Journal of Cellular Biochemistry Supplement 35:115–122 (2000)

Mutant p53: "Gain of Function" Through Perturbation of Nuclear Structure and Function?

Wolfgang Deppert,[1,2]* Thomas Göhler,[1] Hisashi Koga,[1] and Ella Kim[1]

[1]Heinrich-Pette-Institut für Experimentelle Virologie und Immunologie an der Universität Hamburg, Germany

[2]Fachbereich Chemie der Universität Hamburg, Martinistr. 52, D-20251 Hamburg, Germany

Abstract Mutant p53 not simply is an inactivated tumor suppressor, as at least some mutant p53 proteins exhibit oncogenic properties. Mutant p53 thus is the most commonly expressed oncogene in human cancer. Accordingly, the expression of mutant p53 in tumors often correlates with bad prognosis, and expression of mutant p53 in p53-negative tumor cells enhances their transformed phenotype. The molecular basis for this "gain of function" is not yet understood. However, the finding that mutant p53 tightly associates with the nuclear matrix in vivo, and with high affinity binds to nuclear matrix attachment region (MAR) DNA in vitro, suggests that these activities are connected and may result in perturbation of nuclear structure and function in tumor cells. MAR-binding of mutant p53 most likely is due to conformation-selective DNA binding by mutant p53, i.e. the specific interaction of a given mutant p53 protein with regulatory or structural genomic DNA elements that are able to adopt specific non-B-DNA conformations. In support to this assumption, human mutant p53 ($Gly^{245} \rightarrow Ser$) was shown to bind to repetitive DNA elements in vivo that might be part of MAR elements. This further supports a model according to which mutant p53, by interacting with key structural components of the nucleus, exerts its oncogenic activities through perturbation of nuclear structure and function. J. Cell. Biochem. Suppl. 35: 115–122, 2000.

Key words: mutant p53; MAR elements; nuclear matrix; DNA-binding; chromatin remodeling; repetitive DNA elements

The Nuclear Matrix as a Target for Oncogene Action

Loss of mechanisms controlling cell growth and proliferation is a hallmark of cells undergoing neoplastic transformation. Cell growth and proliferation are complex phenomena, and so are the processes controlling it. However, in the end all changes in cellular proliferation and cell growth are reflected by changes in gene transcription, mRNA maturation and export, and DNA replication, i.e. by changes in highly organized, complex nuclear processes. It thus is not circumstantial that alterations in nuclear architecture accompany the process of tumorigenesis, and that nuclear morphology is an important diagnostic criterion in characterizing the pathology of tumors.

Nuclear architecture is built up and maintained by various nuclear structures, with the structural framework of the nucleus, the nuclear matrix, playing the most prominent role. Initially only preparatively defined as a residual, insoluble proteinaceous structure that facilitates the organization of functional components within the nucleus, the nuclear matrix, meanwhile, itself is considered as a major determinant of nuclear function. The concept of the nuclear matrix as a structural framework that

Abbreviations used: CK 2, casein kinase 2; CSB, Cockayne's syndrome B protein; DNA PK, DNA dependent protein kinase; NLS, main nuclear localization signal; RP-A, replication protein A; SV 40, Simian Virus 40; TAF, transcription activating factor; TBP, TATA-Box binding protein; TF, transcription factor; XPB, xeroderma pigmentosum B protein; XPD, xeroderma pigmentosum D protein.

Grant sponsor: Deutsche Forschungsgemein-schaft; Grant sponsor: Deutsche Krebshilfe (Dr. Mildred Scheel Stiftung); Grant sponsor: Fonds der Chemischen Industrie; Grant sponsor: The Heinrich-Pette-Institut is financially supported by the Freie und Hansestadt Hamburg and the Bundesministerium für Gesundheit.

Hisashi Koga's present address is Helix Research Institute, Inc., 1532-3 Yana, Kisarazu-shi, Chiba 292 Japan.

*Correspondence to: Wolfgang Deppert, Heinrich-Pette-Institut für Experimentelle Virologie und Immunologie, Martinistr. 52, D-20251 Hamburg, Germany.
E-mail: deppert@hpi.uni-hamburg.de

Received 21 November 2000; Accepted 29 November 2000

organizes, and at a superior level mediates and controls the complex nuclear processes of gene expression and DNA replication was developed some 25 years ago. It greatly aided in recognizing that the intranuclear distribution of nucleic acids and regulatory factors is tightly linked to architecturally highly organized multicomponent complexes that support and even mediate gene expression and DNA replication. As the multicomponent complexes are highly dynamic in architecture and composition, the nuclear "matrix" also is not a static, but a highly dynamic structure, constantly changing morphology and composition in response to the functional requirements of the cell. Dynamics in structure, composition, and function thus is an important aspect of the nuclear matrix concept.

Given the role and dynamics of the nuclear matrix in organizing and regulating nuclear processes, it seems logical to postulate that the nuclear matrix constitutes an important target for structural and functional alterations during the process of neoplastic transformation of cells. Such alterations have indeed been observed and are even used as markers for this process [reviewed in Deppert, 2000]. However, very little is known about the nature of such alterations. Specifically, it is still an open question of whether the observed alterations in nuclear matrix composition, structure, and function simply are the consequence of transformation processes, or whether such alterations may actively contribute to tumor development. The answer to this question is not trivial, as it requires molecular probes which allow the follow-up of changes in nuclear matrix structure and function, and their correlation with processes relevant to tumorigenesis.

The analysis of nuclear oncogene products, which directly interact with components of the nuclear matrix, and, thereby, may modulate nuclear functions to the requirements of a tumor cell, should provide an experimental approach to address the above question. So far, however, very little is known about such oncogenic proteins, although a number of reports have described the association of certain nuclear oncogene products with the nuclear matrix [reviewed in Deppert, 2000]. As will be outlined below, mutant *p53* appears to be one of the most prominent member of a new family of oncogenes, which exert their oncogenic functions by directly modulating nuclear structure and function.

Mutations in the *p53* Gene May Lead to a "Gain Of Function" Phenotype

The gene of the tumor suppressor p53 is mutated in about 50–60% of all human tumors, rendering it the most frequently mutated single gene in human cancer. Consequently, analysis of p53 as a tumor suppressor has attracted a large number of top scientists worldwide. While the multiple functions of wild-type p53 in maintaining genomic integrity and in protecting cells from all kinds of cellular stress are analyzed in detail, mutant p53 has drawn much less attention, despite the notion that mutant p53 may not simply be an inactivated tumor suppressor. Figure 1 shows the landmarks of p53, indicating the complexity of p53 function, both intrinsic and in cooperation with a variety of protein partners. Figure 1 also shows that p53 has an unusual mutational spectrum. Whereas other tumor suppressors are inactivated by deletions and truncations, or by promoter silencing, the vast majority of mutations in the *p53* gene are single missense point mutations, which cluster in the p53 core domain and thereby eliminate the functions of p53 as a sequence-specific transactivator and a 3′-5′ exonuclease. The mutations lead to the expression of a full length mutant p53 protein with a single amino acid substitution which accumulates in tumor cells, because it is not subjected to rapid degradation by the Mdm2 protein like wild-type p53. The unique mutational spectrum of mutant p53, and its accumulation in tumors imply that these mutations have been selected for during tumor development, and thus that the encoded mutant p53 confers an advantage to tumor cell growth and/or survival.

Very little is known about the molecular basis for the postulated oncogenic function(s) of mutant p53. The oncogenic potential of mutant p53 has become apparent from the analyses of tumor data banks, which indicated that the expression of specific mutant p53 proteins in certain tumors correlates with bad prognosis and/or resistance to chemotherapy, but direct proof for its oncogenic potential in an animal model is still lacking. However, pleiotropic oncogenic effects of mutant p53 could be demonstrated unambiguously in cell culture. There mutant p53 e.g.

(i) promoted cell growth by enhancing the proliferation rate of human Saos-2 or mouse 10(3) cells [Dittmer et al., 1993],

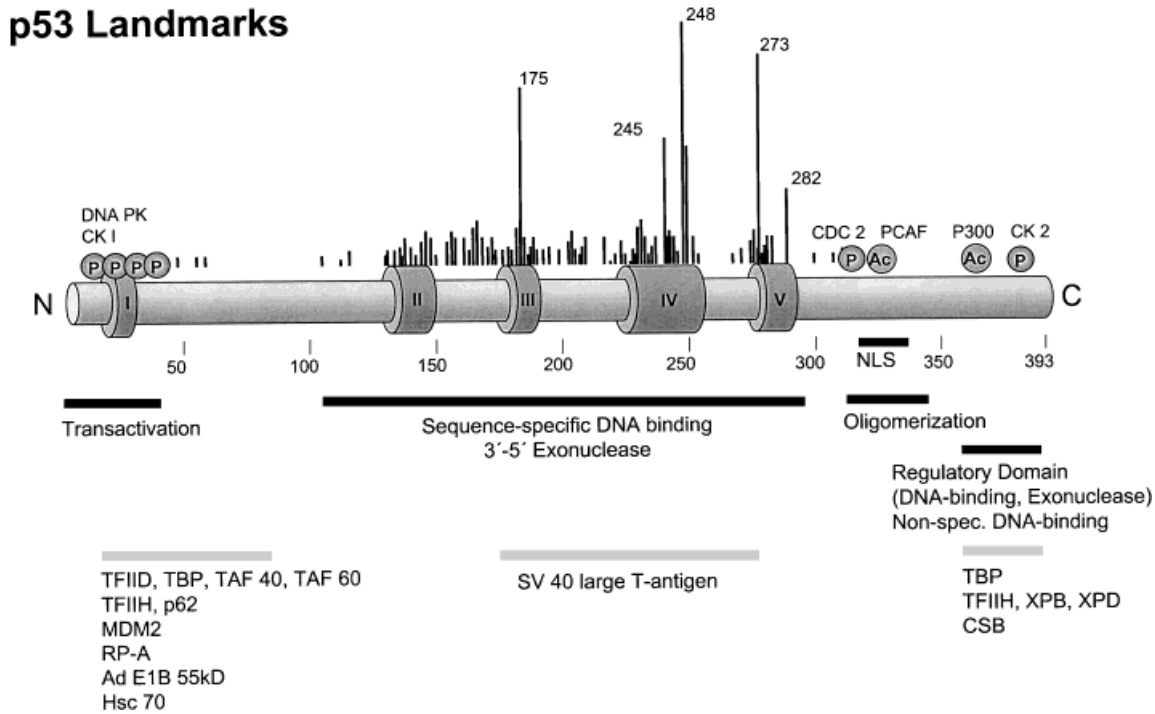

Fig. 1. *p53 Landmarks.* Roman numerals represent the five regions of p53 that are conserved within p53 from all vertebrates. Known phosphorylation (P) and acetylation (Ac) sites are indicated. The vertical bars, clustered in the center of the p53 molecule, indicate amino acid residues mutated in human tumors (hot spots are identified by amino acid number). Shown below and indicated by horizontal bars is the current information concerning various domains of p53 for biological activities, p53 DNA interactions, and p53-protein complex formation.

(ii) conferred a strongly tumorigenic phenotype to the weakly tumorigenic Abelson murine leukemia virus transformed L12 cells [Shaulsky et al., 1991],
(iii) enhanced the metastatic potential of a murine bladder carcinoma cell line [Pohl et al., 1988],
(iv) blocked non-p53-dependent apoptotic pathways in tumor cells after treatment with chemotherapeutic agents [Peled et al., 1996; Blandino et al., 1999].

Deciphering the molecular basis for the "gain of function" of mutant p53 has proven to be difficult, as the observed oncogenic effects are closely associated with the particular system under study. As an example, it has been described that mutant p53 can transcriptionally regulate a variety of genes involved in tumor progression, including the *mdr-1* gene, and the genes encoding the vascular endothelial growth factor (VGEF;) and PCNA [reviewed in Deppert, 2000]. However, detection of the transactivator function of mutant p53 strongly depends on the cellular system and the chosen experimental setup, thus often leading to conflicting result. This already indicates that transactivation by mutant p53 must occur by different mechanisms than transactivation by wild-type p53. In line with this conclusion, no DNA consensus element which could mediate sequence-specific DNA binding of mutant p53 has been identified so far. Despite all these uncertainties, mutant p53-mediated transactivation is an intrinsic property of the mutant p53 protein, as mutations in the N-terminal transactivator domain of p53 that abolish sequence-specific transactivation of wild-type p53, also abolish mutant p53-specific transactivation [Lin et al., 1995].

Mutant p53 Specifically Binds to MAR-DNA Elements In Vitro

The interactions of wild-type p53 with DNA are complex. In addition to sequence-specific DNA binding, p53 binds non-sequence-specific to single- and double- stranded DNA, to DNA ends, it binds to Holliday junctions, and it binds 3-stranded DNA mimicking early recombination intermediates [reviewed in Albrechtsen et al., 1999]. As all these interactions involve

the p53 core domain, mutations in that domain eliminate most of these interactions, with the exception of its binding to single- or double-stranded DNA in a non-sequence specific manner. As p53, additionally, is able to interact with nucleic acids via its basic C-terminal domain, mutant p53 still has the potential to interact specifically with DNA. Indeed, we found that in a variety of different assay systems human and mouse mutant p53, but not wild-type p53 specifically and with high affinity binds to nuclear matrix attachment region DNA (MAR) elements in vitro [Will and Deppert, 1998]. Depending on the binding assay, mutant p53 has a 100–1,000-fold higher affinity for MAR elements than wild-type p53, which suggests that MAR-binding is an activity specific for mutant p53, and as such is suited to form the molecular basis for its dominant oncogenic function. This idea is further supported by the finding that the majority of mutant p53 in tumor cells is tightly associated with the chromatin and the nuclear matrix [Steinmeyer, 1989].

Mutant p53 Preferentially Binds to Repetitive DNA Elements In Vivo

If the MAR-binding of mutant p53 observed in vitro bears any relevance to mutant p53 function, then mutant p53 should also bind to MAR elements in vivo. To test for an in vivo interaction of mutant p53 with MAR elements, we analyzed the in vivo binding of the human p53 "hot spot" mutant $Gly^{245} \rightarrow Ser$ [mutant p53 (Ser^{245})] to DNA in human Onda 11 glioma cells expressing high levels of mutant p53 (Ser^{245}) protein [Koga and Deppert, 2000]. After cross-linking with cisplatin, extraction of non-cross-linked proteins with high salt, and fragmentation of the cellular DNA, mutant p53-DNA complexes were immunoprecipitated [chromatin immunoprecipitation (CHIP) assay]. Then the cross-link was reversed, and the CHIP-DNA fragments were amplified and cloned. A total of 71 clones were analyzed for binding by mutant p53 (Ser^{245}) in vitro, using PCR-EMSA. Thirty-nine clones showed strong, and 15 clones weak, but definite and specific binding to mutant p53 (Ser^{245}). The specificity of the binding to the CHIP-DNA fragments was underscored by the finding that another human hot spot mutant p53, $Arg^{273} \rightarrow His$, did not bind to any of the fragments.

Sequence analysis showed that most of the CHIP-DNA fragments had a high similarity to non-coding repetitive DNA elements. Seven clones had similarity to satellite sequences (gamma: 5, alphoid: 1, satellite III sequence: 1), and seven additional clones similarity to Alu sequences. Other repetitive sequences found by the homology search were MTP, IAP, MMB, and 1MER1B. Five clones contained 12 to 28 CA dinucleotide repeats, although we could not establish a homology to known (CA)n repeats. In summary, the CHIP-DNA fragments identified as possible in vivo targets for mutant p53 in Onda 11 cells did not show any common sequence homology, rendering it extremely unlikely that their specific binding by mutant p53 (Ser^{245}) is mediated by a common sequence element.

Conformation-Selective DNA-Binding of Mutant p53

Though specific, the interaction of mutant p53 with MAR DNA in vitro, and the binding of mutant p53 to repetitive DNA, both in vivo and in vitro, must be fundamentally different from the known sequence-specific DNA binding of wild-type p53. Several lines of evidence support the assumption that mutant p53 recognizes DNA structure, i.e. DNA conformation, rather than DNA sequence:

(i) Mutant p53 specifically interacts with oligonucleotides containing variations of an AATATATTT "unwinding motif", present in certain MAR elements and implicated in MAR function [Will et al., 1998]. Such motifs, also called "base unpairing regions" (BUR) promote structural alterations within the chromatin, including regional base-unpairing [Bode et al., 1992]. Due to their high AT-content, such motifs will not adopt a stable B-DNA conformation, but will form non-B-DNA structures, especially under conditions of superhelical stress.

(ii) Using model oligonucleotide substrates designed to mimick DNA in various non-B-DNA conformations [Kim et al., 1997], we could demonstrate that e.g. mutant p53 (Ser^{245}) is able to bind certain non-B-DNA structures with high affinity (Göhler, T.; Koga, H., Deppert, W., Kim, E., unpublished). Interestingly, mutant p53 (His^{273}) failed to bind to such oligonucleotides. Especially the latter finding supports the concept that the binding of mutant p53 (Ser^{245}) to repetitive DNA elements in vivo and in vitro indeed reflects its interaction

with non-B-DNA, since mutant p53 (His^{273}) also had failed to bind to such elements [Koga and Deppert, 2000].

The data suggest that MAR-binding of mutant p53 most likely is mediated by the binding of mutant p53 to elements within MARs that are able to adopt a non-B-DNA conformation. As MARs contain multiple repetitive DNA elements which are able to form non-B-DNA structures, the possibility exists that several mutant p53 molecules can bind to a single MAR, an assumption that could explain the high affinity of MAR-binding by mutant p53 ($K_D \sim 10^{-10}$ M). Importantly, however, mutant p53 not simply binds to any non-B-DNA structure provided, but selectively to DNA adopting specific, but so far yet undefined non-B-DNA structures (Göhler, T.; Koga, H., Deppert, W., Kim, E., unpublished). We, therefore, have termed this interaction of mutant p53 with DNA *"conformation-selective DNA binding"*. An interesting possibility which has to be further analyzed is that different mutants might differ in their recognition of different non-B-DNA structures. Thus the failure of mutant p53 (His^{273}) to bind to the non-B-DNA oligonucleotides or the repetitive DNA elements recognized by mutant p53 (Ser^{245}) not necessarily implies that mutant p53 (His^{273}) is defective in conformation-selective DNA binding. Rather it might indicate that mutant p53 (His^{273}) binds to different non-B-DNA structures than mutant p53 (Ser^{245}). In fact, our previous findings that mutant p53 (His^{273}) is able to bind to MAR elements supports this assumption. This raises the interesting possibility that the different biological effects ascribed to different mutant p53 proteins may result from their ability to specifically recognize and discriminate between different non-B-DNA structures in MAR elements.

Functional Implications of MAR-DNA Binding by Mutant p53: A Model

MAR elements organize the cellular chromatin into topologically independent loops, thereby providing a structural basis for the independent spatial and temporal regulation of gene expression and initiation of DNA synthesis. It is thought that such a higher order regulatory mechanism is involved in the control of development and differentiation [Herbomel, 1990; Berezney, 1991], i.e. in processes that are grossly disturbed in tumors.

Several possibilities can be envisioned for the functional interaction of mutant p53 with MAR elements. The binding to distinct non-B-DNA structures in MAR elements provides mutant p53 with the potential to modulate gene expression and DNA replication in a positive as well as in a negative way. Initiation of transcription, and initiation of DNA replication both require an open DNA structure for the interaction of transcription/replication factors with DNA. By stabilizing such DNA structures, mutant p53 could mediate the formation of transcription and/or replication complexes at those structures. Formation of such complexes will be further aided by the ability of mutant p53 to recruit a variety of factors required for transcription and replication via its N-terminal transactivation domain. Another important property of mutant p53 relevant to these processes is its ability to tightly associate with the nuclear matrix. Thus mutant p53 could target facultative MAR elements to the nuclear matrix, thereby putting them into a functional state by creating "active" chromatin domains.

Alternatively, or in addition, mutant p53 might promote the dissolution of the non-B-DNA conformation in a bound regulatory element, as mutant p53 binds and activates topoisomerase II [Albor et al., 1998], and thereby could locally reduce the local superhelical density required for maintaining the non-B-DNA structure within a regulatory element. This in turn could prevent the association of the factors that positively or negatively regulate ordered transcription or replication in normal cells, thereby perturbing these processes.

The above model can account for the observation that the specific effects of mutant p53 in transcription (and possibly also in replication) are cell-specific. As the promoter/enhancer elements for a given gene are identical within all cells of a species, cell-type specific gene expression is controlled both by the availability of the appropriate protein factors and by an appropriate chromatin structure. A similar control has to be postulated for the temporal regulation of initiation of DNA replication. The fact that mutant p53 is able to specifically interact with both key players that regulate transcription and replication processes, the nuclear matrix and the MAR elements, thus provides the basis to develop a model for the cell-type specific modulation of gene expression and

replication by mutant p53: accordingly, mutant p53 modulated transcription and replication will be determined firstly by the recognition of regulatory elements within MAR elements. Recognition is determined by sequence and DNA conformation of the regulatory element, the surrounding chromatin structure, and the superhelicity of the DNA. Next, the accessibility of appropriate "docking" surfaces for the association of mutant p53 with the nuclear matrix, and, finally, the ability of mutant p53 to recruit the necessary factors to initiate transcription, replication, or recombination all are requirements for mutant p53 activity. The strong dependence of mutant p53 induced biological effects on such a multitude of parameters not only can explain the cell-type specificity, but also the pleiotropy of mutant p53 effects. Last not least, "fixation" of certain chromatin domains by mutant p53 bound to MAR elements through the tight interaction of mutant p53 both with nuclear matrix proteins and with MAR elements could interfere with differentiation processes, as gene expression, replication, and differentiation are coupled via chromatin organization. In line with this model, it has been described that mutant p53 interferes with differentiation processes [Shaulsky et al., 1991]. Table 1 summarizes the pleiotropic "gain of function" properties of mutant p53 and their possible relation to mutant p53 MAR-binding.

Functional Implications for the Binding of Mutant p53 (Ser^{245}) to Repetitive DNA Elements In Vivo

Due to their mode of isolation, the CHIP-DNA fragments bound by mutant p53 (Ser^{245}) in Onda 11 glioma cells in vivo are rather short and thus do not constitute MAR elements by themselves. Nevertheless, preliminary computer analyses are compatible with the idea that the repetitive DNA sequences isolated could be part of MAR elements. Indeed, the functions speculatively suggested here for the binding of mutant p53 (Ser^{245}) to some exemplary CHIP-sequences are compatible with the MAR concept.

Seven clones showed similarities to three types of satellite DNA sequences (α satellite, γ satellite, satellite III). Satellite sequences frequently cluster at centromeres and play an important role in the correct assembly of kinetochores, which are required for an accurate segregation of chromosomes during mitosis [Murphy and Karpen, 1998]. In this respect, it is of interest that it has been suggested that mutant p53 contributes to the loss of spindle checkpoint controls [Gualberto et al., 1998].

Five independent clones contained (CA)n repeats. This dinucleotide repeat is used as a microsatellite polymorphic marker, is widespread throughout the genome, and is conserved in many loci. Although some functional aspects have emerged, the biological role of (CA)n repeats is not yet understood in detail. However, an important feature of (CA)n repeats is their effect on DNA conformation. (CA)n repeats have the potential to induce a structural transition from a right-handed B-DNA conformation to a left-handed Z-DNA structure [Tripathi and Brahmachari, 1991]. Such a transition is thought to be involved in gene regulation. In line with this assumption, Shimajiri et al., [1999] reported that the promoter activity of the matrix metalloproteinase 9 (MMP-9) gene is dependent on the number of (CA)n repeats located in the promoter region. Floros et al. [1995] described that the number of (CA)n repeats located in intron 4 of the surfactant protein B (SP-B) gene is related to the incidence of the respiratory distress syndrome (RDS) in a large number of clinical samples. (CA)n repeats thus may be important determinants in modulating the expression of the corresponding genes. Therefore, the interaction of mutant p53 with (CA)n repeats might relate to the findings that mutant p53 acts as a transactivator of various cancer-associated genes in tumor progression [Malyapa et al., 1996; Deppert, 2000].

In summary, the DNA sequence information obtained from the analysis of a limited number of genomic DNA fragments serving as target sequences for mutant p53 (Ser^{245}) in Onda 11 cells suggests that the conformation-selective interaction of mutant p53 with non-coding repetitive DNA sequences present within some chromatin-structures (centromeres, telomeres), and around coding regions reflects the interaction of mutant p53 with MAR elements, and that such interactions play an important role in the control of chromatin organization and gene expression. It is tempting to speculate that the ensuing perturbations in chromatin structure and function form the molecular basis for the "gain of function" phenotype of at least some mutant p53 proteins.

TABLE 1. Pleiotropic "Gain of Function" Properties of Mutant p53: Possible Relation to its MAR-Binding Activity

Enhanced cell proliferation
Firing of additional, conditional cellular origins (targeting of replication complexes to conditional origins of replication)
Enhanced expression of growth promoting genes (e.g. PCNA)
Enhanced Tumorigenicity
Enhanced expression of tumor promoting genes (e.g. *mdr1*, VEGF)
Block of cellular differentiation by aberrant "fixation" of differentiation-specific chromatin domains
Block of apoptosis by protecting MAR elements from nucleolytic degradation
Enhanced genetic instability by promotion of recombination events (formation of recombination complexes at MAR elements)
Enhanced genetic instability due to loss of spindle check-point control (interaction of mutant p53 with centromere DNA)

FUTURE AVENUES

The above considerations suggest that the major transforming function of nuclear oncogenes associated with the chromatin and the nuclear matrix is to perturb nuclear structure. As changes in nuclear architecture are a hallmark of neoplastic transformation [Malyapa et al., 1996], an important future task will be to functionally analyze transformation associated changes in nuclear architecture, and their dependence on transforming proteins at the nuclear matrix. Three-dimensional microscopy and imaging techniques, combined with in situ labeling, already successfully applied to relate the dynamic changes in nuclear function to changes in nuclear organization during cell-cycle progression, should provide a powerful tool [Berezney and Wei, 1998]. Concerning the functional analysis of the association of an oncogene product with the nuclear matrix, the discovery that mutant p53 not only interacts with the nuclear matrix, but also specifically and with high affinity binds to MAR elements opened the possibility to develop a model for the biological implications for these interactions which now is put up for functional testing. Last not least, such analyses will pave the way for means of therapeutic intervention, e.g. by specific interference with MAR-binding of mutant p53. That such an intervention seems possible is suggested by the fact that the binding of mutant p53 to MAR elements, as well as to the repetitive DNA elements isolated from Onda 11 cells can be specifically inhibited by addition of a p53-specific monoclonal antibody recognizing an epitope within the C-terminal domain of p53.

ACKNOWLEDGMENTS

We thank Martina Hintz-Malchow for editing the manuscript.

REFERENCES

Albor A, Kaku S, Kulesz-Martin M. 1998. Wild-type and mutant forms of p53 activate human topoisomerase I: A possible mechanism for gain of function in mutants. Cancer Res 58:2091–2094.

Albrechtsen N, Dornreiter I, Grosse F, Kim E, Wiesmüller E, Deppert W. 1999. Maintenance of genomic integrity by p53: complementary roles for activated and non-activated p53. Oncogene 18:7706–7717.

Berezney R. 1991. The nuclear matrix: a heuristic model for investigating genomic organization and function in the cell nucleus. J Cell Biochem 47:109–123.

Berezney R, Wei X. Y. 1998. The new paradigm: integrating genomic function and nuclear architecture. J Cell Biochem Suppl 30–31:238–242.

Blandino G, Levine A. J, Oren M. 1999. Mutant p53 gain of function: differential effects of different p53 mutants on resistance of cultured cells to chemotherapy. Oncogene 18:477–485.

Bode J, Kohwi Y, Dickinson L, Joh T, Klehr D, Mielke C, Kohwi-Shigematsu T. 1992. Biological significance of unwinding capability of nuclear matrix-associating DNAs. Science 255:195–197.

Deppert W. 1996. Binding of MAR-DNA elements by mutant p53: Possible implications for its oncogenic functions. J Cell Biochem 62:172–180.

Deppert W. 2000. The nuclear matrix as a target for viral and cellular oncogenes. Crit Rev Eur Gene Expr 10:45–61.

Dittmer D, Pati S, Zambetti G, Chu S, Teresky A. K, Moore M, Finlay C, Levine A. J. 1993. Gain of function mutations in p53. Nat Genet 4:42–46.

Floros J, Veletza S. V, Kotikalapudi P, Krizkova L, Karinch A. M, Friedman C, Buchter S, Marks K. 1995. Dinucleotide repeats in the human surfactant protein-B gene and respiratory-distress syndrome. Biochem J 305:583–590.

Gualberto A, Aldape K, Kozakiewicz K. 1998. An oncogenic form of p53 confers a dominant, gain-of-function phenotype that disrupts spindle checkpoint control. Proc Natl Acad Sci USA 95:5166–5171.

Herbomel P. 1990. From gene to chromosome: organisation levels defined by the interplay of transcription and replication in vertebrates. New Biol 39:937–945.

Kim E, Albrechtsen N, Deppert W. 1997. DNA-conformation is an important determinant of sequence-specific DNA binding by tumor suppressor p53 Oncogene 15:857–869.

Koga H, Deppert W. 2000. Identification of Genomic DNA Sequences Bound by Mutant p53 Protein (Gly245- > Ser) in vivo. Oncogene 19:4178–4183.

Lin J, Teresky A. K, Levine A. J. 1995. Two critical hydrophobic amino acids in the N-terminal domain of the p53 protein are required for the gain of function phenotypes of human p53 mutants. Oncogene 0:2387–2390.

Malyapa R. S, Wright W. D, Taylor Y. C, Roti Roti J. L. 1996. DNA supercoiling changes and nuclear matrix-associated proteins: possible role in oncogene-mediated radioresistance. Int J Rad Oncol Biol Phys 35:963–973.

Murphy T. D, Karpen G. H. 1998. Centromeres take flight: alpha satellite and the quest for the human centromere. Cell 93:317–320.

Peled A, Zipori D, Rotter V. 1996. Cooperation between p53-dependent and p53-independent apoptotic pathways in myeloid cells. Cancer Res 56:2148–2156.

Pohl J, Goldfinger N, Radler-Pohl A, Rotter V, Schirrmacher V. 1988. p53 increases experimental metastatic capacity of murine carcinoma cells. Mol Cell Biol 8:2078–2081.

Shaulsky G, Goldfinger N, Rotter V. 1991. Alterations in tumor development in vivo mediated by expression of wild type or mutant p53 proteins. Cancer Res 51:5232–5237.

Shimajiri S, Arima N, Tanimoto A, Murata Y, Hamada T, Wang K. Y, Sasaguri Y. 1999. Shortened microsatellite d(CA)21 sequence down-regulates promoter activity of matrix metalloproteinase 9 gene. FEBS Lett 455:70–74.

Steinmeyer K. 1989. Charakterisierung nukleärer Subklassen des zellulären Tumorantigens. In: Biology. Ulm: Universität Ulm. p. 53.

Tripathi J, Brahmachari S. K. 1991. Distribution of simple repetitive (TG/CA)n and (CT/AG)n sequences in human and rodent genomes. J Biol Struct Dyn 9:387–397.

Will K, Deppert W. 1998. Analysis of mutant p53 for MAR-DNA binding: determining the dominant-oncogenic functio of mutant p53. Gene Ther Mol Biol 1:543–549.

Will K, Warnecke G, Wiesmüller L, Deppert W. 1998. Specific interaction of mutant p53 with regions of matrix attachment region DNA elements (MARs) with a high potential for base-unpairing Proc Natl Acad Sci USA 95:13681–13686.