Zeitschrift: Archives des sciences et compte rendu des séances de la Société
Herausgeber: Société de Physique et d'Histoire Naturelle de Genève
Band: 47 (1994)
Heft: 3: Archives des Sciences

Artikel: New trends in polarity. II. Nuclocentric origin of intrinsic cell polarity
Autor: Turian, Gilber / Barja, Francisco
DOI: https://doi.org/10.5169/seals-740188

Archs Sci. Genève	Vol. 47	Fasc. 3	pp. 243-254	Décembre 1994

NEW TRENDS IN POLARITY
II. NUCLEOCENTRIC ORIGIN OF INTRINSIC CELL POLARITY[1]

BY

Gilbert TURIAN & Francisco BARJA*

(Ms soumis le 7.6.1994, accepté le 5.7.1994)

ABSTRACT

New trends in polarity. II. Nucleocentric origin of intrinsic cell polarity. – Intracellular polarization is shown to find its starting point in the nucleolus, the internally polarized nuclear organite, site of DNA transcription into the RNAs. These macromolecules are then vectorially exported – as reviewed – through the nuclear matrix to the presumptive site(s) of cytoplasmic outgrowth, genetically monopolar in *Neurospora* conidial cells.

INTRODUCTION

A chain of transfer of morphogenetic information polarly starting from the nucleus has early been assumed (see Kiermayer, 1981) in which membranes of the endoplasmic reticulum (ER) receive information by a sequence DNA-RNA-ribosomes (R)-protein. Then intervenes in the cytoplasm a membrane flow from the ER and "transitorial vesicles" carry information to the membranes of the Golgi (G) system. From the G dictyosomes, special vesicles are formed that finally transfer the information to the plasma membrane where it is realized.

A. *Nucleolus*

The nucleus appears to be internally ordered and can be subdivided into DNA- and RNA-related functional domains. Such ordering is also that of the transient nucleolus-dependent ribosome biogenesis (Warner, 1990) which involves DNA transcription by RNA polymerase I (pol I) and the processing of ribosomal RNAs (Sommerville, 1986; Scheer and Benavente, 1990). Small nucleolar ribonucleoproteins (snoRNPs) appear to play a role in the processing of pre-ribosomal RNAs (pre-rRNAs) and ribosome

[1] Chapter IV. E and F (Table of contents, Turian, 1989-92)

* Laboratoire de Microbiologie générale, Sciences III, 30 quai Ernest-Ansermet, CH-1211Genève 4

assembly (Tollervey & Hurt, 1990). The most abundant snoRNA, U3 is hydrogen-bonded to pre-ribosomal RNA. Like the spliceosomal snRNAs, the snoRNAs exist as ribonucleoprotein (RNP) particles which appear to assemble into a large multi-RNA RNP complex for pre-rRNA maturation (Fournier & Maxwell, 1993).

In the nucleolus, synthesis of RNA occurs along the loops of lampbrush chromosomes according to a pattern which suggests that the DNA is being unwound at one end of the loop, spun in at the other and transcribed only when exposed in the loop. The postulated movement of the loop occurs in a *polarized* direction (Callan, 1963). It is important to trace the active rRNA genes within the nucleolar body but the results obtained to date remain somewhat controversial as well as the important question of where nucleolar transcription takes place (Jordan, 1987). Nevertheless, nucleolus organizer regions (NORs) are known to be greatly involved in nuclear polarity (Manuelidis & Borden, 1988) and there is also evidence for polarity between different NORs, and between NORs and the nuclear envelope. These points which underline the importance of nucleolar polarity are further discussed in Hernandez-Verdun (1991). Of special interest is her observation that, "in the metaphasic plate, the NOR-bearing chromosomes are closer to each other than would be expected from a random chromosomal distribution". This would indicate that there is certainly "a *polarized* arrangement of the NOR-bearing chromosomes in the interphasic nuclei".

Three morphologically distinct nucleolar components can usually be recognized by thin section electron microscopy: the fibrillar centers and the dense fibrillar and granular components (Goessens, 1984). The granular part contains the ribosomal processing intermediates (pre-rRNP) (Fakan & Puvion, 1980). According to Scheer & Benavente (1990) "there is a general consensus that ribosome biogenesis is a *vectorial* process which begins within the confines of the fibrillar components and continues into the surrounding granular component of the nucleolus". Starr & Hanover (1990) have suggested that the fibrils which extend from the nuclear surfaces of the nuclear pore complex (NPC) are involved in regulating RNA export.

The central element of the nucleolus, upon which all the rest of the nucleolar structure assembles, is the tandemly repeated ribosomal DNA that codes for the large rRNA precursors (Reeder, 1990). Electron microscopic localization of ribosomal DNA in rat liver nucleoli by nonisotopic *in situ* hybridization has been further studied and ribosomal DNA sequences found to be enriched in the dense fibrillar component of the rat liver nucleolus (Jiménez-García *et al.*, 1993).

Nucleoli are known to be sensitive to high temperatures and an heat-shock (HS) of cells provokes the intranuclear formation of "dense corpuscules" associated with the nucleolus in root tip meristems of onion and corn (Risueno *et al.*, 1973 and Fransolet *et al.*, 1979 in Nover *et al.*, 1989). Perinucleolar dense spots have also been found around the nuclei of heat-treated (46°C) macroconidia of the red mold *Neurospora crassa* (Ton-That & Turian, 1978) and their RNP composition cytochemically demonstrated (Ton-That *et al.*, 1981). Thermal retention of transportable rRNAs (ribosome subunits) has also been observed by Wunderlich *et al.* (1984) in nuclei from animal cells. Other

types of perichromatin granules and so-called "nuclear bodies", first studied in the 60's by Bernhard's group and then by Arrigo *et al.* (1980), have been "rediscovered" in animal cells by Brasch and Ochs (1992) and one class of these multifunctional organelles coiled bodies shown to be nucleolus-derived and involved in the processing or transport of both pre-mRNA and pre-rRNA.

Nucleoli can be transiently damaged by heat-shock which disrupts pre-ribosomal RNPs in someway (Simard *et al.*, 1974 in Pelham, 1984) and their export of ribosomes is blocked for several hours, in contrast with what occurs in normal conditions in which pre-ribosomes are chased out of nucleoli (Pelham, 1984). With the onset of HS the formation of mature ribosomal subunits stops immediately and completely (Scharf and Nover, 1982, 1987 in Nover *et al.*, 1989). Preribosomal ribonucleoprotein (prerRNP) processing is evidently the most heat-sentitive step of ribosome biosynthesis.

Certain heat shock proteins (HSPs) have been suggested to be present in association with cytoskeletal frameworks (Wang *et al.*, 1981). The major heat shock protein HSP70 could catalyze reassembly of damaged preribosomes and other RNPs after heat shock (Pelham, 1984) and a massive accumulation of this HSP in the granular material of the nucleolus could exert a protective effect allowing immediate recovery after the stress period (Neumann *et al.*, 1987 in Nover *et al.*, 1989).

Heat-shock produces changes in both the number and size of the granular RNP components, and finally reorganization of the nucleolar fibrillar reticulum in rat fibroblasts. Parallely, cytoplasmic actin is induced to translocate into nucleus where it forms paracrystal-like structures (Pekkala *et al.*, 1984; Welch & Suhan, 1985). These findings have be extended to the HSP90 and HSP100, both actin-binding proteins, which could cross-link intranuclear actin filaments (Koyasu *et al.*, 1986), with the expected consequence that such bundling of actin would interfere with its function in nucleocytoplasmic transport. Both these works, contrarily to Iida's *et al.* (1986) suggestions, provide a possible cue to the understanding of the cytochalasin-prevention of RNP nuclear export in heat-shocked cells (Barja & Turian, 1993, see B).

B. *Nuclear actin and RNA export*

The shift down from 46° to 25°C of heat-shocked conidia of *Neurospora crassa* provoked the migration of ^{3}H-uridine labelled RNP, presumed preribosomal components, from the nucleus to the cytoplasm paralleled by the disintegration of the dense spots (Ton-That & Turian, 1984). On the basis of a rapidly labelling technique of nuclear RNA, Nakayasu & Ueda (1985) have further suggested that nuclear actin holds nascent RNAs and thus might also play a role in processing and transport of these RNAs. A role of nuclear actin filaments in the RNA transport was further investigated by Ueyama *et al.* (1987) who used phalloidin as an inhibitor of actin filament depolymerization and found that it had no effect on the system. They concluded therefore that actin filament depolymerization may not be involved in the transport of RNA. Our recent finding of perinucleolar RNP dense spots in cytochalasin-treated

conidia (Turian *et al.*, 1992) similar to those previously found in heat-shocked conidia of *N. crassa* rather leads us now to consider that it is the polymerization of actin which is involved in this export process, F-actin tracks thus playing a guiding role[a]) for the transport of pre-ribosomal particles from the nucleoli to the conidial cytoplasm (Barja & Turian, 1993). This interpretation is supported by the suggestive microscopical observations of RNP-like particles bound to nuclear actin cables made of tightly associated microfilaments as previously observed by Gounon and Karsenti (1981). Following Singer's suggestion (1992) that mRNAs also become associated with actin filaments, Rosbash & Singer (1993) have studied the tracks from DNA to cytoplasm and the compartimentalization of certain mRNAs has been shown by Zachary & Rozengurt (1992) to occur via binding of their 3' untranslated regions to cytoskeletal microfilaments (Kislauskis & Singer, 1992).

It had been suggested by Sanger *et al.* (1980) that actin is able to translocate across the nuclear pore complex. The radius of the nuclear pore would be variable thereby enhancing transport rates (Paine *et al.*, 1975) and suggesting that it may function in the manner of a diaphragm composed of ATP-dependent contractile proteins. Among the possible functions of such nuclear contractile proteins, LeStourgeon (1978) proposed that "actin may be required for the efficient and directed transport of RNA to the cytoplasm" and derived from that an actomyosin-based pore model. A nuclear contractile protein could thus serve in the transport of precursors such as preribosomal particles and/or HnRNPs from various regions of the nucleus or nucleolus to the cytoplasm (Bremer *et al.*, 1981). Actin and myosin have further been suggested by Schindler & Jiang (1986) to participate to nucleocytoplasmic transport mechanisms. They studied such nuclear pore-mediated transport with both phalloidin and cytochalasin D which individually inhibited the ATP-dependent protein contractility. However, the nuclear contractile system includes not only actin but α-spectrin, myosin light chain kinase (MLCK) and caldesmon. Actin as well as MLKC are present not only in nuclear matrix, envelope and pores but also in nucleoli and this nuclear contractile system could be activated by nuclear calmodulin (Bachs *et al.*, 1990). As suggested by Forbes (1992), it would be of high interest to detect an actin-type of protein in the nuclear pores (first indication in Turian & Barja, 1995).

The transport out of the nucleus involves ribonucleoproteins (RNPs) and not naked RNAs. The component of the transport machinery would confer selectivity on RNA movement and act by recognition of RNA, protein or both. In this problem of transport of RNA between nucleus and cytoplasm, basic questions thus remain to be answered such as "whether export requires association of the RNA with specific proteins" (Izaurralde & Mattaj, 1992).

---

[a] As anti-actin drugs, cytochalasins might also enforce the intranuclear confinement of the RNA transcripts of retroviruses (AIDS, etc.) and thereby provide a new preventive strategy against their intercellular transmission.

C. *The role of the nuclear pore complex*

The nuclear envelope plays a dynamic role in nucleocytoplasmic transport (Stewart *et al.*, 1991). Along this pathway it implicates a relationship between nuclear pores, lamina fibers and pore-connecting fibrils. The intermediate filament proteins lamins located at the nucleoplasmic surface of the inner nuclear membrane concour to the assembly-disassembly of the nuclear lamina (Nigg, 1992).

Nucleocytoplasmic transport of either proteins (Garcia-Bustos *et al.*, 1991; Silver, 1991) or both proteins and RNA (Goldfarb & Michaud, 1991; Nigg *et al.*, 1991) has been recently reviewed and shown to be bidirectional through an asymmetric nuclear pore complex (NPC). The NPC is a 125'000 kDa machine (Reichelt *et al.*, 1990 in Goldfarb & Michaud, 1991) that transports both proteins and RNAs bidirectionally across the nuclear envelope. The two faces of NPC are not identical. This asymmetry could reflect a functional differentiation of the two NPC halves with respect to the mechanics of substrate binding (Goldfarb & Michaud, 1991), a fact which raises questions relevant to the problem of *vectorial* translocation. Both protein import and RNA export can be seen in low resolution micrographs to occur through the nuclear pore (Dworetzky & Feldherr, 1988).

Nucleocytoplasmic transport may be an important regulatory point in the control of gene expression (Davis, 1992). According to Zapp (1992), the transport of RNAs from the nucleus to the cytoplasm is an obligatory step in such gene expression and may also be a target for regulation. The cellular machinery has the capacity to export a myriad of RNA transcripts, which differ significantly in sequence and structure. Two hypothetical models for a two-step mechanism for receptor mediated RNA export have been proposed: (a) the RNA, but not the receptor is transported to the cytoplasm; (b) the RNA/receptor complex is transported to the cytoplasm. In the cytoplasm, the RNA/receptor complex dissociates and the receptor is imported back into the nucleus. However, "the actual mechanism by which the pore complex mediates RNA export remains unclear". From their recent results, Schmidt-Zachmann *et al.* (1993) rather favor the (a) proposal, declaring that "a protein does not require positively acting export signals to be transported from nucleus to cytoplasm".

The mediated transport of macromolecules across the NPC could be considered as a vectorial process using certain signals specifying transport in a cytoplasm to nuclear direction, and others in a nuclear to cytoplasmic direction (Gerace, 1992). In this respect, advances in understanding the structure of the NPC are starting to provide further information on the export of RNA from the nucleus to the cytoplasm (Forbes, 1992). In plants where nuclear targeting also occurs (Raikehl, 1992), NPC contains the major sites of bidirectional protein and nucleic acid traffic. Individual NPCs are known to carry out both import of protein (ribosomal proteins, etc.) and export of RNA. Nuclear protein import is a selective process (Silver, 1991). Certain proteins destined to the nucleus contain active nuclear localization sequences. The study of the nuclear signal sequence receptor(s) should further contribute to understand the initial steps in protein import (Forbes, 1992).

A specific premessenger ribonucleoprotein particle is translocated through the nuclear pore in the salivary glands of the dipteran *Chironomus tentans*. Its study with electron microscope tomography (Mehlin *et al.*, 1992) has led to the suggestion that a specific recognition of the RNP particle at the nuclear pore is involved in this highly ordered – and *polarized* – process.

Proteins other than ribosomal ones probably have roles not only in maturation and packaging, but in the transport of ribosomal particles even though "how they accomplish this is not clear" (Reeder, 1990). One such proteins is nucleolin which binds to nucleic acids in general and appears to shuttle between the nucleus and cytoplasm (Borer *et al.*, 1989), and is a substrate for the mitotic cdc2 kinase (Peter *et al.*, 1990). Another abundant nucleolar 38 Kd protein (NO38, B23, numatrin, nucleophosmin) has been implicated in packaging and transport of preribosomal particles (Busch, 1984; Jordan, 1987; Schmidt-Zachmann *et al.*, 1987; Borer *et al.*, 1989) and also proposed to have a role in the transport of ribosomes (Reeder, 1990). Fibrillarin is also a major protein (36 Kd), conserved from yeasts to man, present in the dense fibrillar region of nucleoli (Tollervey & Hurt, 1990), and from which depends pre-rRNA processing among major postranscriptional activities (Tollervey *et al.*, 1993).

In spite of all the above chemo-descriptive progresses, the actual mechanisms by which the pore complex mediates RNA export remain unclear. A shuttling of pre-mRNA binding proteins could also intervene between nucleus and cytoplasm as proposed by Piñol-Roma & Dreyfuss (1992). On such intracellular pathway, several nucleolar proteins have been found to be shuttled between the nucleus and the cytoplasm. These proteins have been referred as shuttling proteins because they migrate constantly back and forth between nucleus and cytoplasm (Laskey & Dingwall, 1993). Among them, nucleolin might not only function within the nucleus but might also be involved in the translocation of ribosomal components across the nuclear envelope (Borer *et al.*, 1989). A stable retention of proteins in cultured cell nuclei contrasts with the shuttling behavior of protein kinase or fragments of nucleolin (Laskey & Dingwall, 1993). The transport of RNAs from the nucleus to the cytoplasm is an obligatory step in gene expression and may also be a target for regulation (Zapp, 1992). Small nuclear ribonucleoproteins (snRNPs) have been shown to be associated with actin in the nuclear matrix, suggesting that both actin and snRNPs may be involved in the processing and transport of transcripts (Sahlas *et al.*, 1993). Prominent actin aggregates have frequently been associated with the nucleolar periphery (Milankov & De Boni, 1993).

The role of microfilaments and thus presumably actin suggest that some type of interactions occurs between the intracellular compartments, from the nucleus to the peripheral cell membranes involving protein kinase C-sensitive reversible cross-bridges between actin cytoskeleton and the plasma membrane (Aderem, 1992). Interactions indeed occur at the cytoskeleton-plasma membrane level (Luna & Hitt, 1992) and progress has recently been done in identifying specific RNA sequences that target developmentally important messages to cytoplasmic locations. It has been reported that large mRNP complexes are transported to the cytoplasm in a polar manner starting with

the 5' and finishing with the 3' end (Daneholt cited by Nigg *et al.*, 1991). This would suggest a signaling role of the mRNA's 5'm^{7}G cap.

D. *Final overview*

It is now known that specific mRNAs are concentrated in localized regions of the cytoplasm and this has led to an interest in the cytoskeleton as examplified by a possible association of actin mRNAs with actin filaments. There would thus be a stringent requirement of microfilaments in polar sorting of actin messenger RNA (Sundell & Singer, 1991) which might exit the nucleus in an asymmetric manner thereby intracellularly positioning genes for apical versus basal proteins (Singer, 1992). The question remains "how does a cell know where to send the messenger RNA" as asked by Fulton (1993) who suspects that the mRNA itself contains information to direct it to the proper cytoskeletal address.

As for the nucleofugal polarization of the cell, its "know-how" could be insured by a *vectorial* process initiated by the polarized transcription of RNAs in the nucleolus organizer region within the confines of the fibrillar zone and continued into the surrounding granular component of the nucleolus (Scheer & Benavente, 1990, see A). This vectorial process would require tracks of fibronectin fibers and actin filament bundles known to be oriented along the long axis of the cell (immunofluorescent micrographs by Hynes & Destree, 1978, in Alberts *et al.*, 1983). Of further relevance, fibronectin RNA frequently accumulates in elongated tracks localized to the nuclear interior while extending well beyond the site of transcription (Xing *et al.*, 1993) and actin has been detected not only in the nucleus and the cytoplasm of animal cells (ref. in Milankov & De Boni, 1993) but also in the fungus *Neurospora crassa* (Barja *et al.*, 1993; Turian & Barja, 1995).

This whole nucleofugally polarized process can then tentatively be illustrated with the remnants of the RNA dense spots of heat-treated conidial cells polarly exported when such cells are rescued at normal temperature (Fig. 1a). If such polar export of the RNA – presumably as preribosomal units – follows the vectorial route "fibrillar → granular zone of the nucleolus" could authorize a guess as for the localization of the ribosome-rich site of fungal germ tube outgrowth (Fig. 1b).

By the same token, nuclear actin might be considered as the mediator of such monoaxial intracellular continuity from the polarized nucleolus, through the nuclear matrix, toward proximate nuclear pores to reach cytoskeletal tracks leading to the cellular border. There, the internal driving force would first initiate the actin-rich conidial bump (Barja *et al.*, 1993) and then the elongating germ tube, with the parallel coucour of stringently required intact microtubules (Caesar-Ton That *et al.*, 1988) according to a scheme also proposed for germinating cells of *Candida albicans* by Tanaka's group (Akashi *et al.*, 1994) and sporangiospores of *Mucor rouxii* by Hasek and Bartnicki-Garcia (1994).

We are grateful to Dr. T.C. Caesar-Ton That for authorization to reproduce the two electron micrographs from her thesis (Université de Genève, 1978) and to Ariane Fehr for typing the manuscript.

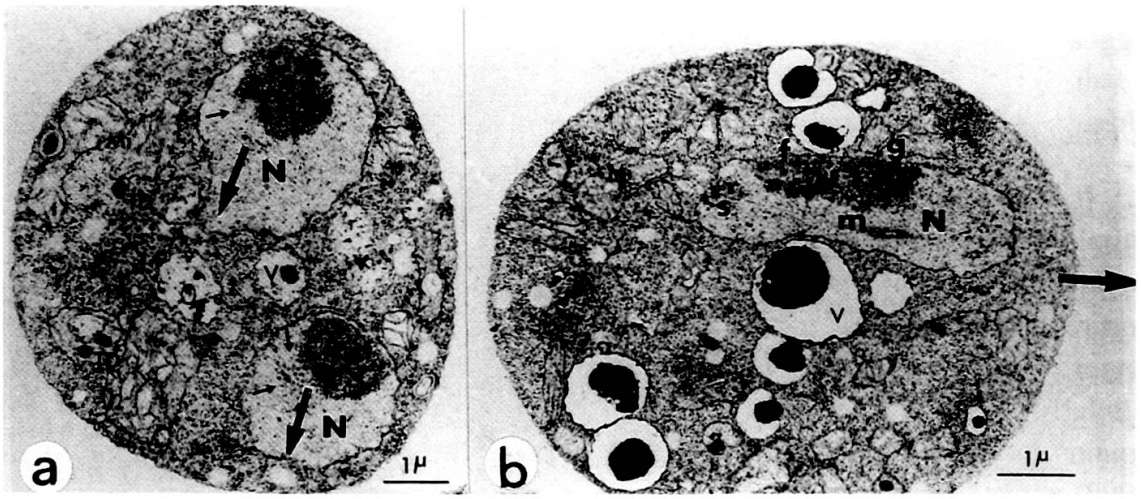

FIG. 1.

Electron micrographs of ultrathin-sectioned conidial cells of *Neurospora crassa*. a) Incubated in Vogel's medium for 10 h at 46°C, then shifted-down to 25°C for 2 h: preribosomal remnants (small arrows) of RNA dense spots polarly oriented (larger arrows) toward the nuclear (N) membrane. b) Primarily axiated after 4 h shift-down, premitotic stage of one nucleus (= N, duplicated spindle pole body = s, first spindle microtubules = m); nucleolus still complete, with its fibrillar part followed by the granular (g) zone polarly oriented toward the presumed site of germ tube outgrowth (large arrow).

REFERENCES

ADEREM, A. (1992). Signal transduction and the actin cytoskeleton: the roles of MARKS and profilin. *Trends Biol. Sci.* 17: 438-443.

AGUTTER, P.S. (1991). *Between Nucleus and Cytoplasm*. 1st ed. Chapman & Hall. London, New York. 148 pp.

AKASHI, T., T. KANBE & K. TANAKA (1994). The role of the cytoskeleton in the polarized growth of the germ tube in *Candida albicans*. *Microbiology* 140: 271-280.

ALBERTS, B., D. BRAY, J. LEWIS, M. RAFF, K. ROBERTS & J.D. WATSON (1983). *Molecular Biology of the Cell*. Garland Publ., New York, London. 1146 pp.

ARRIGO, A.P., S. FAKAN & A. TISSIERES (1980). Localization of the heat shock-induced proteins in *Drosophila melanogaster* tissue culture cells. *Dev. Biol.* 78: 86-103.

BACHS, O., L. LANINI, J. SERRATOSA, M.J. COLL, R. BASTOS, R. ALIGUE, E. RIUS & E. CARAFOLI (1990). Calmodulin-binding proteins in the nuclei of quiescent and proliferatively activated rat liver cells. *J. Biol. Chem.* 265: 18595-18600.

BARJA, F. & G. TURIAN (1993). Cytochalasin B-sensitive actin-mediated nuclear RNA export in germination conidia of *Neurospora crassa*. *Cell Biol. Intern.* 17: 985-988.

BARJA, F., M.L. CHAPPUIS & G. TURIAN (1993). Differential effects of anticytoskeletal compounds on the localization and chemical patterns of actin in germinating conidia of *Neurospora crassa*. *FEMS Microbiol, Lett.* 107: 261-266.

BORER R.A., C.F. LEHNER, H.M. EPPENBERGER & E.A NIGG (1989). Major nucleolar proteins shuttle between nucleus and cytoplasm. *Cell* 56: 379-390.

BRASCH, K. & R.L. OCHS (1992). Nuclear bodies (NBs): a newly "rediscovered" organelle. *Exp. Cell Res.* 202: 211-223.

BREMER, J.W., H. BUSCH & L.C. YEOMAN (1981). Evidence for a species of nuclear actin distinct from cytoplasmic and muscle actins. *Biochemistry* 20: 2013-2017.

BUSCH, H. (1984). Nucleolar proteins: purification, isolation and functional analyses. In *Chromosomal Nonhistone Proteins*. Vol. 4, pp. 233-286. L.S. Hnilinca (ed.). CRS Press, Boca Raton, Florida.

CAESAR-TON THAT, T.C., C. ROSSIER, F. BARJA, G. TURIAN & U.-P. ROOS (1988). Induction of multiple germ tubes in *Neurospora crassa* by antitubulin agents. *Eur. J. Cell Biol.* 46: 68-79.

CALLAN, H.G. (1963). The nature of lampbrush chromosomes. *Int. Rev. Cytol.* 15: 1-34.

CLARK, T.G. & J.L. ROSENBAUM (1979). An actin filament matrix in hand-isolated nuclei of *X. laevis* oocytes. *Cell* 18: 1101-1108.

COOPER, J.A. (1987). Effects of cytochalasin and phalloidin on actin. *J. Cell Biol.* 105: 1473-1478.

DAVIS, L.I. (1992). Control of nucleocytoplasmic transport. *Curr. Opin. Cell Biol.* 4: 424-429.

DOUVAS, A.S., C.A. HARRINGTON & J. BONNER (1975). Major nonhistone proteins of rat liver chromatin: preliminary identification of myosin, actin, tubulin, tropomyosin. *Proc. Natl. Acad. Sci. U.S.A.* 72: 3902-3906.

DWORETZKY, S.I. & C.M. FELDHERR (1988). Translocation of RNA-coated gold particles through the nuclear pores of oocytes. *J. Cell Biol.* 106: 575-584.

FAKAN, S. & E. PUVION (1980). The ultrastructural visualization of nucleolar and extranucleolar RNA synthesis and distribution. *Int. Rev. Cytol.* 65: 255-299.

FELDHERR, C. (1992). *Nuclear Trafficking*. C. Feldherr (ed.) Academic. New York. 370 pp.

FORBES, D.J. (1992). Structure and function of the nuclear pore complex. *Annu. Rev. Cell Biol.* 8: 495-527.

FOURNIER, M.J. & E.S. MAXWELL (1993). The nucleolar snRNAs: catching up with the spliceosomal snRNAs. *Trends Biol. Sci.* 18: 131-135.

FULTON, A.B. (1993). Small wonder. *The Sciences* May/June: 21-25.

GARCIA-BUSTOS, J., J. HEITMAN & M.N. HALL (1991). Nuclear protein localization. *Biochim. Biophys. Acta* 1071: 83-101.

GERACE, L. (1992). Molecular trafficking across the nuclear pore complex. *Curr. Opin. Cell Biol.* 4: 637-645.

GOESSENS, G. (1984). Nucleolar structure. *Int. Rev. Cytol.* 87: 107-158.

GOLDFARB, D. & N. MICHAUD (1991). Pathways for the nuclear transport of proteins and RNAs. *Trends Cell Biol.* 1: 20-24.

GOUNON, P. & E. KARSENTI (1981). Involvement of contractile proteins in the changes in consistency of oocyte nucleoplasm of the newt *Pleurodeles waltlii*. *J. Cell Biol.* 88: 410-421.

HADJIOLOV, A.A. (1985). *The Nucleous and Ribosome Biogenesis*. Cell Biology Monographs Vol. 12. Springer-Verlag, Wien, New York. 255 pp.

HASEK, J. & S. BARTNICKI-GARCIA (1994). The arrangement of F-actin and microtubules during germination of *Mucor rouxii* sporangiospores. *Arch. Microbiol.* 161: 363-369.

HERNANDEZ-VERDUN, D. (1991). The nucleolus today. *J. Cell Sci.* 99: 465-471.

IIDA, K., H. IIDA & I. YAHARA (1986). Heat shock induction of intranuclear actin rods in cultured mammalian cells. *Exp. Cell Res.* 165: 207-215.

IZAURRALDE, E. & I.W. MATTAJ (1992). Transport of RNA between nucleus and cytoplasm. *Semin. Cell Biol.* 3: 279-288.

JIMENEZ-GARCIA, L.F., M. DE L. SEGURA-VALDEZ, R.L. OCHS, O.M. ECHEVERRIA, G.H. VASQUEZ-NIN & H. BUSCH (1993). Electron microscopic localization of ribosomal DNA in rat liver nucleoli by nonisotopic in situ hybridization. *Exp. Cell Res.* 207: 220-225.

JOCKUSCH, B., D.F. BROWN & H.P. RUSCH (1971). Synthesis and some properties of an actin-like nuclear protein in the slime mold *Physarum polycephalum*. *J. Bacteriol.* 108: 705-714.

JORDAN, G. (1987). At the heart of the nucleolus. *Nature* 329: 489-490.

KIERMAYER, O. (1981). Cytoplasmic basis of morphogenesis in *Micrasterias*. *In Cytomorphogenesis in Plants. Cell Biol. Monogr.* Vol. 8, pp. 147-189. O. Kiermayer (ed.), Springer-Verlag, Wien, New York. 439 pp.

KISLAUSKIS, E.H. & R.H. SINGER (1992). Determinants of mRNA localization. *Curr. Opin. Cell Biol.* 4: 975-978.

KOYASU, S., E. NISHIDA, T. KADOWAKI, F. MATSUZAKI, K. IIDA, F. HARADA, M. KASUGA, H. SAKAI & I. YAHARA (1986). Two mammalian heat shok proteins, HSP90 and HSP100, are actin-binding proteins. *Proc. Natl. Acad. Sci. U.S.A.* 83: 8054-8058.

LASKEY, R.A. & C. DINGWALL (1993). Nuclear shuttling: the default pathway for nuclear proteins? *Cell* 74: 585-586.

LESTOURGEON, W.M. (1978). The occurrence of contractile proteins in nuclei and their possible functions. In *The Cell Nucleus Chromatin.* Vol. 6, pp. 305-326. H. Busch (ed.). Academic Press, New York.

LESTOURGEON, W.M., A. FORER, Y.-Z. YANG, J.S. BETRAM & H.P. RUSCH (1978). Major components of nuclear and chromosome non-histone proteins. *Biochim. Biophys. Acta* 379: 529-552.

LUNA, E.J. & A.L. HITT (1992). Cytoskeleton-plasma membrane interactions. *Science* 258: 955-964.

MANUELIDIS, L. & J. BORDEN (1988). Reproductible compartmentalitazion of individual chromosome domains in human CNS cells revealed by *in situ* hybridization and three-dimensional reconstruction. *Chromosoma* 96: 397-410.

MARTIN, K. & A. HELENIUS (1991). Nuclear transport of influenza virus ribonucleoproteins; the viral matrix protein (M1) promotes export and inhibits import. *Cell* 67: 117-130.

MEHLIN, H., B. DANEHOLT & U. SKOGLUND (1992). Translocation of a specific premessenger ribonucleoprotein particle through the nuclear pore studied with eletron microscope tomography. *Cell* 69: 605-613.

MILANKOV, K. & U. DE BONI (1993). Cytochemical localization of actin and myosin aggregates in interphase nuclei *in situ*. *Exp.Cell Res.* 209: 189-199.

NAKAYASU, H. & K. UEDA (1981). Isolation and characterization of bovine lymphocyte nuclear matrix. *Cell Struc. Funct.* 6: 181-190.

NAKAYASU, H. & K. UEDA (1985). Association of rapidly-labelled RNAs with actin in nuclear matrix from mouse L5178Y cells. *Exp. Cell Res.* 160: 319-330.

NEWMEYER, D.D. (1993). The nuclear pore complex and nucleocytoplasmic transport. *Curr. Opin. Cell Biol.* 5: 395-407.

NIGG, E.A. (1992). Assembly-disassembly of the nuclear lamina. *Curr. Opin. Cell Biol.* 4: 105-109.

NIGG, E.A., P.A. BAEURLE & R. LÜHRMANN (1991). Nuclear import-export: in search of signals and mechanisms. *Cell* 66: 15-22.

NOVER, L., D. NEUMANN & K.-D. SCHARF (eds) (1989). *Heat Shock and other Stress Response systems of Plants.* Springer-Verlag, Berlin. 155 pp.

OHMORI, H., S. TOYAMA & A. TOYAMA (1992). Direct proof that the primary site of action of cytochalasin on cell motility processes is actin. *J. Cell Biol.* 116: 933-941.

OHNISHI, T., H. KAWAMURA & T. YAMAMOTO (1963). Extraktion eines dem Aktin ähnlichen Proteins aus dem Zellkern des Kalbsthymus. *J. Biochem.* 54: 298-300.

ORNELLES, D.A., E.G. FREY & S. PENMAN (1986). Cytochalasin releases mRNA from the cytoskeletal framework and inhibits protein synthesis. *Mol. Cell. Biol.* 6: 1650-1662.

PAINE, P.L., L.C. MOORE & S.B. HOROWITZ (1975). Nuclear envelope permeability. *Nature* 254: 109-114.

PEKKALA, D., I.B. HEATH & J.C. SILVER (1984). Changes in chromatin and the phosphorylation of nuclear proteins during heat shock of *Achlya ambisexualis*. *Mol. Cell Biol.* 4: 1198-1205.

PELHAM, H.R.B. (1984). Hsp 70 accelerates the recovery of nucleolar morphology after heat shock. *EMBO J.* 3: 3095-3100.

PETER, M., J. NAKAGAWA, M. DOREE, J.C. LABBÉ & E.A. NIGG (1990). Identification of major nucleolar proteins as candidate mitotic substrates of cdc2 kinase. *Cell* 60: 791-801.

PIÑOL-ROMA, S. & G. DREYFUSS (1992). Suttling of pre-mRNA binding proteins between nucleus and cytoplasm. *Nature* 255: 730-732.

RAIKEHL, N. (1992). Nuclear targeting in plants. *Plant Physiol.* 100: 1627-1632.

RAMPAL, A.L., H.B. PINKOFSKY & C.Y. JUNG (1980). Structure of cytochalasins and cytochalasin B binding sites in human erythrocyte membranes. *Biochemistry* 19: 679-683.

REEDER, R.H. (1990). rRNA synthesis in the nucleus. *Trends Genet.* 6: 390-395.

REISLER, E. (1993). Actin molecular structure and function. *Curr. Opin. Cell Biol.* 5: 41-47.

ROSBASH, M. & R.H. SINGER (1993). RNA travel: Tracks from DNA to cytoplasm. *Cell* 75: 399-401.

SAHLAS, D.J, K. MILANKOV, P.C. PARK & U. DE BONI (1993). Distribution of snRNPs, splicing factor SC-35 and actin in interphase nuclei: immunocytochemical evidence for differential distribution during changes in functional states. *J. Cell Sci.* 105: 347-357.

SANGER, J.W., J.M. SANGER, T.E. KREIS & B.M. JOCKUSCH (1980). Reversible translocation of cytoplasmic actin into the nucleus caused by dimethylsulfoxide. *Proc. Natl. Acad. Sci. U.S.A.* 77: 5268-5272.

SCHEER, U. & R. BENAVENTE (1990). Functional and dynamic aspects of mammalian nucleolus. *BioEssays* 12: 14-21.

SCHINDLER, M. & L.-W. JIANG (1986). Nuclear actin and myosin as control elements in nucleocytoplasmic transport. *J. Cell Biol.* 102: 859-862.

SCHLIWA, M. (1982). Action of cytochalasin D on cytoskeletal networks. *J. Cell Biol.* 92: 79-91.

SCHMIDT-ZACHMANN, M.S, B. HÜGLE-DÖRR & W.W. FRANKE (1987). A constitutive nucleolar protein indentified as a member of the nucleoplasmin familiy. *EMBO J.* 6: 1881-1890.

SCHMIDT-ZACHMANN, M.S., C. DARGEMONT, L.C. KÜHN & E.A. NIGG (1993). Nuclear export of proteins: the role of nuclear retention. *Cell* 74: 493-504.

SILVER, P.A. (1991). How proteins enter the nucleus. *Cell* 64: 489-497.

SINGER, R.H. (1992). The cytoskeleton and mRNA localization. *Curr. Opin. Cell Biol.* 4: 15-19.

SOMMERVILLE, J. (1986). Nucleolar structure and ribosome biogenesis. *Trends Biol. Sci.* 11: 438-442.

STARR, C.M. & J.A. HANOVER (1990). Structure and function of the nuclear pore complex: new perspectives. *BioEssays* 12: 323-330.

STEWART, N., S. WHYTOCK & R.D. MOIR (1991). Nuclear envelope dynamics and nucleocytoplasmic transport. *J. Cell Sci. Suppl.* 14: 79-82.

SUNDELL, C.L & R.H. SINGER (1991). Requirement of microfilaments in sorting of actin messenger RNA. *Science* 253: 1275-1277.

SUZUKI, N. & K. MIHASHI (1991). Binding mode of cytochalasin B to F-actin is altered by lateral binding of regulatory proteins. *J. Biochem.* 109: 19-23.

TOLLERVEY, D & E.C. HURT (1990). The role of small nucleolar ribonucleoproteins in ribosome synthesis. *Mol. Biol. Rep.* 14: 103-106.

TOLLERVEY, D., H. LEHTONEN, R. JANSEN, H. KERN & E.C. HURT (1993). Temperature-sensitive mutations demonstrate roles for yeast fibrillarin in pre-rRNA processing, pre-rRNA methylation, and ribosome assembly. *Cell* 72: 443-457.

TON-THAT, T.C. & G. TURIAN (1978). Ultrastructural study of microcyclic macroconidiation in *Neurospora crassa*. *Arch. Microbiol.* 116: 279-288.

TON-THAT, T.C. & G. TURIAN (1984). High-resolution autoradiography of nuclear modifications during and after heat treatment of *Neurospora crassa*. *Protoplasma* 120: 165-171.

TON-THAT, T.C., G. TURIAN, J. FAKAN & A. GAUTIER (1981). Ultrastructural cytochemistry of perinucleolar dense spots in heat-treated macroconidia of *Neurospora crassa*. *Eur. J. Cell Biol.* 24: 317-319.

TURIAN, G. (1989-92). Polarity. From Dipoles to Biopolarizations. *Archs Sci. Genève* 42: 1-323. Addenda II-IV pp. 324-529.

TURIAN, G. (1994). *Polarity*. Verlag Dr. Kovac, Hamburg. 525 pp.

TURIAN, G. & BARJA, F. (1995). Nuclear actin and RNA export in conidial cells of *Neurospora crassa*. *Cell Biol. Intern.* (accepted).

TURIAN, G., F. BARJA & T.C. CAESAR-TON THAT (1992). Nucleolar dense granules in cytochalasinn-treated conidia of *Neurospora crassa*. *Cell Biol. Intern. Rep.* 16: 1265-1266.

UEYAMA, H., H. NAKAYASU & K. UEDA (1987). Nuclear actin and transport of RNA. *Cell Biol. Interrn. Rep.* 11: 671- 677.

WANG, C., R.H. GOMER & E. LAZARIDES (1981). Heat shock proteins are methylated in avian annd mammalian cells. *Proc. Natl. Acad. Sci. U.S.A.* 78: 3531-3535.

WARNER, J.R. (1990). The nucleolus and ribosome formation. *Curr. Opin. Cell Biol.* 2: 521-527.

WELCH, W.J. & J.P. SUHAN (1985). Morphological study of the mammalian stress responsse: characterization of changes in cytoplasmic organelles, cytoskeleton, and nucleoli, and appearanoce of intranuclear actin filaments in rat fibroblasts after heat-shock treatment. *J. Cell Biol.* 1011: 1198-1211.

WUNDERLICH, F., G. GIESE & V. SPETH (1984). Thermal diminution and augmentation of the retentioon of transportable rRNA in nuclear envelope-free nuclei. *Biochim. Biophys. Acta* 782: 187-194.

XING, Y., C.V. JOHNSON, P.R. DOBNER & J.B. LAWRENCE (1993). Higher level organization oof individual gene transcription and RNA splicing. *Science* 259: 1326-1330.

YANG, C.H., E.J. LAMBIE, J. HARDIN, J. CRAFT & M. SNYDER (1989). Higher order structure is present iin the yeast nucleus: autoantibody probes demonstrate that the nucleolus lies opposite the spindlle pole body. *Chromosoma* 98: 123-128.

ZACHARY, I. & E ROZENGURT (1992). Focal adhesion kinase (pl25^{FAK}): A point of convergence in thhe action of neuropeptides, integrins, and oncogenes. *Cell* 71: 891-894.

ZAPP, M.L. (1992). RNA nucleocytoplasmic transport. *Sem. Cell Biol.* 3: 289-297.