Available online at www.sciencedirect.com
SCIENCE DIRECT®
Biochemical and Biophysical Research Communications 321 (2004) 21–25
BBRC
www.elsevier.com/locate/ybbrc

# Cadmium inhibits human DNA mismatch repair in vivo

Anne Lützen, Sascha Emilie Liberti, Lene Juel Rasmussen *

*Department of Life Sciences and Chemistry, Roskilde University, DK-4000 Roskilde, Denmark*

Received 16 June 2004
Available online 3 July 2004

**Abstract**

The heavy metal cadmium (Cd) is a human carcinogen that inhibits DNA repair activities. We show that DNA mismatch repair (MMR)-mediated cell cycle arrest after alkylation damage is suppressed by exposure to Cd and that this effect is reversed by preincubation with excess of zinc (Zn). We show that Cd-mediated inactivation of MMR activity is not caused by disruption of complex formation between the MMR proteins hEXO1-hMutSα and hEXO1-hMutLα nor does Cd inhibit 5′-exonuclease activity of hEXO1 in vitro. Thus, our studies show that exposure of human cells to Cd suppresses MMR activity, a repair activity known to play an important role in colon cancer and that this effect can be reversed by Zn treatment.


*Keywords:* hEXO1; hMSH2; DNA mismatch repair; Cancer; Cadmium

DNA repair pathways remove the majority of harmful lesions. However, if DNA repair fails, for instance if cells are exposed to cadmium (Cd), the result is DNA damage accumulation and one of the symptoms of DNA repair deficiency is development of cancer. One example of linkage between insufficient repair and cancer is the associations between defective DNA mismatch repair (MMR) and hereditary nonpolyposis colorectal cancer (HNPCC) [1]. Inactivation of MMR activity results in genetic instability, which is assumed to promote tumorigenesis [2]. Besides correcting replication errors, the MMR system is also implicated in other cellular processes such as recombination, activation of cell cycle checkpoints, and apoptosis. Following alkylation damage MMR-proficient cells, in contrast to MMR-deficient cells, arrest at the G2/M cell cycle checkpoint and, if the damage is extensive, induce apoptosis [3]. Cd is classified as a human carcinogen and is widely found in our environment [4]. The way Cd mediates toxicity and genotoxicity remains to be elucidated but several mechanisms have been suggested such as interaction with DNA and proteins, by generating reactive oxygen species, altering gene expression, impairing the cellular antioxidant defense system, interfering with signal transduction, activating cellular proto-oncogenes, and inhibiting DNA repair mechanisms [4]. Recent studies in yeast have shown that the MMR pathway is a target for Cd and that this metal causes genomic instability by inactivating yet unidentified factors in this key repair pathway [5]. In this report we extend the microbial studies to include human cells and show that MMR-mediated cell cycle arrest after alkylation damage is suppressed by exposure to cadmium. The Cd-mediated inactivation of MMR activity could be reversed by preincubation with zinc (Zn). It is known that Zn treatment reduces or abolishes the adverse effects of Cd [6]. However, the mechanism underlying this phenomenon is unclear. Cadmium competes with zinc for a variety of important binding sites in cells, such as zinc-finger motifs [7]. We show that Cd-mediated inactivation of MMR activity is not caused by disruption of specific MMR complexes nor does it inhibit hEXO1 activity. Thus, exposure of human cells to Cd suppresses MMR

* Corresponding author. Fax: +45-46-74-30-11.
*E-mail address:* ljr@ruc.dk (L.J. Rasmussen).

0006-291X/$ - see front matter © 2004 Elsevier Inc. All rights reserved.
doi:10.1016/j.bbrc.2004.06.102

activity, a repair activity known to play an important role in colon cancer, and this inhibition of MMR activity can be reversed by Zn treatment.

## Materials and methods

*Cell culture and flow cytometry*. Human 293T-Tet-Off-hMLH1 cells were maintained as described in [8]. For each cell line (293T-Tet-Off-hMLH1 + 50 ng/ml doxycycline [hMLH1−] or 293T-Tet-Off-hMLH1 − doxycycline [hMLH1+]), $2 \times 10^6$ cells were seeded per 75 $cm^2$ flasks in the presence of 0 or 20 μM zinc chloride ($ZnCl_2$) (Sigma–Aldrich). The indicated amount of cadmium chloride ($CdCl_2$) (Sigma–Aldrich) was added after 8 h. The next day cells were treated with 0 or 5 μM *N*-methyl-*N*′-nitro-*N*-nitrosoguanidine (MNNG) in serum-free medium, containing the indicated amount of $CdCl_2$ and/or $ZnCl_2$, for 60 min at 37 °C and 5% $CO_2$. After treatment cells were grown in serum-containing medium containing the indicated amount of $CdCl_2$ for 48 h. Attached cells were harvested by trypsin and pooled with detached cells. Cells were prepared for FACS analysis as described in [9]. For each sample $10^4$ cells were analyzed.

*Preparation of DNA substrate and exonuclease assays*. We used a 42 bp 5′-labeled double-stranded homoduplex substrate (A2/A9) described in Nielsen et al. [10] as well as a corresponding heteroduplex (A2(G23)/A9) substrate containing a G/T mismatch at position 23. The top strand oligonucleotides (A2 or A2(G23)) were labeled on the 5′-end using T4 polynucleotide kinase and [γ-$^{32}$P]ATP. Excess ATP was removed by using NucTrap Probe Purification Columns (Stratagene), and the labeled strand was annealed to an equal molar concentration of the complementary oligonucleotide (A9) in 20 mM Tris–HCl, pH 7.6, 0.01 mM EDTA, and 0.1 mM DTT by heating to 70 °C for 10 min followed by cooling to room temperature for 30 min and finally incubated on ice for 5 min. The nucleotide sequences of the oligonucleotides are: (A2) 5′-TAG AGG ATC CCC GCT AGC GGG TAC CGA GCT CGA ATT CAC TGG-3′, (A2(G23)) 5′-TAG AGG ATC CCC GCT AGC GGG TGC CGA GCT CGA ATT CAC TGG-3′, and (A9) 5′-CCA GTG AAT TCG AGC TCG GTA CCC GCT AGC GGG GAT CCT CTA-3′. Nuclease assays were performed as described in Nielsen et al. [10]. The reactions contained 10 pmol of purified hEXO1 recombinant protein, 0.25 pmol end-labeled DNA substrate, and 0, 5, 15, or 50 μM $CdCl_2$ in a final volume of 40 μl containing 20 mM Hepes, pH 7.5, 50 mM KCl, 0.5 mM DTT, 5 mM $MgCl_2$, 0.05% Triton X-100, 100 μg/ml BSA, and 5% glycerol. Reactions were carried out at 37 °C for 60 min and stopped immediately by adding 8 μl of formamide loading buffer and heating at 90 °C for 5 min. Aliquots were subsequently analyzed on a 12% polyacrylamide–8 M urea denaturing gel to determine the degree of conversion of substrate to mononucleotide product. Images were obtained using a STORM 840 Phosphoimager (Amersham Pharmacia Biotech).

*Pull-down assay*. All in vitro transcription translation reactions were carried out using the TNT coupled reticulocyte lysate system (Promega). Briefly, lysates were incubated with 1 μg DNA, amino acid mix lacking cysteine, [$^{35}$S]cysteine, and T7 RNA polymerase, for 90 min at 30 °C, according to manufacturer's instructions. The fusion proteins were purified as described by Jäger et al. [11], using noninduced *Escherichia coli* cultures. The pull-down assays were performed essentially as described by Nielsen et al. [10] with few modifications. Ten microliters of (~1 μg) GST-hMLH1 (100 μg/ml) or GST-hMSH2 (100 μg/ml) was bound to GST-beads (Glutathione–Sepharose 4B, Amersham Biosciences) by incubation for 2 h at 4 °C. Samples were transferred to a 5-ml tube (Falcon), diluted with binding buffer to approximately 50 μl GST-beads/ml, and incubated for 30 min at 4 °C. $CdCl_2$ was added simultaneously with 10 μl IVTT-synthesized proteins (hEXO1b, hMSH6, and hPMS2). The reaction mixture was incubated for 2 h at 4 °C. The GST-beads were washed three times with binding buffer before samples were resolved on 12% SDS–polyacrylamide gel (Cambrex Bio Science Rockland) and exposed to X-ray films (HyperfilmMP, Amersham Biosciences). Values for relative band intensities were determined using the Molecular Analyst software (Bio-Rad).

## Results

### *Cell cycle analysis of Cd-treated human cells*

MMR-proficient cells arrest in the G2 phase after treatment with alkylating agents such as MNNG, whereas MMR-deficient cells continue through the cell cycle [12]. We evaluated the cell cycle distribution of the human cells expressing hMLH1 from a doxycycline regulated promoter [8] after Cd and MNNG treatment (Figs. 1 and 2). Treatment of MMR+ or MMR− cells with Cd had no effect on the distribution of the viable cells in the cell cycle (Fig. 1). However, Cd treatment increased the fraction of cells in sub-G1 representing dead cells (data not shown). For both MMR+ and MMR− cells, we detected approximately 18% sub-G1 cells in samples treated with 2 μM Cd and approximately 30% sub-G1 cells in samples treated with 5 μM Cd. MMR-proficient cells accumulated in the G2 phase of the cell

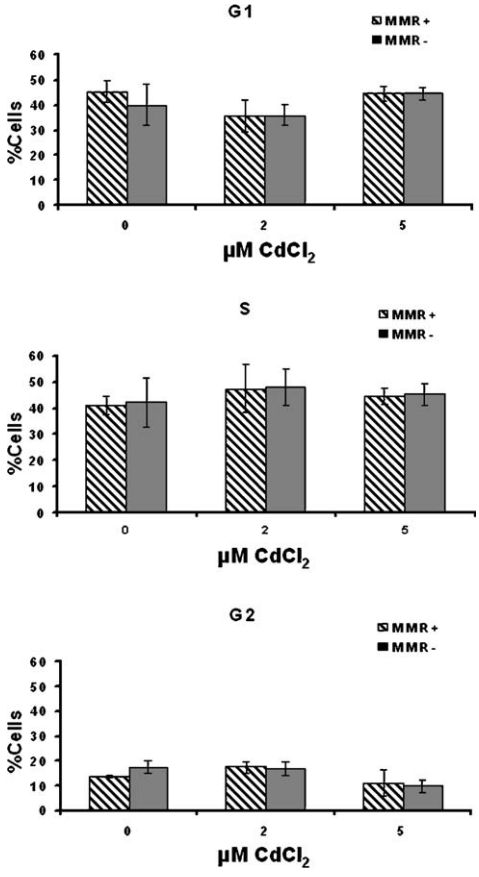


Fig. 1. Flowcytometric analysis of MMR+ and MMR− cells treated with Cd. The percentage of viable cells in various phases of the cell cycle is shown. Standard deviations are shown. The cells were grown and treated as described in Materials and methods.

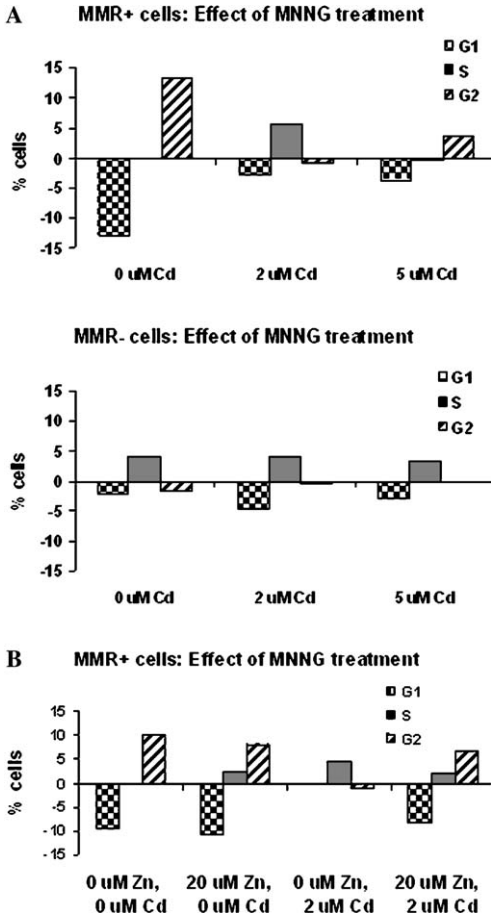


Fig. 2. Flowcytometric analysis of cells treated with Zn, Cd, and MNNG. (A) MMR+ and MMR− cells were treated with Cd as well as 0 or 5 μM MNNG. The percentage of cells in each phase of the cell cycle of samples not treated with MNNG was subtracted from the corresponding percentages in MNNG-treated samples, to illustrate the effect of MNNG on the cell cycle. The G2 arrest is significantly lowered by Cd treatment as determined by the one-tailed Mann–Whitney test ($\alpha = 0.05$). (B) MMR+ cells were treated with Zn and Cd as well as 0 or 5 μM MNNG. The effect of MNNG (as described in A) is presented. Each bar in this figure represents the average from at least three independent experiments.

cycle after treatment with MNNG (Fig. 2A) whereas MMR-proficient cells treated with both Cd and MNNG did not. This could indicate that Cd suppresses the MMR system resulting in MNNG tolerance. Thus, our results suggest (i) that Cd inactivates the human MMR system and (ii) that Cd causes cell death that is independent of MMR status.

It is known that Zn can suppress or abrogate the effects of Cd in many biological systems. In order to investigate whether Zn reverses the suppressive effect of Cd on the MMR system, we preincubated MMR+ cells with excess of Zn before addition of Cd. The cells did not accumulate in the sub-G1 fraction (data not shown) and the alkylation-induced G2 checkpoint response was restored (Fig. 2B), suggesting that Zn suppresses the effect of Cd on the MMR system.

*Does Cd disrupt protein–protein interactions in MMR complexes or does it affect exonuclease activity of hEXO1?*

Complex formations between MMR proteins are essential for repair activity and it is, therefore, possible that Cd inactivates MMR by preventing or disrupting

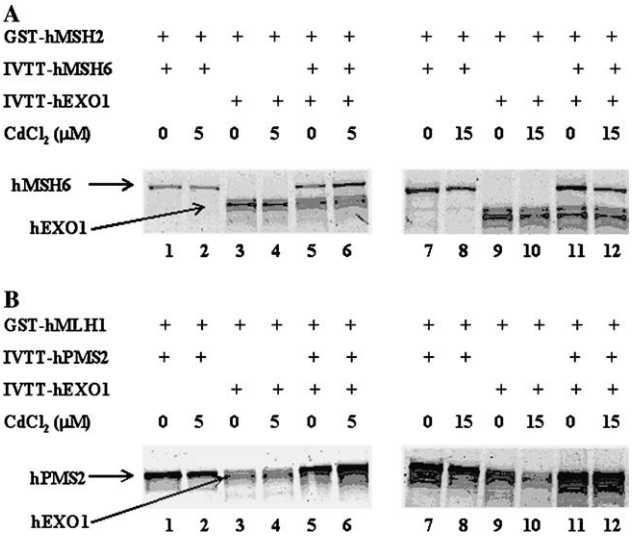


Fig. 3. GST-fusion protein assay to study the influence of Cd on interactions between MMR complexes. Proteins were purified and reactions were performed as described in Materials and methods. (A) Influence of Cd on the interactions between hMSH2, hMSH6, and hEXO1. Lane 1, GST-hMSH2 and labeled IVTT hMSH6; lane 2, GST-hMSH2, labeled IVTT hMSH6, and 5 μM Cd; lane 3, GST-hMSH2 and labeled IVTT hEXO1; lane 4, GST-hMSH2, labeled IVTT hEXO1, and 5 μM Cd; lane 5, GST-hMSH2, labeled IVTT hMSH6, and labeled IVTT hEXO1 (hMSH6 and hEXO1 were added to hMSH2 simultaneously); lane 6, GST-hMSH2, labeled IVTT hMSH6, labeled IVTT hEXO1, and 5 μM Cd (Cd, hMSH6, and hEXO1 were added to hMSH2 simultaneously); lane 7, GST-hMSH2 and labeled IVTT hMSH6; lane 8, GST-hMSH2, labeled IVTT hMSH6, and 15 μM Cd; lane 9, GST-hMSH2 and labeled IVTT hEXO1; lane 10, GST-hMSH2, labeled IVTT hEXO1, and 15 μM Cd; lane 11, GST-hMSH2, labeled IVTT hMSH6, and labeled IVTT hEXO1 (hMSH6 and hEXO1 were added to hMSH2 simultaneously); and lane 12, GST-hMSH2, labeled IVTT hMSH6, labeled IVTT hEXO1, and 15 μM Cd (Cd, hMSH6, and hEXO1 were added to hMSH2 simultaneously). (B) Influence of Cd on the interactions between hMLH1, hPMS2, and hEXO1. Lane 1, GST-hMLH1 and labeled IVTT hPMS2; lane 2, GST-hMLH1, labeled IVTT hPMS2, and 5 μM Cd; lane 3, GST-hMLH1 and labeled IVTT hEXO1; lane 4, GST-hMLH1, labeled IVTT hEXO1, and 5 μM Cd; lane 5, GST-hMLH1, labeled IVTT hPMS2, and labeled IVTT hEXO1 (hPMS2 and hEXO1 were added to hMLH1 simultaneously); lane 6, GST-hMLH1, labeled IVTT hPMS2, labeled IVTT hEXO1, and 5 μM Cd (Cd, hPMS2, and hEXO1 were added to hMLH1 simultaneously); lane 7, GST-hMLH1 and labeled IVTT hPMS2; lane 8, GST-hMLH1, labeled IVTT hPMS2, and 15 μM Cd; lane 9, GST-hMLH1 and labeled IVTT hEXO1; lane 10, GST-hMLH1, labeled IVTT hEXO1, and 15 μM Cd; lane 11, GST-hMLH1, labeled IVTT hPMS2, and labeled IVTT hEXO1 (hPMS2 and hEXO1 were added to hMLH1 simultaneously); and lane 12, GST-hMLH1, labeled IVTT hPMS2, labeled IVTT hEXO1, and 15 μM Cd (Cd, hPMS2, and hEXO1 were added to hMLH1 simultaneously).

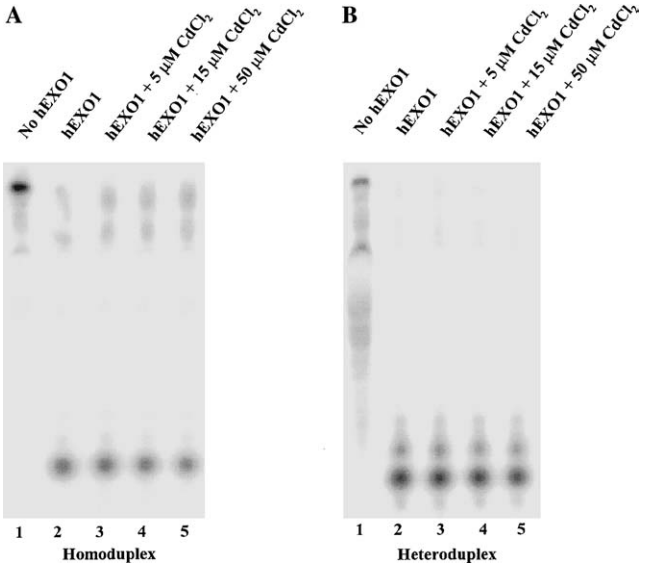


Fig. 4. Exonuclease 1 activity on a 5′ $^{32}$P-labeled double-stranded (A) homoduplex and (B) heteroduplex DNA. Lane 1, 0 pmol hEXO1; lane 2, 10 pmol hEXO1; lane 3, 10 pmol hEXO1 + 5 μM $CdCl_2$; lane 4, 10 pmol hEXO1 + 15 μM $CdCl_2$; and lane 5, 10 pmol hEXO1 + 50 μM $CdCl_2$.

such complexes. By applying a GST-fusion interaction assay, we have previously shown that hEXO1 and hMutLα proteins likely form a ternary complex [10]. In this report we expand these observations to include hEXO1 and hMutSα (Fig. 3A, lanes 5 and 11). Control experiments performed essentially as described in [10] showed that under the experimental conditions employed in Fig. 3 the GST-hMSH2 as well as the GST-hMLH1 proteins, which are bound to the beads, are saturated with the first added protein before the second protein is added to the reaction mixture (data not shown). However, it cannot be completely excluded that some of the preincubated protein may dissociate and become exchanged with the second protein. Furthermore, we did not detect any binding of labeled hEXO1, hMSH6 or hPMS2 to the GST-beads (data not shown). Results presented in Fig. 3 demonstrate that protein interactions between hEXO1 and hMutLα as well as between hEXO1 and hMutSα are not disrupted in the presence of Cd. To test whether Cd could inhibit the excision step of the MMR process, we compared the 5′-exonuclease activity of hEXO1 on homoduplex and G/T containing heteroduplex substrates after addition of Cd. Our results show that Cd at a concentration up to 50 μM did not influence exonuclease activity of hEXO1 (Fig. 4) under the experimental conditions used in this study.

## Discussion

We have shown that treatment of human MMR-proficient cells with the environmental carcinogen Cd abrogates MMR-mediated G2 checkpoint arrest following MNNG treatment (Figs. 1 and 2). Cejka et al. [8] reported that these cells require the full complement of hMLH1 for checkpoint activation. Thus, one possible explanation for the effect of Cd on MMR could be reduced expression of one or more MMR proteins. However, Western blot analysis of cells treated with Cd showed no difference in protein levels of hMLH1, hMSH2, hMSH6, and hPMS2 (data not shown). Therefore, we do not expect that Cd impedes MMR-mediated G2 arrest after alkylation damage by reducing the levels of these MMR proteins in the cells.

We found no effect of Cd treatment on complex formation between MMR proteins in vitro (Fig. 3). Since the GST-fusion interaction assay provides mainly qualitative information, it cannot be ruled out that the MMR inhibitory effect of Cd is not a result of stoichiometric changes in the MMR protein complexes in vivo. It is expected that even modest alterations in the assembly of the MMR complexes cause destabilization of the repair machinery and increased mutability. Furthermore, Cd might function by increasing binding affinities between MMR proteins, thereby, sequestering these and retaining them from functioning.

To test whether Cd could inhibit the excision step of the MMR process, we compared the 5′-exonuclease activity of hEXO1 on homoduplex and G/T containing heteroduplex substrates after addition of Cd. We found no difference in exonuclease activity between hEXO1 incubated with Cd on neither substrate (Fig. 4). One important prerequisite for the initiation of repair is the binding of repair proteins to DNA mismatches and DNA lesions. Since Cd has been shown to inhibit DNA–protein interactions essential for the initiation of nucleotide excision repair (NER) [13], we investigated whether Cd was capable of inhibiting hMutSα binding to a G/T mismatch. Our preliminary results show that cadmium does not interfere with mismatch binding of hMutSα (data not shown). However, it is possible that Cd would have an effect on mismatch recognition by hMutSα under different experimental conditions.

We found that preincubation of cells with excess of Zn before Cd treatment could abrogate the suppressive effect of cadmium on the MMR system. The mechanism of the protective effect of Zn on Cd-treated cells is largely unknown. One possible mechanism is that Zn can induce the synthesis of the metal-binding protein, metallothionein (MT), which binds Cd with high affinity, thus sequestering Cd and thereby decreasing the adverse effect of Cd [14]. Cd has been reported to induce the synthesis of MT [15] and stably transfected cells overexpressing MT are more resistant to MNNG compared to parental cells [16]. Thus, it could be argued that the effect of Cd reported in this paper is not due to a suppressive effect of MMR but rather is

due to an upregulation of MT, which in turn confers resistance to MNNG. However, the level of MT does not seem to parallel the degree of protection from MNNG [16]. On the other hand, Zn pretreatment causing a concentration-dependent increase in MT was indeed able to protect cells from MNNG-induced killing [17]. If MT is responsible for the phenotypes presented in this work, we would expect to see a protective effect of Zn on MNNG-induced cell cycle effects. However, we do not observe such an effect (Fig. 2).

Another possible target for Cd is the replication protein A (RPA), a protein required for complete MMR [18]. The RPA protein contains a conserved zinc-finger motif in the large subunit, Rpa1, where Zn complexes four cysteines and it was demonstrated that MMR requires an intact zinc finger in RPA to be active [19]. Since Cd shows high affinity for zinc-finger domains and the function of zinc-finger proteins is inhibited by low concentrations of Cd it can be speculated that Cd suppresses MMR by inhibiting the RPA protein.

In summary, our results indicate that environmentally relevant concentrations of Cd are able to sufficiently suppress the MMR system to abrogate the G2 cell cycle checkpoint arrest following alkylation damage. Normally, colonocytes injured by DNA alkylating agents would be expected to arrest at the G2 checkpoint, and if the DNA damage is beyond repair, initiate apoptosis to eliminate damaged cells. Our results suggest that if cells are exposed to Cd they may escape G2 arrest and/or apoptosis following DNA damage, thus continuing proliferation of cells with genetic changes, the hallmark of cancer cells. Furthermore, it is possible that Cd, independent of MMR inhibition, activates proto-oncogenes, such as c-*fos*, c-*myc*, and c-*jun* [4]. Along these lines, it has been shown that Cd inhibits apoptosis in various cell types [4], which in combination with inhibition of MMR could further enhance the expansion of cells with genetic changes resulting from chemically or spontaneously induced DNA damage.

## Acknowledgments

We are indebted to Dr. Dmitry Gordenin for encouragement, advice, suggestions, and critical reading of the manuscript. We are grateful to Anne Marie Bundgaard for expert technical assistance. We are grateful to Dr. Josef Jiricny for the 293T cell line. The Danish Cancer Society (L.J.R., S.E.L.), Danish Research Council (L.J.R.), Danish Cancer Research Foundation (L.J.R.), Else and Aage Gronbech-Olsen Foundation (A.L.), and Jens and Korna Christensens Foundation (A.L.) supported this work.

## References

[1] P. Peltomaki, Role of DNA mismatch repair defects in the pathogenesis of human cancers, J. Clin. Oncol. 21 (2003) 1174–1179.
[2] C.R. Boland, Molecular genetics of hereditary nonpolyposis colorectal cancer, Ann. N. Y. Acad. Sci. 910 (2000) 50–59.
[3] A. Bellacosa, Functional interactions and signaling properties of mammalian DNA mismatch repair proteins, Cell Death Differ. 8 (2001) 1076–1092.
[4] M. Waisberg, P. Joseph, B. Hale, D. Beyersmann, Molecular and cellular mechanisms of cadmium carcinogenesis, Toxicology 192 (2003) 95–117.
[5] Y.H. Jin, A.B. Clark, R.J.C. Slebos, H. Al-Refai, J.A. Taylor, T.A. Kunkel, M.A. Resnick, D.A. Gordenin, Cadmium is a mutagen that acts by inhibiting mismatch repair, Nat. Genet. 34 (2003) 326–329.
[6] A.B. Fischer, Y. Skreb, In vitro toxicology of heavy metals using mammalian cells: an overview of collaborative research data, Arh. Hig. Rada. Toksikol. 52 (2001) 333–354.
[7] A. Hartwig, Zinc finger proteins as potential targets for toxic metal ions: differential effects on structure and function, Antioxid. Redox Signal. 3 (2001) 625–634.
[8] P. Cejka, L. Stojic, N. Mojas, A.M. Russell, K. Heinimann, E. Cannavo, M. di Pietro, G. Marra, J. Jiricny, Methylation-induced G(2)/M arrest requires a full complement of the mismatch repair protein hMLH1, EMBO J. 22 (2003) 2245–2254.
[9] A. Lützen, H.C. Bisgaard, L.J. Rasmussen, Differential Cyclin D1 expression and apoptosis in DNA mismatch repair-deficient cells upon methylation and UV-C damage, Exp. Cell Res. 292 (2004) 123–134.
[10] F.C. Nielsen, A.C. Jäger, A. Lützen, J.R. Bundgaard, L.J. Rasmussen, Characterization of human exonuclease 1 in complex with mismatch repair proteins, subcellular localization and association with PCNA, Oncogene 23 (2004) 1457–1468.
[11] A.C. Jäger, M. Rasmussen, H.C. Bisgaard, K.K. Singh, F.C. Nielsen, L.J. Rasmussen, HNPCC mutations in the human DNA mismatch repair gene hMLH1 influence assembly of hMutLalpha and hMLH1-hEXO1 complexes, Oncogene 20 (2001) 3590–3595.
[12] M.T. Hawn, A. Umar, J.M. Carethers, G. Marra, T.A. Kunkel, C.R. Boland, M. Koi, Evidence for a connection between the mismatch repair system and the G2 cell cycle checkpoint, Cancer Res. 55 (1995) 3721–3725.
[13] M. Hartmann, A. Hartwig, Disturbance of DNA damage recognition after UV-irradiation by nickel(II) and cadmium(II) in mammalian cells, Carcinogenesis 19 (1998) 617–621.
[14] C.D. Klaassen, S. Liu, S. Choudhuri, Metallothionein: an intracellular protein to protect against cadmium toxicity, Annu. Rev. Pharmacol. Toxicol. 39 (1999) 267–294.
[15] S.L. Samson, L. Gedamu, Molecular analyses of metallothionein gene regulation, Prog. Nucleic Acid Res. Mol. Biol. 59 (1998) 257–288.
[16] B. Kaina, H. Lohrer, M. Karin, P. Herrlich, Overexpressed human metallothionein IIA gene protects Chinese hamster ovary cells from killing by alkylating agents, Proc. Natl. Acad. Sci. USA 87 (1990) 2710–2714.
[17] P. Moffatt, G. Plaa, F. Denizeau, Rat hepatocytes with elevated metallothionein expression are resistant to *N*-methyl-*N*′-nitro-*N*-nitrosoguanidine cytotoxicity, Toxicol. Appl. Pharmacol. 136 (1996) 200–207.
[18] J. Genschel, P. Modrich, Mechanism of 5′-directed excision in human mismatch repair, Mol. Cell 12 (2003) 1077–1086.
[19] Y.-L. Lin, K.K. Shivji, C. Chen, R. Kolodner, R.D. Woods, A. Dutta, The evolutionarily conserved zinc finger motif in the largest subunit of human replication protein A is required for DNA replication and mismatch repair but not for nucleotide excision repair, J. Biol. Chem. 273 (1998) 1453–1461.