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REVIEW PAPER

Concerted control of DNA double strand break repair and cell cycle progression in X-irradiated mammalian cells

Barbara Sochanowicz, Irena Szumiel

Abstract Upon examination of cell cycle regulation in a damaged cell, relations were discovered of the cell cycle control mechanisms with a complicated web of signalling pathways, eventually called the genome surveillance system. After infliction of DNA double strand breaks (DSB), the signalling is initiated by sensor proteins and transducer protein kinase ATM. This kinase phosphorylates downstream effector proteins, such as checkpoint kinases CHK1 and CHK2, which initiate the pathways leading to cell cycle arrest. In contrast with the older model of linear transmission of signals in a certain sequence, it is now accepted that the damage signalling system is branched and contains feedback loops. DSB's presence is signalled by sensor proteins (MRE11-RAD50-nibrin complex, MRN) to ATM and the signal is amplified through adaptor proteins, MDC1/NFBD1 or 53BP1 (Tp53 binding protein). MRN contains a forkhead-associated (FHA) domain and BRCA1 carboxyl-terminal (BRCT) domain. The combination of the FHA/BRCT domains has a crucial role for the binding of nibrin to the H2AX histone, assembling the components of repair foci. These domains also are important for interaction of other proteins localised in the foci. For example, MDC1/NFBD1 contains a FHA domain and two BRCT domains which are involved in protein interactions. The signal generated at DSBs is amplified and transduced to recruit components of DNA repair systems. In a concerted way with the sequential recruitment of components of repair foci, activation of transcription of genes takes place, that is necessary for blocking progression through the cell cycle, for DNA repair or apoptosis.

Key words ATM kinase • DNA double strand break repair • RAD50 • cell cycle arrest • repair foci • BRCT domain

Introduction

Cells are targets of environmental factors and agents that damage DNA and may lead to its erroneous replication or transcription. The biological consequences depend on the cellular phenotype and the extent of damage, and range from various types of heritable damage to cell death. Also, replication and recombination may be a source of errors. Hence, cellular defence mechanisms are very important. A striking feature of the cellular response to DNA damaging agents is the concerted activation of DNA repair processes and arrest of the cell's progression through cell cycle phases until the repair is completed.

Progression through the cell cycle can be treated as a chain of events in a strictly defined sequence until the cell divides into two daughter cells. When the event B can take place only after the event A is completed, and this relation is abolished by mutation, this indicates that there is a checkpoint in the A → B transition [49]. Such transition may depend on numerous factors, among them – synthesis of signalling proteins, their posttranslational modification (usually phosphorylation) or translocation between cell compartments, synthesis of substrates and enzymes needed for DNA replication. The web of control mechanisms has been called genome surveillance system. Most of its components have struc-

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tural features of striking similarity from yeast to mammals. Owing to the structure-function relation, amazingly conserved during evolution, experiments on yeast cells usually supply data that can be taken as clues for studies of mammalian cells. Human homologues of yeast proteins have the same or closely related function in the stress response.

Double strand breaks (DSB) constitute ionising radiation-induced DNA lesions most difficult for faultless repair; they are lethal, if left unrepaired or misrepaired. In this paper, some features of the cellular response to this type of DNA damage are reviewed, with emphasis on coordination of DSB repair and checkpoint control.

Generation of the damage signal

Figure 1 shows diagrammatically the standard concept of the sequence of events that take place in the mammalian cell after infliction of DSB (after [57], modified). The diagram also lists the most important proteins of this phase of cellular response according to their function. These proteins were classified by Melo and Toczyski [31], O'Connell *et al.* [35] and McGarry [30]. They can be divided into the following groups:

- phosphatidyl inositol-3 kinase like (PIKL) protein kinase: ATM, ataxia telangiectasia mutated protein; ATR, ataxia telangiectasia and Rad3-related protein; the latter is mainly responding to stalled replication forks and will be omitted from the further discussion;
- trimer forming proteins similar to PCNA (proliferating cell nuclear antigen), Rad9, Rad1, Hus1 (the 9-1-1 complex);
- serine-threonine-specific kinases or effector kinases: CHK1 i CHK2 (checkpoint protein kinases 1 and 2);
- adaptor proteins (claspin, BRCA1, 53BP1, NFBD1, a "nuclear factor with an amino-terminal FHA domain and a tandem repeat of BRCT domains" also called MDC1 (mediator of DNA damage checkpoint 1).

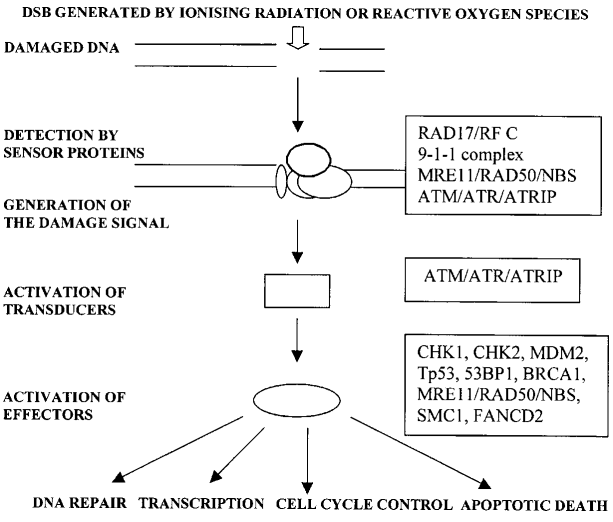

Fig. 1. The standard concept of the sequence of events that take place in the mammalian cell after infliction of DSB (after Zhou and Elledge [57], modified).

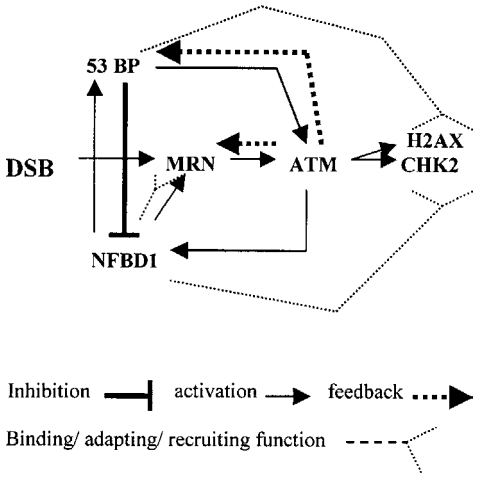

Fig. 2. Main pathways of DSB detection and signalling accounting for the recent observations (according to Mochan *et al.* [32] with considerable modifications). See the list of abbreviations for the names of the participating proteins.

Interestingly, this classification does not include the so-called MRN complex (MRE11, RAD50, NBS) which was long considered a DNA repair complex rather than an actor of the early events in the cell's response to DSB generation. In fact, both the classification of proteins and the linear sequence of activation events shown in the diagram (Fig. 1) is an oversimplification. Notably, Fig. 1 lists ATM and ATR both in the group of sensors and transducers; the trimer-forming proteins seem to act as adaptors rather than sensors [38]. Some of the adaptor proteins behave like direct participants of the repair processes. The putative sensors, RAD17/RF-C and the 9-1-1 complex, according to a recent review, act as adaptors in stalled replication forks and recruit ATR-associated with a specific protein – ATRIP (ATR interacting protein) [38], whereas 53BP1 according to Iwabuchi *et al.* [17] also is a repair protein. The function of the effector proteins SMC1 and FANCD2 will be discussed in the section concerning the S phase checkpoints.

The surprising observation that ATM can be found both upstream and downstream of the adaptor proteins (reviewed in [32]), lead to revision of the linear signalling pathway. Figure 2 shows a modified diagram of the early steps of DSB signalling; the sequential steps and the roles of particular proteins are discussed in the further text. In particular, Fig. 3 explains the mechanism of DSB detection, and the function of adaptor proteins is described in a separate section preceding the concluding remarks.

Detection of DSB

It is assumed that every type of damage, including DSB, alters chromatin conformation and this alteration is sensed by proteins that are placed in strategic sites of the chromatin, very much like a spider in the centre of his web detects the weakest movement of its threads. To function effectively, a damage sensor must be able to

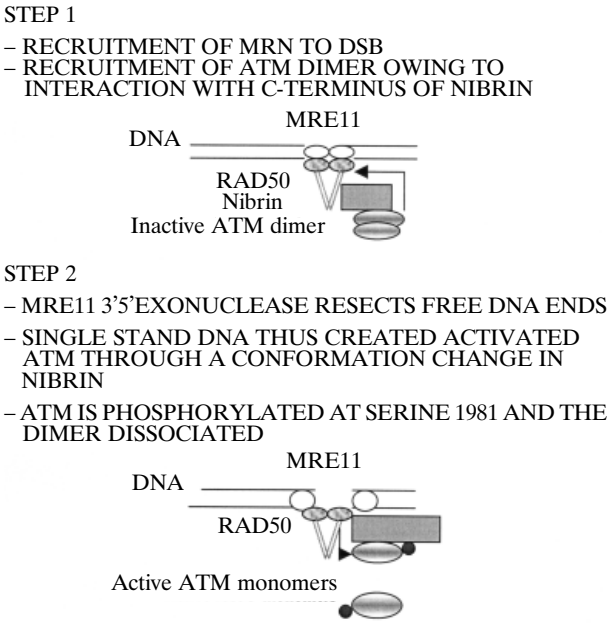

Fig. 3. The two-step model of DSB detection carried out by the MRN complex and ATM (according to Abraham [1] modified). See text for explanations.

detect very small numbers of DSB, since even a single DSB can cause apoptosis, or a lethal chromosomal aberration. Furthermore, the sensor should trigger events that lead to an amplification of the initial signal.

There are several candidate proteins to which the sensor function has been ascribed, however, recent discoveries have shown that most of them have more than one function. The secret of DSB detection has been solved only recently. According to the latest experiments [22], the sensor function is carried out by the MRN complex and ATM. This has been documented in carefully planned experiments with reconstruction *in vitro* of the events that lead to ATM activation. Figure 3 shows the two steps of this process.

– Step 1 consists of recruitment of the MRN complex to DSB. M is MRE11, shown as white ovals; R is RAD50, a very unusual protein that finds DSB and binds to the free DNA ends (Fig. 4 shows its structure). RAD50 contains 2 long coiled-coil structures (forming V in the diagram); at the apex of the coiled coils, pairs of conserved Cys-X-X-Cys motifs form "hooks" that bind one Zn^{2+} ion. ATPase domains are at the ends of the molecule and have a globular form; they are close to the MRE11 binding sites and are shown as shaded grey ovals in the diagram. ATP hydrolysis supplies energy for a conformation change in RAD50 that probably facilitates separation of DNA strands and the enzymatic function of MRE11. It is assumed that the conformation change in RAD50 also results in a concerted change in nibrin conformation. Then, recruitment of the ATM dimer takes place, owing to interaction with the C-terminus of nibrin (shown in the diagram as a grey rectangle).
– In step 2, the MRE11 3',5'exonuclease resects and unwinds free DNA ends; single strand DNA thus created is the "genuine alarm signal". As it activates ATM through a conformation change in nibrin, ATM becomes phosphorylated at serine 1981 and the dimer dissociates. The monomeric ATM molecule has protein kinase activity. As follows from the further text, this is the key event in the control of DSB repair and cell cycle progression.

It should be noted, however, that in contrast to Lukas *et al*. [26, 27], Cerosaletti and Concannon [8] claim that nibrin is not necessary for this initial step in ATM activation. This controversy has not yet been solved.

Repair foci

One of the earliest events after dissociation of the ATM dimers and activation of its kinase function is phosphorylation of the H2AX histone [7, 41]. This triggers a marked change in chromatin structure in every DNA region containing a DSB. The change comprises a DNA region corresponding, on the average, to 1–2 Mbp (mega base pairs) per DSB [40]. In these regions, adaptor proteins, repair enzymes and checkpoint control kinases and proteins are forming focal assemblies which can be visualised in a fluorescence microscope as "repair foci". A review published in *Nukleonika* [23] gives a more detailed information on this subject.

Using specific antibodies conjugated with fluorescent dyes, it is possible to follow the sequence in which the proteins are assembled into foci or become dislocated [39]. Such foci correspond in number to DSB and thus, it is possible to have DSB visualised and directly counted. Figure 5 presents an example of microscopic image of control and X-irradiated cells with repair foci visualised with anti-H2AX histone monoclonal antibody. This is the basis of a recently established method of estimation of the initial number of DSB after exposure to ionising radiation and DSB repair kinetics, usually with the use of anti-H2AX histone monoclonal antibody.

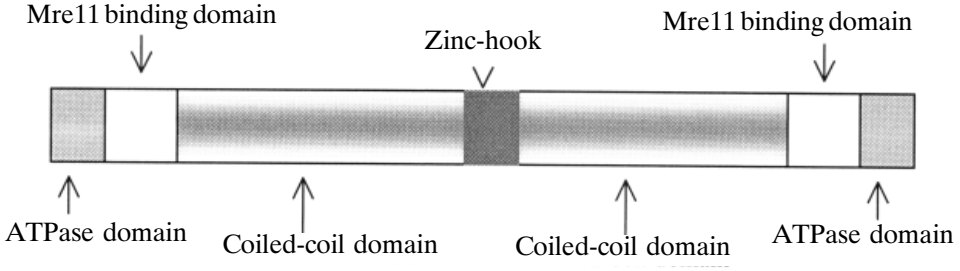

Fig. 4. Schematic structure of the RAD50 molecule. The drawing is not to scale.

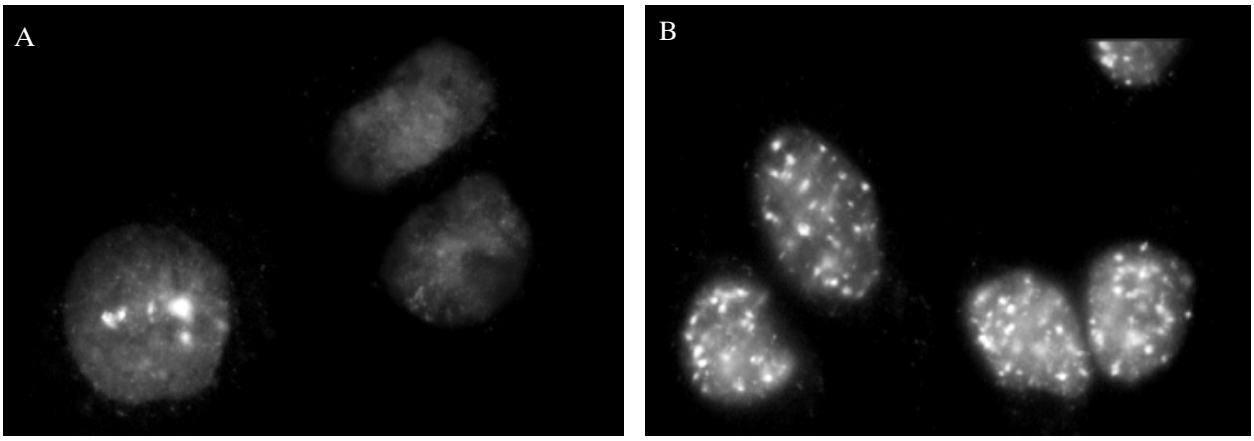

Fig. 5. Microscopic image of control and X-irradiated (2 Gy) Chinese hamster ovary cell nuclei with repair foci visualised with anti-H2AX histone monoclonal antibody. A – control cell nuclei which occasionally show foci during S phase; B – irradiated cells with numerous foci. Courtesy of Dr M. Wojewódzka.

It should be noted that not only DSB induce foci formation. The protein composition of the repair foci depends on the nature of the DNA lesion. Notwithstanding the type of damage, the function of the focal assembly of repair proteins is co-ordinated with the cell's position in the cell cycle. In the case of DSB repair, the choice of repair system is between NHEJ (non-homologous end-joining) and HRR (homologous recombination repair). NHEJ may act during the whole cell cycle but is essential in G1 and early S phases. Later in S phase, where lesions are present in duplicated DNA, HRR predominates, since the undamaged homologous DNA strand serves as template for repair replication. After completion of repair, the system that signals the presence of DNA breaks is switched off. Then, the cell with restored DNA integrity is able to resume its progress through the cell cycle.

The central role of ATM kinase

Substrates of ATM kinase are listed in Table 1, whereas a very simplified diagram of the most important ATM functions is presented in Fig. 6. It can be seen that directly or indirectly, ATM controls DNA repair,

Table 1. The most important substrates of ATM kinase (based on the papers [10, 21, 25])

Substrate	Function in
Histone H2AX	Repair foci assembly
MRN, MRE11/RAD50/NBS complex	DSB detection, DSB repair, S arrest
53BP1 (see abbreviations)	Repair foci assembly, signal amplification
NFBD1/MDC1 (see abbreviations)	Repair foci assembly, signal amplification
Tp53, tumor supressor protein 53	G1/S arrest, G2 arrest, apoptosis
c-ABL kinase	G1/S arrest, apoptosis, HRR
CHK1, checkpoint kinase 1	G2 arrest
CHK2, checkpoint kinase 2	G1/S and G2 arrest
FANCD2	S arrest
RAD9, part of the DNA damage sensor complex, 9-1-1	S arrest
SMC1, structural maintenance of chromosomes protein 1, a chromosomal ATPase with a role in sister chromatid cohesion	S arrest
RPA2, replication protein 2	S arrest
BRCA1, breast cancer susceptibility 1	DNA repair, S and G2 arrest, scaffold for ATM substrates
c-JUN, transcription factor	Transcription
IKK, NF-kappaB inhibitor (IkappaB) kinase	Transcription
HDAC, histone deacetylase	Transcription
TRF1, telemeric repeat binding factor	Telemere function
MDM2, ubiquitin ligase	Degradation of Tp53
E2F1, transcription factor	Apoptosis, transcription

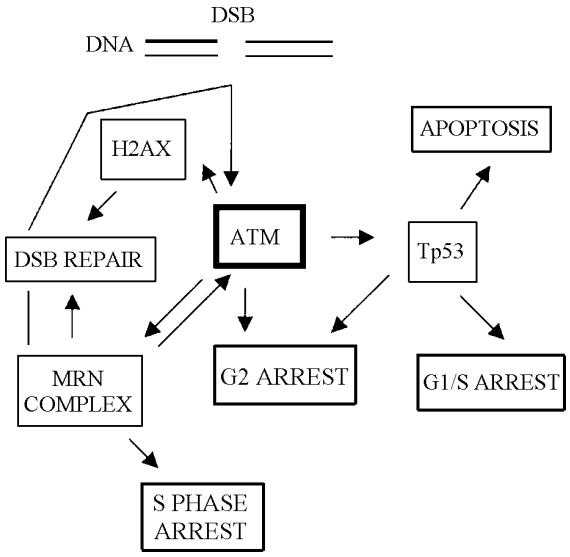

Fig. 6. A simplified diagram showing the central role of ATM kinase in the cellular response to DSB). See the list of abbreviations for the names of the participating proteins.

apoptosis and cell cycle arrests damage induced by DNA. In the downstream signalling, a similar central role is played by Tp53 (tumor supressor protein 53) which controls G1/S arrest, G2 arrest and apoptosis (reviews in [15, 16].

The G_1/S checkpoint

After DSB induction, ATM and ATM-activated checkpoint kinases phosphorylate Tp53 [34]. This brings numerous consequences, some of which are shown in Fig. 7 (review in [15, 16]). Tp53 degradation stops, its molecules become stabilised and increase in number. Further post-translational modifications allow Tp53 to play the role of transcription factor for several genes. Activated transcription and expression of CDKN1A (also known as p21(cip1/waf1)), inhibitor of cyclin-dependent kinases leads to arrest at the G1/S phase boundary, because inhibition of G1 phase specific kinases (CDK/cyclin E and CDK4/cyclin D1) prevents phosphorylation of the retinoblastoma protein, thus blocking the E2F transcription factor. The latter is necessary for transcription of S phase specific genes. Additionally, CDKN1A binds PCNA, impairing DNA-polymerase δ. As long as the presence of DSB is signalled, the cell stays in G1 phase. This mechanism prevents replication of damaged DNA and leaves time necessary for repair. Also transcription of genes that control apoptosis is activated or inhibited by Tp53. Such an effect takes place when the repair of DNA damage is delayed or impaired. Defect in checkpoint control may also be a source of "death signal" activating the Tp53-dependent apoptosis.

The S checkpoints

Similarly to other checkpoints, the S checkpoints were studied mostly in X-irradiated mammalian cells. DNA

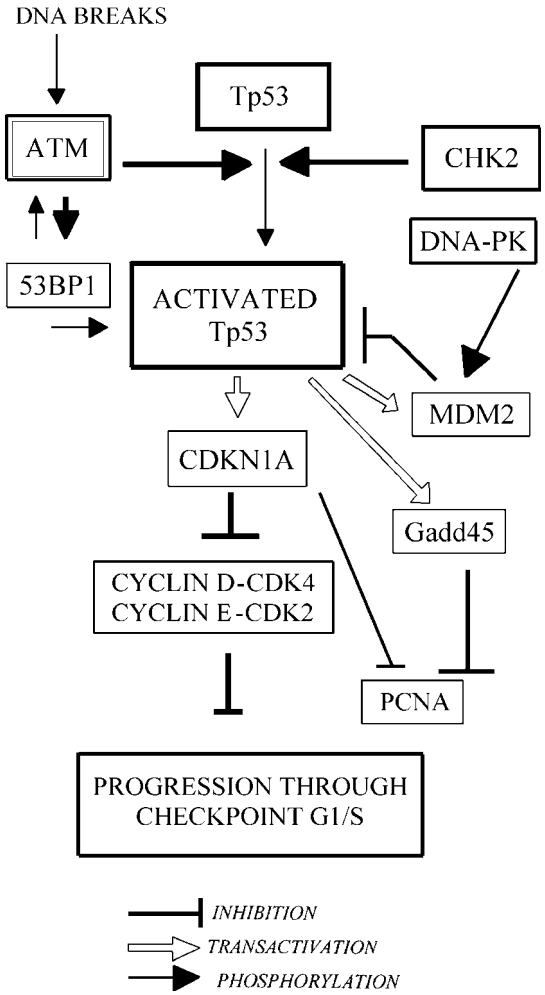

Fig. 7. Signalling pathways involving Tp53 and affecting progression through the cell cycle (based on the data from [15, 16, 26]. See the list of abbreviations for the names of the participating proteins.

replication stops a few minutes after irradiation. The dependence of inhibition percentage on radiation dose (dose-effect curve) is bi-phasic. The initial fast decrease with increasing radiation dose is caused by inhibition of replicon initiation; the second slower phase corresponds with elongation inhibition in replicons where initiation took place before exposure [37, 48]. About 30 years ago this relation was considered as a direct effect of DNA breakage. Then, Wang *et al.* [46] have shown that inhibition is due to regulatory factors: plasmid DNA was replicated *in vitro* by extracts from control cells, but not from irradiated cells. The plasmid DNA was not irradiated, hence, it did not contain any lesions that could inhibit replication.

Bartek *et al.* [5] describe the three checkpoints of the S phase as follows: ..."the DNA-damage-induced, replication-independent, intra-S-phase checkpoint (the 'intra-S-phase checkpoint'); the replication-dependent S-phase checkpoint (the 'replication checkpoint'); the replication-dependent S–M checkpoint (the 'S–M checkpoint'), which prevents mitotic entry when DNA is incompletely replicated".

The list of proteins that are necessary for post-irradiation inhibition of DNA replication is still expanding.

At present it comprises ATM [36], calmodulin-dependent kinase II [11], nibrin [14], BRCA1 [12, 28, 50, 51], RAD17, CHK2 [12], SMC1 and FANCD2 (reviewed by Bartek *et al*. [5], Kobayashi *et al*. [20]). Some of the S checkpoint proteins also directly participate in DSB repair, like nibrin [2, 20] and BRCA1 [56]. The best characterised intra-S-phase checkpoint involves the combined action of the ATM/ATR–CHK2/CHK1–CDC25A–CDK2 pathway and the ATM–NBS–FANCD2/SMC1 pathway [5]. Ubiquitin-dependent degradation of the CDC25A phosphatase and the consequent inhibition of cyclin E/CDK2 kinase activity are the main features of the first of the intra-S-phase checkpoint pathways mentioned above. The second pathway is presented in Fig. 8. It involves the Fanconi anaemia protein, FANCD2 and the MRN complex, in particular, the NBS protein. In response to DNA strand breakage induced by ionising radiation, ATM phosphorylates the NBS protein. Phosphorylation of NBS is required for FANCD2 phosphorylation by ATM at serine 222 (S222), and ultimately for activation of the intra-S-phase checkpoint response [5]. Similarly, SMC1 phosphorylation by ATM at serine residues S957 and S966 requires functional NBS [18].

A relatively new actor among S phase regulatory factors is the multiple BRCT-domain protein TopBP1. It functions in the initiation of DNA replication and participates in checkpoint responses in S phase (review in [13]). This protein is phosphorylated by ATM after irradiation [55]. It contains eight BRCT motifs and upon infliction of DNA damage it interacts with the S phase specific transcription factor E2F1. The interaction depends on the N-terminus of E2F1 and the sixth BRCT domain of TopBP1. Moreover, in cells with damaged DNA, TopBP1 recruits E2F1 to repair foci [24].

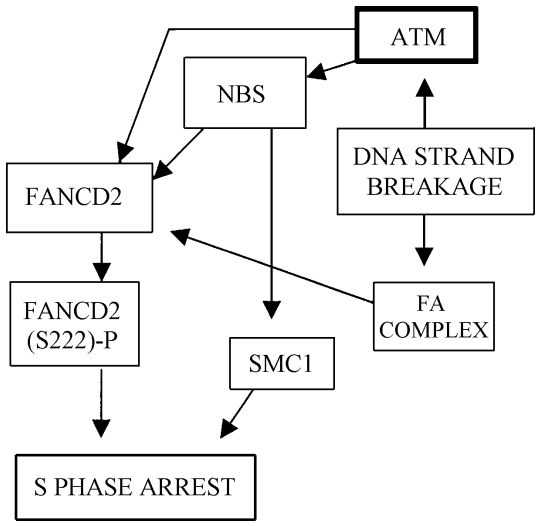

Fig. 8. Signalling related to the intra-S-phase checkpoint – the ATM–NBS–FANCD2/SMC1 pathway. In response to DNA strand breakage, ATM phosphorylates the NBS protein. Phosphorylation of NBS is required for FANCD2 phosphorylation by ATM at serine 222 (S222), and ultimately for activation of the intra-S-phase checkpoint response. Similarly, SMC1 phosphorylation by ATM at serine residues S957 and S966 requires functional NBS (based on [18] and the review of Bartek *et al*. [5]).

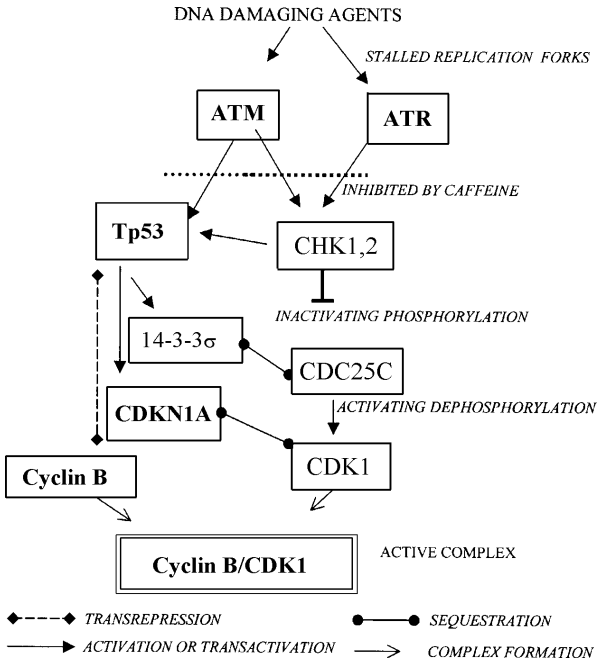

Fig. 9. Signalling involved in G2 arrest in cells with damaged DNA (based on the data from [15, 16, 26, 35]. See the list of abbreviations for the names of the participating proteins.

The G2 checkpoint

The G2 arrest is an important defence mechanism, since it prevents cell division when DNA integrity is compromised. The main signalling is initiated by PI3K-like kinases, as shown in Fig. 9. The diagram presents the sequestration of phosphatase CDC25 by cytoplasmic 14-3-3s protein, and that of kinase CDK1 by the inhibitor CDKN1A (p21). Additionally, Tp53 represses the synthesis of cyclin B. The most important mechanism of the G2 arrest is inhibition of CDK1 kinase in complex with cyclin B. Its activity depends on dephosphorylation of tyrosine 15. This tyrosine residue stays phosphorylated as long as the ATM and ATR kinases signal the presence of DNA breaks or other lesions. Activated CHK kinases phosphorylate the CDC25C phosphatase, thus inhibiting it and favouring its sequestration by 14-3-3 protein in the cytoplasm. A detailed review, also including the role of RAD17 and the 9-1-1 complex can be found in [26, 27, 35].

The essential role of adaptor proteins in amplification of the damage signal

The sole presence of wild type ATM kinase is sufficient for the key event in DSB detection and generation of the damage signal, that is, dissociation of the ATM dimer and activation of kinase. However, the generated damage signal is too weak to evoke a sufficient response. Furthermore, the protein substrates of ATM need to be adequately placed for efficient phosphorylation. For example, mutations in MRE11 result in the radiosensitivity disorder called ataxia-telangiectasia-like disorder (ATLD). Cells from ATLD patients are deficient in activation of ATM and phosphorylation of

downstream ATM targets following irradiation [8]. A weaker activation of ATM reflected in defective check-point control also is noted in the absence of RAD17 and the 9-1-1 complex [3, 4]. According to Kitagawa *et al*. [18], NBS and BRCA1 are required for the recruitment of previously activated ATM to the sites of DNA breaks after irradiation.

The features of the genome surveillance system presented so far give some notion on the degree of its complication both in the number of components and the necessity of co-ordination of their actions. These logistic problems are solved by the cell owing to self-assembly of components in repair foci and to specialised functions of adaptor proteins that bring together substrates and enzymes or serve as platforms for binding several proteins in adequate steric locations. Such a platform is the BRCA1 protein which assembles several proteins of the genome surveillance system listed in Table 2 and known as BASC (BRCA1-associated genome surveillance complex). As discussed below, also MDC1/NFBD1 functions as a platform in the self-assembly of protein components of the repair foci.

Specific structural features of the interacting proteins are critical for the proper self-assembly of repair foci components and the perfect co-ordination of their function. BRCA1 carboxyl-terminal (BRCT) and forkhead-associated (FHA) domains are present in a number of repair and checkpoint proteins, suggesting their role in co-ordination of the response to DNA damage. There are several proteins with C-terminal tandem BRCT domains in the human proteome – among them: BRCA1, 53BP1, MDC1/NFBD1, TopBP1 and microcephalin (MCPH1) [24, 52–54]. All of them are involved in DNA damage response, localise to repair foci after X or γ irradiation and the first four are phosphorylated by ATM in response to DNA strand breakage. Analysis of the tandem BRCT domain structure lead to the conclusion that it functions as a phosphoserine- or phosphothreonine-specific binding module that recognises proteins phosphorylated by ATM or ATR in response to ionising radiation and brings them into repair foci [29].

In the last 3 years, special importance of adaptor proteins 53BP1 and MDC1/NFBD1 and their interaction have been revealed (reviewed by Stucki and Jackson [43, 44]). Within minutes after DSB generation by exposure to ionising radiation, 53BP1 becomes bound to the DSB-containing chromatin region. The assembly of MDC1/NFBD1 at DSB takes place even faster. Change in chromatin conformation due to DNA strand breaks unmasks methylated lysine 79 residues in histone H3 in the vicinity of the break. These residues are recognised and bound by 53BP1. Depletion of MDC1/NFBD1 by small interfering RNA radiosensitises mammalian cells, impairs 53BP1 redistribution to DSB and triggers premature dislocation of 53BP1 from repair foci [6, 43] as well as reduces the binding of NBS in the foci [26, 27]. 53BP1 is required for CHK2 activation and is involved in S and G2/M checkpoint controls [45].

MDC1/NFBD1 contains one FHA domain and two BRCT domains which are involved in protein interactions [42], whereas 53BP1 contains two BRCT domains and tandem Tudor domains [9, 33]. The Tudor domain is an approximately 60-amino acid structure motif forming three beta-stranded core regions with a negatively charged surface. The tandem Tudor domains interact with methylated lysine 79 in histone H3 and this is a critical step in recruiting 53BP1 to the repair foci (review in [43]). The MRN complex which plays the roles of sensor, checkpoint and repair factors, contains a FHA domain and a BRCT domain [19].

The combination of the FHA/BRCT domains is crucial for the binding of nibrin to the H2AX histone. Mutant proteins devoid of these domains prevent foci formation; similar effects are obtained by over-expression of FHA or BRCT-containing truncated MDC1/NFBD1 protein [53, 54]. Over-expressed FHA and BRCT domain-containing fragments of NFBD1/MDC1 abolish ionising radiation-induced foci formation by full length NFBD1/MDC1 protein, MRE11, nibrin, 53BP1, CHK2 phosphorylated at tyrosine 68, phosphorylated H2AX, and putative ATM substrates recognised by anti-phospho-SQ/TQ antibody (SQ/TQ is a serine-glutamine/tyrosine-glutamine motif recognised by PI3K like kinases). These results suggest that MDC1/NFBD1 is a platform that recruits DNA checkpoint signalling and repair proteins to the sites of DNA damage [53].

The reports mentioned above point to the importance of adaptor proteins in the response to DSB induc-

Table 2. Proteins identified so far as components of BASC (BRCA1-associated genom surveillance complex) [47]

Acronim	Full name and function
ATM	Ataxia telangiectasia (AT) mutated, a PI3-K (phosphatidyl inositol 3-kinase) like protein kinase
ATR	ATM-RAD3-related, a PI3-K like kinase
NBS	Nibrin, mutated in Nijmegen chromosome breakage syndrome, in complex with MRE11-RAD50, active in DSB repair and S-phase checkpoint
MRE11, RAD50	Components of the MRN complex (MRE11-RAD50-NBS), active in DSB repair and S-phase checkpoint
BLM	Helicase mutated in Bloom's syndrome
MSH2-6, MLH1	Mismatch repair proteins
BRCA1	Breast cancer susceptibility gene 1
RFC	Replication factor C, acting with DNA polymerase δ

tion. It seems that their main role is damage signal amplification and bringing together the various protein substrates and the respective kinases that are involved in the response. The reader is referred again to the diagram in Fig. 2 which illustrates the main interactions between the most important actors of the genome surveillance system.

Concluding remarks

A characteristic feature of the genome surveillance web is redundancy: the mammalian cell usually has more than one mechanism for specific functions; when one signalling pathway is blocked, e.g. in mutant cells, other "backup" pathways become activated and the cell's defence may be weaker but still allows a fraction of cell population to survive.

The last years brought a wealth of information on relations between malignancy and the function of the genome surveillance system. Apart from the function of control of the cell cycle and its co-ordination with DNA repair processes, the system participates in telomere maintenance and regulation of transcription of some gene groups, in particular those involved in programmed cell death (reviewed in [58]). The complexity of the emerging picture is a challenge for the new generation of radiation biologists whose aim is to understand the cellular response to ionising radiation in molecular terms.

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Abbreviations

9-1-1 – RAD9-RAD1-HUS1 complex,
53BP1 – Tp53 binding protein,
ATLD – ataxia-telangiectasia-like disorder, a disease with the same clinical features as ataxia-telangiectasia but with a milder clinical course, due to defective MRE11,
ATM – ataxia telangiectasia mutated,
ATR – ATM and Rad3 related,
ATRIP – ATR-interacting protein,
BASC – BRCA1-associated genome surveillance complex,
BLM – Bloom syndrome helicase,
BRCA1 – breast cancer susceptibility gene 1,
BRCA2 – breast cancer susceptibility gene 2,
BRCT, BRCA-1 – carboxyl-terminal protein domain,
c-ABL – Abelson oncogene-coded tyrosine kinase E2F, E2F1 transcription factors specific or genes involved in S-phase of the cell cycle,
CDC25A – cell division cycle 25A phosphatase,
CDK – cyclin-dependent protein kinase,
CHK1 – checkpoint protein kinase 1,
CHK2 – checkpoint protein kinase 2,
DNA-PK – DNA-dependent protein kinase,
DSB – DNA double strand break,
FANCD2 – Fanconi anemia complementation group D2,
FHA – forkhead-associated domain,
Gadd45 – growth arrest, DNA damage protein 45,
H2AX – variant of histone H2A, phosphorylated upon DSB infliction,
HDAC – histone deacetylase,
HRR – homologous recombination repair,
HUS1 – hydroxyurea sensitive, yeast *Saccharomyces pombe* checkpoint protein,

IKK – inhibitor kappaB kinase,
MCPH1 – microcephalin, microcephaly, primary autosomal recessive 1,
MDC1 – mediator of DNA damage checkpoint 1,
MDM2 – murine double minute 2,
MLH1 – mutL homologue 1 (bacterial mismatch repair protein),
MRE11 – meiotic-recombination protein-11,
MRN – MRE11-RAD50-NBS complex,
MSH 2-6 – MutS homologues (misrepair proteins) 2-6,
NBS – Nijmegen breakage syndrome (nibrin),
NFBD1 – a "nuclear factor with an amino-terminal FHA domain and a tandem repeat of BRCT domains" also called MDC1 (mediator of DNA damage checkpoint 1),
NHEJ – non-homologous end-joining,
PCNA – proliferating cell nuclear antigen,
PI3K – phosphatidyl inositol-3 kinase RCF, replication factor C,
PIKL – PI3K-like kinases,
RAD1, RAD9, RAD17, RAD51 – human analogues of yeast Rad proteins responsible for ionising radiation resistance,
RPA2 – replication protein A2,
SMC1 – structural maintenance of chromosomes protein 1,
TopBP1 – topoisomerase II beta binding protein 1,
Tp53 – tumor suppressor protein 53,
TRF1 – telomeric repeat binding protein 1.