tan 1991). Tan (1991) suggested that alterations in the time and tissue-specificity of expression of the RBP-*lacZ* depend on the chromosome neighboring it in the vicinity of its integration site. Another illustrative case is the transgenic expression of a human keratin18/lacZ fusion gene. The reporter gene in transgenic mice was expressed in a parent-specific fashion: the correctly expressed transgene (in liver cells) was inherited from the mother, while the ectopically expressed (embryonic mesoderm, retina) was of paternal derivation (Thorey et al. 1992). It is significant that this transgene integrated into a site unrelated to endogenous imprinting site (Thorey et al. 1992). Many other examples can be cited. Particular attention deserves the observation that the reporter genes under the control of the "weak" promoters (for example, the thymidine gene of the *Herpes simples virus*) are uniquely expressed in different transgenic founders (Allen et al. 1988). This applies to both the activation time in development and the tissue-specific expression of the transgene. All in all, from the numerous data obtained with transgenic animals it may be concluded that the spatiotemporal parameters of transgene expression are under the strong influence of the chromosomal context. This prompts the appealing suggestion that chromosomal rearrangement may potentially modify the temporal and spatial parameters of the expression of genes subject to rearrangement.

Yet another aspect of chromosome control of development merits closer scrutiny-cell (Lyko and Paro 1999) or chromosome memory (Serov et al. 2003). Different differentiation-associated epigenetic events take place in development: adjustments in the length of telomeric DNA, local or global changes in DNA methylation, increase in the time of the cell cycle, among others. Features common to the chromosomes in development are an increase in the degree to which they are compacted within heterochromatin and, as Gasser (2002) believes, constraint imposed on the motion of the chromosome elements in the interphase nucleus. Chromosome compaction implies a local or a global increase in the packaging density of chromatin. The described silencing taking place under the gene position effect in *Drosophila* or occasionally in transgenic animals is also closely associated with process(es) of this kind. It is noteworthy that a change in the local organization of chromatin is inherited clonally. Lyko and Paro (1999) have defined it as cell memory, i.e., the ability to transmit the gene state through successive cell generations. The mechanisms underlying cell memory are unclear. What is known is that proteins of the *Polycomb* type and *bithorax* type can stabilize either the silenced state of chromatin or open its configuration when the cells go through mitosis and even meiosis (Lyko and Paro 1999). DNA methylation is doubtlessly another important mechanism in epigenetic inheritance, too (Lyko and Paro 1999). A similar inference can be made from studies on the epigenetic mechanisms that maintain alternative states of the transgenes in transgenic animals. For example, analysis of the expression of the transgene including the IgμH (the gene coding for a heavy chain of immunoglobulin) without LCR showed that different transgenic states are inherited under control of *cis*-acting epigenetic mechanisms (Ronai et al. 2002). The LCR deletion from the intron of the endogenous IgμH locus resulted in mosaic expression in the transformed cells, i.e., the

transgene was active in one cell population, silent in another. Both cell types were fused with myeloma cells carrying a normal expressed allele at the IgμH locus. The original state of the transgene (active or silent) persisted in the hybrid cells during numerous passages. This provided strong evidence for the different transgene states being due to the *cis*-acting regulatory mechanism(s). It should be noted that treatment of the hybrid cells with 5-azacytidine, the demethylation agent, reactivated the silent transgene. The latter indicated that the DNA methylation was responsible for silencing the transgene in the original transformed cells (Ronai et al. 2002).

It is becoming increasing clear that, in order to understand how the different epigenetic status is maintained in the chromosome, it is essential to know more about how it is *cis*-regulated. In fact, recent evidence pointed to *cis*-regulation as a major factor in this maintenance (Serov et al. 2003). In hybrid cells produced by fusion of embryonic stem cells with splenocytes from an adult animal, the homologous chromosomes were in alternative states, a pluripotent or a differentiated state. Sharing the same nucleus, the hybrid cells of the parental genomes could readily exchange *trans*-acting signals, giving opportunities to evaluate the role of *cis*- and *trans*-factors in the maintenance of the original ontogenetic differences in the homologous chromosomes. Segregation data showed that the hybrid cells preferentially lost the chromosomes of the somatic partner. This provided evidence that the original differences in the organization of the parental chromosomes were completely or partly preserved in the hybrid genome. The derivates of the former hybrid cells showed a pluripotency comparable to embryonic stem cells. The observations pointed to *cis*-regulation as a crucial factor in the maintenance of both the pluripotent and differentiated states of the parental chromosomes through the mediation of a mechanism referred to as chromosome memory (Serov et al. 2003). It this way, ontogenetic chromosome memory makes feasible the transmission of the entire set of epigenetic signs, including information on the status of all the genes, from one generation to the next. The behavior of the active and the inactive X chromosomes in mammalian development can be regarded as a particular case of the more general chromosome memory.

Future studies will yield intimation about how the chromosome inherent "ontogenetic memory" may function. The importance of the chromosomal level in the developmental regulation of gene loci is beyond question. Hopes are high in a research area rushing headlong in the past decade.

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RESUMO

Durante a última década houve imenso progresso na pesquisa sobre as bases moleculares da regulação gênica durante o desenvolvimento. Foram identificados grupos de genes funcionando sob controle hierárquico, sistemas de genes conservados ao longo da evolução atuando na transmissão célula a célula de sinais transmembrana e com uma função central na morfogênese foram intensamente estudados e o conceito de redes genômicas regulatórias coordenando a expressão de diversos genes foi introduzido, para citar apenas alguns dos principais avanços. Deve-se notar que os parâmetros tempo e tecido-específicos da expressão gênica são corretamente regulados durante o desenvolvimento apenas no contexto do cromossomo e que são amplamente dependentes da posição do gene no cromossomo ou no núcleo em interfase. Além do mais, a herança epigenética dos estados gênicos através de sucessivas gerações celulares foi conduzida exclusivamente ao nível cromossômico em função da memória celular ou cromossômica. A memória ontogenética é uma propriedade inerente do cromossomo e a regulação em *cis* tem um papel crucial em sua manutenção.

Palavras-chave: regulação epigenética, genes de desenvolvimento, redes genéticas, efeito posicional variegado, territórios cromossômicos, organização do núcleo interfásico.

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