Published in final edited form as:
Cell Signal. 2013 July ; 25(7): 1625–1634. doi:10.1016/j.cellsig.2013.04.004.

MicroRNA and Signal Transduction Pathways in Tumor Radiation Response

Luqing Zhao[1], **Xiongbin Lu**[2], and **Ya Cao**[1]

[1]Cancer Research Institute, Xiangya School of Medicine, Central South University, Changsha 410078, China

[2]Department of Cancer Biology, The University of Texas, MD Anderson Cancer Center, Houston, TX 77030, USA

Abstract

Tumor radiation response is an essential issue in radiotherapy and a core determining factor of tumor radioresistance or radiosensitivity. Multiple factors can influence tumor radiation response, and among them tumor genetic and epigenetic background, tumor microenvironment and tumor blood flow status may take a leading role. During the whole process of tumor radiation response, tumor radiosensitivity can be regulated in an orderly manner through some classical signal transduction pathways. Although these pathways have already owned multiple biological functions and involved in the process of carcinogenesis, their regulatory roles in tumor radiation response can not be ignored. MicroRNA (miRNA) is a class of non-coding RNA of about 22 nucleotides in length, which binds to the 3'-untranslated region (3'-UTR) of target gene and controls the expression of it at the post-transcriptional level. MiRNA participates in numerous physiology and pathology processes and acts as oncogene or tumor suppressor to affect cancer progression. Through interplaying with the key components in radiation related signal transduction pathways, miRNA could effectively activate the expression of DNA damage response genes and cell cycle related genes in nucleus, and play a critical role in the modulation of radiation response and radiosensitivity in tumor cells. In this review, we mainly elucidate the regulatory mechanisms and functions of miRNA in these radiation related signal transduction pathways from three different aspects which include the upstream receptors, midstream transducer pathways, and downstream effector genes.

Keywords

microRNA (miRNA); signal transduction pathways; tumor radiation response

1. Introduction

MicroRNA (miRNA) is a "multifunctional molecule" which has been heatedly studied in cancer research field nowadays. It is a class of short non-coding RNA, which consists of about 22 nucleotides in length. Through its "seed sequences" (7–8 nucleotides), miRNA

Conflict of interest statement
None declared.

binds to the 3'-UTR of target mRNA and inhibit or block the expression of target gene at the post-transcriptional level [1, 2]. One miRNA has multiple potential target genes, while one gene can be regulated by various miRNAs, thus miRNA and target genes form a complicated regulatory network to affect the gene expression at different levels [3]. Many miRNAs are located at the "fragile sites" of the chromosome, which are frequently deleted in cancer, so the dysregulation of miRNA will lead to the tumorigenesis. Some miRNAs act as the function of oncogene, which are named as "OncomiR"; while, others play a role like tumor suppressor gene, which are called "tumor suppressive miRNA" [4, 5]. Moreover, a new type of "epi-miRNA" which directly targets key enzymatic effectors of the epigenetic machinery (such as DNA methyltransferases, histone deacetylases, and polycomb genes) has come into sight [6]. MiRNA owns multiple functions and is involved in almost every physiology and pathology processes, such as DNA damage response, autophagy, apoptosis, differentiation, metabolism and inflammation [7, 8]. Also, it participates in the initiation and progression of numerous diseases and cancers, and regulates many malignant hallmarks of cancer, which include tumor growth, proliferation, cell cycle checkpoint, genomic instability, metabolism, invasion, metastasis, radiation response and chemoresistance [9, 10].

Radiotherapy is one of the major treatments in various tumors, especially the squamous epithelium originated carcinomas, which owns good therapeutic effects when combined with other treatments like chemotherapy or surgery [11]. The radiation response of tumor is the determining factor of the radiotherapeutic effect. If the radiation response of tumor cells is highly sensitive, the killing effect of radiotherapy will be greatly improved, allowing low dose of ionizing radiation to achieve the same results. Otherwise, if the tumor owns the resistance to radiation or has insensitive radiation response, the outcome of radiotherapy will be quite different. So how to elevate tumor radiation response and improve radiosensitivity has become a core issue in the radiotherapeutic field [12]. It has been reported that four classical signal transduction pathways, including PI3K/AKT, MAPK/ERK, NF-κB, TGF-β, have participated in the regulation of tumor radiation response, and they can be activated either through ionizing radiation or through the receptor tyrosine kinase (RTK) of EGFR and IGFR [13–15]. Among these four pathways, PI3K/AKT, MAPK/ERK, NF-κB pathways are closely related to non-homologous end joining (NHEJ) of DNA damage repair process, while TGF-β pathway is necessary for the full activation of ATM gene and it is involved in both processes of NHEJ and homologous recombination (HR) in DNA damage response [16, 17]. All these pathways will finally affect the expression of crucial genes in nucleus which take part in the processes of DNA damage response (DDR) and cell cycle and apoptosis, including ATM, DNA-PK, NBS1, RAD51, BRCA1, Chk1, Chk2, CDK2, CDC25, BAD, BIM, MCL1 and so on [18–20]. MiRNA plays a critical role in the regulation of tumor radiation response. Through interplaying with the key factors in the radiation related signal transduction pathways, miRNA could modulate tumor radiation response from three different aspects: upstream receptors, midstream pathways and downstream genes, and form a complicated epigenetic-genetic interaction network to influence the expression of radiation related genes [21].

In this review, we mainly focus on the role of miRNA and signal transduction pathways in tumor radiation response, and highlight the critical regulatory role of miRNA in the three layers of these radiation related signal transduction pathways.

2. MiRNA and the upstream receptors of radiation related signal transduction pathways

2.1. Epidermal Growth Factor Receptor (EGFR)

Emerging evidences have demonstrated that the overexpression or the mutation of the EGFR is closely associated with resistance of tumor cells to both chemo- and radiotherapy. It is frequently expressed in tumors of epithelial origin, and is a crucial determinant of tumor responses to ionizing radiation [22]. Owing to the activation of survival or cell proliferation pathways, such as PI3K/AKT, MAPK/ERK pathways, EGFR may confer tumor resistance to radiation [23]. Apart from this mechanism, a novel function of EGFR involved in radiation-induced nuclear translocation and interaction with the DNA-dependent protein kinase (DNA-PK) has been widely acknowledged [24]. DNA-PK is an essential component of NHEJ in DNA repair pathway, which a predominant process for the repair of radiation-induced DNA double-strand breaks (DSBs) [25]. A series of latest reports have indicated that EGFR and its downstream components could modulate NHEJ through directly interplaying with multiple DNA damage repair enzymes [26, 27].

In human cancer cells, over-expression of miR-7 not only down-regulates the expression of EGFR, activates EGFR-associated signaling like the EGFR-PI3K-AKT pathways, but also prolongs the radiation-induced γH2AX formation and reduces the expression of DNA-PKcs [28]. MiR-133 targets the expression of EGFR so as to inhibit tumor cell proliferation, invasion and migration, especially in hormone-independent prostate cancer cell lines [29]. Similarly as miR-133, through targeting the EGFR pathway, miR-146a suppresses tumor growth and progression by inhibiting the expression of MMP2, meanwhile, this process is closely related to the down-regulation of p-ERK expression [30]. Moreover, miR-21 and EGFR consists of a regulatory loop during ionizing radiation. The radiation could stimulate the expression of miR-21 through the EGFR/STAT3 pathway, while the up-regulation of miR-21 can also activate the EGFR pathway and its downstream target genes [31]. All of these regulatory mechanisms between miRNA and EGFR feedback loop reveal the possible molecular basis of tumor radiation response. Targeting the DNA repair function of EGFR or blocking its downstream signaling components may serve as a promising therapeutic approach for sensitizing tumors to radiotherapy.

2.2. Insulin-like Growth Factor Receptor (IGFR)

It is evident that insulin-like growth factor (IGF) signaling is intimately correlated with the function of endocrine system and could tightly regulate glucose and insulin metabolism. Due to the activation of type 1 insulin-like growth factor receptor (IGF-1R), the IGF ligands could stimulate cellular proliferation signals and promote cell growth and development [32, 33]. Also the IGF signaling system is implicated to play a critical role in the pathogenesis of multiple cancers. Besides the role of directly influencing tumor development, IGF-1R also confers the resistance to chemo- or radiotherapy and appears to be an essential determinant of response to various cancer therapies [34, 35]. In the field of tumor radiation response, IGF-1R modulates DSB repair via HR process [36]. Depletion or blockage the expression of IGF-1R will sensitize tumor cells to ionizing radiation and suppress cell cycle-dependent processes, especially the G2/M phase arrest [37]. Therefore, using several drugs such as monoclonal antibodies (mAB), small molecule tyrosine kinase inhibitors (RTKIs), anti-sense oligonucleotids (ASOs) and IGF-binding proteins (IGFBPs) to target the functional binding sites of IGF-1R and inhibit its downstream signaling, will enhance the radiation response and radiosensitivity of tumors [38, 39].

IGFR is reported to be a potential target of miR-7, which is regarded as a tumor suppressor miRNA in multiple tumors. The down-regulation of miR-7 enhances the expression of IGFR

at both mRNA and protein levels [40]. Besides targeting IGFR, miR-7 also targets the substrates of IGFR, such as IRS1 and IRS2, and triggers the PI3K/AKT pathway so as to regulate the survival and proliferation of tumor cells [41]. MiR-223 targets both a-subunit and β -subunit of IGFR in cell membrane, and inhibits the proliferation and growth of tumor cells through suppressing downstream AKT/mTOR/p70S6K signal transduction pathway in cytoplasm. IGFR is just the functional target of miR-223 and the direct target of this miRNA is Rasa1. However, the downstream of this gene doesn't have the exact biological functions, so IGFR and its downstream factors in this pathway take the leading role and worth doing further researches [42].

It has also been demonstrated that miR-375 interacts with IGFR and has strong tumor-suppressive effects through inhibiting tumor colony formation, growth, proliferation, migration and metastasis. The expression of IGFR negatively correlates with miR-375 expression. Meanwhile, the methylation status in the promoter region of miR-375 greatly determines the expression level of this miRNA [43]. Definitely, the genetic and epigenetic regulation in the upstream of miRNA genes effectively influence the biological functions of downstream components and its signal transduction pathways, which also reveals new insights for understanding the mechanisms of tumor initiation and progression [44]. All in all, the essential roles of miRNA played in the IGFR-induced signal transduction pathways will provide effective therapeutic targets to improve tumor radiation response.

3. MiRNA and the midstream transducers of radiation related signal transduction pathways

3.1. PI3K/AKT pathway

PI3K/AKT signaling pathway constitutes an essential pathway and regulates various biological processes such as cell growth, proliferation, apoptosis, invasion and metabolism [45]. The activation of AKT could further modulate the function of numerous substrates involved in the regulation of cell survival and cell cycle progression [46]. It has also been reported that the components in this pathway are frequently altered in human cancers and may decisively contribute to the chemo- or radiotherapy resistant phenotype [47]. PI3K/AKT pathway is closely associated with three major radiation resistance mechanisms, which are the tumor intrinsic radiosensitivity, tumor cell proliferation ability, and the hypoxia microenvironment [48]. Effectively inhibition the activity of PI3K and its downstream component mammalian target of rapamycin (mTOR) will help to maintain the DNA damage status and increase the numbers of γH2AX foci, together with the enhanced G2 phase cell cycle delay after the ionizing radiation treatment [49].

To be specific, miR-7 is shown to act as a tumor suppressor and regulate the PI3K/AKT pathway through targeting key molecules in this pathway, such as PIK3CD, mTOR and p70S6K, so as to inhibit tumorigenesis and reverse the metastasis process of tumor cells [50]. The miR-17–92 cluster targets protein phosphatase PHLPP2, a negative regulator of PI3K/AKT pathway, accompanied with another two targets PTEN and BIM. Over-expression of this miRNA activates the PI3K/AKT pathway, while down-regulation of miR-17–92 inhibits the function of this pathway, suppresses tumor growth and affects the chemoresistance [51]. MiR-126 directly targets p85 beta, regulates PI3K signaling and influences the phosphorylated AKT expression in order to suppress tumor growth [52]. Meanwhile, there are still some other miRNAs exist which could target the key factors in PI3K/AKT pathway. MiR-221 and miR-222 directly target PTEN gene through their seed sequences, and efficiently affect downstream biological processes of tumors such as cell growth, invasion, migration and radiosensitivity via regulating the expression of PTEN [53]. Besides, miR-486 can also directly targets PTEN and Foxo1a, which are two crucial

negative regulators in PI3K/AKT signaling pathway, and inhibits the translation of these two genes, leading to the phosphorylation of AKT so as to activate the AKT signaling. On the other hand, the activation of AKT results in the phosphorylation of GSK3β and the inhibition of Foxo1a activity, so the function of PI3K/AKT pathway can be greatly suppressed. This negative feedback loop initiating by miR-486 helps to modulate the expression of various genes in the PI3K/AKT pathway [54]. Thus, directly targeting the roles of miRNA and suppressing the activation of PI3K/AKT signaling pathway may become a valid approach to treat human malignancies and overcome the resistance of radiation therapy, finally increasing tumor radiosensitivity and providing novel targets for radiosensitization and drug discovery.

3.2. MAPK/ERK pathway

The mitogen-activated protein kinases (MAPK)/extracellular-signal-regulated kinases (ERK) pathway is known to play important roles in diverse cellular events, including cell growth, proliferation, apoptosis, differentiation and senescence [55]. Depending on the regulation of various growth factor receptors, the activation of this pathway can be simultaneously induced by ionizing radiation or a variety of other toxic stresses [56]. Meanwhile, this pathway is essential for the transmission of cellular signals through transduction systems (ligands, transmembrane receptors and cytoplasmic secondary messengers) to the nucleus, where the expressions of multiple downstream target genes can be greatly influenced [57]. Inflicted by ionizing radiation, MAPK pathway could activate the downstream of death receptors and procaspases, and DNA damage signals, such as the JNK, p38 MAPK and NF-κB pathways [58]. Moreover, through MAPK signaling, ionizing radiation can induce the initiation of EGFR-ERK signaling and upregulate the expression of DNA repair genes XRCC1 and ERCC1 in an ERK1/2-dependent fashion. Decreasing the activity of ERK1/2 could lead to an increase in XRCC1/ERCC1 expression. Besides, inhibition of the function of ERK will led to the persistence of apurinic/apyrimidinic (AP) sites of DNA damage so as to increase cellular killing effect [59]. Obviously, a complex control of DNA damage repair process may be largely dependent on the activation of MAPK/ERK1/2 signaling [60].

It has been reported that four miRNAs, including miR-7-3, miR-34a, miR-181d, and miR-193b, are closely associated with MAPK activity. When MAPK is activated, the expression of miR-7-3 is further up-regulated, while the other three miRNAs expression are down-regulated in the same way. Over-expression of MAPK-associated miRNAs inhibit the proliferation of tumor cells, among them miR-193b leads the most obvious suppressive role. All of these miRNAs inhibit the expression of multiple downstream target genes, along with the activation of upstream MAPK activity, in order to form a complicated regulatory network of MAPK pathway and play a significant role in numerous tumor malignance behaviors [61]. MiR-17-5p is usually over-expressed and acts as an oncogene in tumor, in order to effectively activate the p38 MAPK pathway and elevate the phosphorylation level of heat shock protein 27 (HSP27). A new signal transduction pathway “miR-17-5p--- p38--- HSP27” has been established to regulate tumor proliferation and migration [62]. Through interacting with the p38 MAPK pathway, miR-17-5p owns a potential clinical application in anti-cancer therapies.

MiR-21 takes an essential part in various aspects of carcinogenesis and is involved in the ERK/NF-κB signal pathways. Once the expression of miR-21 is up-regulated, this pathway will be activated. MiR-21 has a feedback effect in regulating ERK activity, and the activation degree of ERK/NF-κB pathway is largely depended on the ROS level [63]. This phenomenon indicates that miR-21 may be closely related to the tumor metabolic states and has the potential to become a metabolism-associated target to affect tumor progression. Additionally, miR-21 can directly target PTEN gene, leading to the activation of AKT and

ERK 1/2 signaling pathway, and enhance the expression of HIF-1α and VEGF, so as to induce tumor angiogenesis and further promote tumor growth and metastasis [64]. Hence, blocking the functional exertion of miRNA and interfering its relationship with the MAPK/ERK signaling pathway, will help to better control of tumor radiation response and exploit effective strategies to improve radiotherapeutic effects.

3.3. NF-κB pathway

Transcription factor NF-κB and its downstream pathway are frequently activated in tumor cells and contribute to aggressive tumor growth, proliferation, apoptosis and the resistance to chemotherapy or ionizing radiation during cancer treatment [65]. Inhibition of the activity of this transcription factor will increase the sensitivity of tumor cells to the apoptotic action of chemotherapeutic agents and to radiation exposure [66]. Mounting evidences indicate that poly (ADP-ribose) polymerase-1 (PARP-1) activity is essential in the upstream regulation of ionizing radiation (IR) induced NF-κB activation and sensitizes cancer cells to IR-induced cellular killing, together with the inhibited XIAP expression and the increased caspase-3 activity [67]. Besides, it has been demonstrated that ataxia telangiectasia mutated (ATM) has a critical role in the activation of NF-κB following the DNA damage and in response to numerous genotoxic stresses. Meanwhile, NF-κB is found to be defective in cells from patients with A–T (ataxia-telangiectasia) who are highly sensitive to DNA damage induced by ionizing radiation [68]. Also, ATM itself is a key regulator of the cellular response to DSB and a sensor protein to other DNA damaging agents in a manner of activating a wide variety of effectors involved in multiple signaling pathways and controlling cell cycle checkpoints, apoptosis and DNA repair processes [69]. Blocking the expressions of both ATM and NF-κB will contribute to the increased tumor sensitivity to DSB and radiation response [70].

As we all known that NF-κB consists of three subunits, p50, p65 and IκB, and recent reports have implicated that miRNA could regulate the expression status of any of these subunits [71, 72]. MiR-31 negatively modulates the function of NF-κB signal pathway through targeting NF-κB inducing kinase (NIK). The loss of miR-31 expression triggers the activation of this oncogenic signaling pathway. MiR-31 expression level can also be controlled by the upstream polycomb protein in an epigenetic model. Owing to this epigenetic modulation, the upregulation of polycomb protein leads to the downregulation of miR-31, resulting in the activation of NF-κB pathway and influencing multiple downstream biological functions such as proliferation, apoptosis, and inflammatory responses [73].

IκB is a most crucial factor for the activation of NF-κB, as IκB can tightly bind to NF-κB in the cytoplasm in order to prevent it from translocating into the nucleus to further activate the expressions of downstream genes [74]. MiR-30e* directly targets IκB and suppresses its expression, which leads to the hyper-activation of NF-κB and promotes the expression of NF-κB-regulated genes so as to elevate the invasiveness and metastasis ability of tumor cells. The miR-30e*-mediated constitutive activation of NF-κB pathway is largely depended on the disruption of NF-κB/IκB negative feedback loop in an epigenetic mechanism, which can finally let tumor gain an aggressive phenotype [75].

Besides, miR-301a down-regulates the expression of NF-κB repressing factor (NKRF) in order to promote the activation of NF-κB. On the other hand, NF-κB could also regulate the activities of the promoter regions of miR-301a and elevate its transcriptional levels accordingly. A positive feedback loop of miR-301a and NF-κB is formed to maintain the persistence of NF-κB activation, which indicates a new mechanism for the post-transcriptional regulation of NF-κB activity [76]. In sum, identification of the molecular regulatory mechanisms of miRNA in the NF-κB signaling will lead to a better

understanding of tumor radiation response and provide novel promising targets and potential therapeutic approaches to overcome radioresistance in cancer treatment (Figure 1).

3.4. TGF-β pathway

Transforming growth factor (TGF)-β signaling plays a critical role in several different biological processes involving cell growth, differentiation, apoptosis, motility, angiogenesis, invasion, epithelial mesenchymal transition (EMT), extracellular matrix production and cellular immune responses [77]. This pathway is also quite essential in tumorigenesis and demonstrates paradoxical actions. During the early phase, TGF-β signaling acts as a tumor suppressor and function to suppress tumor progression, exemplified by deletions or mutations in the core components of the TGF-β signaling pathway. On the contrary, it can also act as a pro-metastatic pathway and facilitate malignant transformation such as cell invasion, dissemination, and immune evasion in the late-stages [78]. Accumulating evidences show that the function of TGF-β signaling is closely correlated with the activities of SMAD family, such as SMAD4 and SMAD7 [79]. Mutation or blockage of TGF-β type II receptor (TβRII) and SMAD4 expressions can help to overcome the growth promoting effects of TGF-β and loss its responsiveness to growth suppression [80]. Besides, TGF-β signaling is a good modifier of radiation responses and plays an important role in protecting cells from radiation [81]. Inducing the function of TGF-β receptor or constitutively activating SMAD family will reduce DNA fragmentation, Caspase-3 cleavage and γH2AX foci formation in irradiated cells [82]. Meanwhile, owing to the protection of TGF-β under radiation, there will be a decreased DNA damage, reduced apoptosis and thereby an enhanced cell survival rate [83]. Moreover, during the DDR process, the roles of SMAD proteins can have crosstalk between TGF-β and ATM pathways. SMAD2 and SMAD7 respectively contribute to IR-induced DSB signaling in an ATM or TGF-β receptor 1 (TGFβR1) dependent manner [84].

The latest studies have shown that miRNA is involved in the regulation of TGF-β mediated signal transduction pathways. MiR-520/373 family acts as tumor suppressors in tumors by affecting the function of NF-κB and TGF-β pathways, and it further leads to the malignance characteristics of tumor through promoting growth, proliferation, metastasis, inflammation and progression. MiR-520c is closely correlated with the stages of lymph node metastasis, so targeting this miRNA offers us effective measures to prevent tumor from metastasizing [85]. MiR-21 is a key “OncomiR” in the regulatory process of EMT mediated by TGF-β, and this miRNA is closely related to the functional exertion of TGF-β signaling pathways [86]. After exposed in TGF-β, the expression of miR-21 is up-regulated and it further affects the ability of epithelial cells so as to have a potential impact on epithelium homeostasis and tumorigenesis [87]. MiR-17–92 cluster inhibits cell proliferation and regulates collagen synthesis through TGF-β pathway by directly targeting downstream genes such as TGFBR2, SMAD2 and SMAD4, which greatly pave the way to study the function of palatal mesenchymal cells and promote the normal palatal development [88].

MiR-181 also has a close relationship with the TGF-β pathway, and the expression level of miR-181 can be elevated by TGF-β at the post-transcriptional level [89]. The expression of ATM, a direct target of miR-181, is interfered by TGF-β exposure. Through this interaction between miRNA and the TGF-β signaling pathway, the properties of cancer stem cells can be regulated and the stem cell-like features can be controlled [90]. Moreover, miR-106b-25 cluster directly targets SMAD7, increases the levels of TGF-β type 1 receptor and further activates the downstream of the TGF-β signaling. It induces an EMT transition and a tumor initiating-cell like phenotype, which are both required for the activation of downstream Six1 gene. This crucial correlation between miR-106b-25 cluster, Six1 and the TGF-β signaling, offers a new molecular mechanism for us to modulate tumor malignance phenotype and shift tumor aggressive behavior [91]. Thus, elucidating the functional interactions between

miRNA and the TGF-β signaling may offer a sound rationale for sensitizing tumor cells to radiation and help to tailor appropriate adjuvant radiotherapeutic strategies to improve the treatment of solid tumors.

4. MiRNA and the downstream effector genes of radiation related signal transduction pathways

4.1. DNA damage response genes

In the downstream of four radiation related signal transduction pathways, majorities of nucleus genes are involved in the DDR process, which mainly include ATM, DNA-PK, BRCA1, NBS1, RAD51 and so on [92–94]. DDR is an essential process during the ionizing radiation, which can promote faithful transmission of genomes in dividing cells by reversing the extrinsic and intrinsic DNA damage and is indispensable in cell survival during DNA replication [95]. Accordingly, cells have evolved diverse DDR pathways to monitor the integrity of their genome. Specifically, DDR pathways contain three major components: sensors, signal transducers, and effectors. H2AX and NBS1 act as the core sensors and initiate the beginning step of DDR [96, 97]. At the level of transducers, ATM and ATR (ATM-Rad3-related) are proximal kinases in the central of the entire DDR and can be used to detect various forms of damaged DNA and trigger the downstream DDR cascades [98, 99]. Moreover, numerous factors play an important role as effectors in the DDR pathways, including DNA-PK, BRCA1, BRCA2, RAD51, RAD52, Chk1, Chk2, p53, and are separately involved in the regulation of multiple biological processes covering two types of DNA damage repair (NHEJ and HR), cell cycle checkpoint and apoptosis control [100–104] (Figure 2). In addition, poly (ADP-ribose) polymerase (PARP-1) is another crucial effector in DDR pathways and responsible for cellular survival [105, 106]. Using the inhibitor of PARP-1 demonstrates selectively killing effects on the cells with defects of HR, particularly in the context of BRCA1/2 mutations. Successfully inhibiting the expression of PARP-1 in BRCA1/2 deficient tumors will effectively prevent cancer progression [107, 108]. The PARP inhibitor-induced HR is abolished in ATM, but not DNA-PK, inhibited cells. Also, ATM is activated following the inhibition of PARP and may function in the upstream of HR in order to repair certain types of DSB [109]. Besides PARP-1, adopting other inhibitors to target major components of the DDR, such as ATM, ATR, DNA-PK, Chk1 and Chk2, can also confer radio- and/or chemosensitivity in cancer cells [110].

Meanwhile, the expression levels of above components and their downstream factors are also regulated by miRNAs. MiR-18a directly targets ATM gene and down-regulates its expression so as to affect DNA damage repair ability and HR efficiency of DSBs. Owing to this regulatory model, the phorsphorylation levels and nuclear foci formations of ATM downstream substrates H2AX and 53BP1 are greatly changed, which finally contributes to the radiation response and radiosensitivity of tumor cells [111]. Some studies further indicate that miR-101 could target the 3’-UTR regions of DNA-PK and ATM mRNA, and further efficiently reduces the expression of these genes in order to influence both NHEJ and HR processes in DNA damage repair and sensitize tumor cells to radiation at the same time [112]. Apart from ATM and DNA-PK, BRCA1 is another essential tumor suppressor gene which plays an essential role in DNA damage repair pathway [113]. BRCA1 could repress the expression of miR-155 in an epigenetic model through regulating the expression of HDAC2 and deacetylating histones H2A and H3 which are in the promoter region of miR-155. This phenomenon reveals the intimate relationship between miR-155 and BRCA1, suggesting that miR-155 can be used to treat BRCA1-associated tumors and modulate DDR or radiosensitivity of some tumors [114]. To sums up, deeply exploring the potential regulatory mechanisms between miRNA and DNA damage response genes, will open new avenues to investigate into tumor radiation response and help to find novel and promising

targets in clinical trials to improve tumor radiosensitivity and elevate radiotherapeutic effects.

4.2. Cell cycle and apoptosis related genes

The cell cycle and apoptosis related genes, such as Chk1, Chk2, CDK2, CDC25, Cyclin E, BAD, BIM, and MCL1, are also under the regulation of four radiation related signal transduction pathways. Interfering with the process of cell cycle and apoptosis contributes to the tumor radiation response either [115, 116]. Cell cycle checkpoints are pivotal for safeguarding genome stability, and if they are defect in cells, there will be an increasingly rate of genome instability and neoplastic transformation. Besides their role of maintaining genomic stability or implementing cell cycle arrest, the cell cycle checkpoint signaling also mediates the recruitment of DNA repair pathways. When the transducers like ATM and ATR in DDR pathways gain their functions, the downstream cell cycle checkpoint effector kinases termed Chk1 and Chk2 will be further activated accordingly [117, 118]. The final effect of DDR pathway can be divided into two major branches. One is the ATM/Chk2 pathway that is activated after DSBs in response to IR, the other is the ATR/Chk1 pathway which responds primarily to DNA single strand breaks or bulky lesions and is modified by ATR in response to replication inhibition and UV-induced damage [119, 120]. So the Chk2 and Chk1 might have been involved in channeling the DNA damage signal from ATM and ATR, respectively. However, these two parallel branches of the DDR pathway also show a high degree of crosstalk and connectivity. Both pathways will converge on CDC25, a positive regulator of cell cycle progression, which is inhibited by Chk1-mediated or Chk2-mediated phosphorylation [121]. Thus, an ATM/ATR-Chk2/Chk1-CDC25-CDK axis will be established to underlie the molecular basis of the replication checkpoint, the intra-S phase checkpoint, and the G2 DNA damage checkpoint [122]. Furthermore, the proto-oncogene c-Myc has been reported to regulate tumor radioresistance through transcriptional activation of Chk1 and Chk2 checkpoint kinases by direct binding to the Chk1 and Chk2 promoters. Inhibition of the c-Myc-Chk1/Chk2 pathway could regulate DDR checkpoints and stem cell characteristics, and reveal potential therapeutic applications in reversal of DDR processes and tumor radioresistance [123]. Therefore, using small inhibitors to target the expression of Chk1 and Chk2 will effectively disturb the progression of cell cycle checkpoint and suppress tumor radiation response in DDR pathways.

Mounting evidences have shown that the expressions of various cell cycle and apoptosis related genes are greatly affected by the activities of miRNAs in the radiation related signal transduction pathways. MiR-25 regulates the function of intrinsic apoptosis pathway by directly targeting the expression of pro-apoptotic protein BIM, and indirectly modulating other two pro-apoptotic proteins BAX and caspase-3 so as to inhibit apoptosis process. The inverse relationship between miR-25 and BIM, indicates that miR-25 can effectively interfere apoptosis process and prevent cancer progression [124]. In addition, miR-29b directly targets MCL1 mRNA, promotes the activation of caspase-3 and further induces apoptosis process [125]. MiR-29a is also found to modulate apoptosis through inhibiting MCL1 expression. The down-regulation of miR-29a greatly contributes to the over-expression of MCL1, an anti-apoptotic protein, which promotes tumor cell survival through suppressing apoptosis process [126]. Moreover, miR-193b negatively regulates the expression of MCL1 in order to disturb apoptosis process and inhibit tumor cell proliferation. The down-regulation of miR-193b can be regarded as an early event in tumor progression, and it will help to do early detection and treatment of tumors [127]. To conclude, these findings suggest that miRNA has an intimate relationship with the cell cycle and apoptosis related genes and plays a critical role in tumor radiation response. Making best use of miRNAs as novel diagnostic markers and therapeutic targets will greatly improve tumor radiotherapeutic effects.

5. Conclusion

Tumor radiation response is a major factor in determining radiotherapeutic effect and closely related to tumor radiosensitivity and radioresistance. Numerous biological processes are participated in the regulation of tumor radiation response, including the DNA damage response and repair, cell cycle checkpoint and apoptosis control, and microenvironment and metabolism reprogramming. Interestingly, some classical multifunctional signal transduction pathways have recently been found to play an essential role in modulating tumor radiation response. Among them, four signal transduction pathways, such as the PI3K/AKT, MAPK/ERK, NF-κB, and TGF-β pathways, have attracted our attention. MiRNA is a class of small non-coding RNA that modulates tumorigenesis from different aspects and has multiple clinical prospects in tumor diagnosis, treatment and prognosis. In this review, we extensively explore the critical regulatory role of miRNA in the radiation related signal transduction pathways, and illustrate their interactions with key components in these pathways from three different aspects, which are indicated in the upstream receptors, midstream pathways and downstream target genes (Figure 3 and Table 1). We successfully find that through interplaying with crucial factors in the radiation related pathways, the biological functions of miRNAs are critical enough to determine the radiation response and radiosensitivity of tumor cells. Deeply studying the potential mechanisms of miRNA in the radiation related signal transduction pathways, will not only offer a new insight to regulate tumor radiosensitivity at the post-transcriptional level, but also provide various novel diagnostic markers and therapeutic targets to improve the efficacy of radiotherapy. Furthermore, with the development of translational medicine which advocates the transition from “bench to bedside”, the clinical application of miRNA in tumor radiation response will finally bring more hope and gospel for tumor patients.

Acknowledgments

This work was supported by the National Key Basic Research Program of China (2011CB504300), the Key Project of National Natural Science Foundation of China (30930101), the Hunan Provincial Innovation Foundation for Postgraduate (CX2012A005), and the Scholarship Award for Excellent Doctoral Student granted by Ministry of Education of China, the National Institutes of Health of American (R01CA136549) and the American Cancer Society (119135-RSG-10-185-01-TBE).

References

1. van Kouwenhove M, Kedde M, Agami R. MicroRNA regulation by RNA-binding proteins and its implications for cancer. Nat Rev Cancer. 2011; 11:644–656. [PubMed: 21822212]
2. Ryan BM, Robles AI, Harris CC. Genetic variation in microRNA networks: the implications for cancer research. Nat Rev Cancer. 2010; 10:389–402. [PubMed: 20495573]
3. Esteller M. Non-coding RNAs in human disease. Nat Rev Genet. 2011; 12:861–874. [PubMed: 22094949]
4. Kasinski AL, Slack FJ. MicroRNAs en route to the clinic: progress in validating and targeting microRNAs for cancer therapy. Nat Rev Cancer. 2011; 11:849–864. [PubMed: 22113163]
5. Iorio MV, Croce CM. microRNA involvement in human cancer. Carcinogenesis. 2012; 33:1126–1133. [PubMed: 22491715]
6. Fabbri M, Calore F, Paone A, Galli R, Calin GA. Epigenetic regulation of miRNAs in cancer. Adv Exp Med Biol. 2013; 754:137–148. [PubMed: 22956499]
7. Mendell JT, Olson EN. MicroRNAs in stress signaling and human disease. Cell. 2012; 148:1172–1187. [PubMed: 22424228]
8. Abdellatif M. Differential expression of microRNAs in different disease states. Circ Res. 2012; 110:638–650. [PubMed: 22343558]
9. Lovat F, Valeri N, Croce CM. MicroRNAs in the pathogenesis of cancer. Semin Oncol. 2011; 38:724–733. [PubMed: 22082758]

10. Liu X, Liu L, Xu Q, Wu P, Zuo X, Ji A. MicroRNA as a novel drug target for cancer therapy. Expert Opin Biol Ther. 2012; 12:573–580. [PubMed: 22428844]
11. Joubert A, Vogin G, Devic C, Granzotto A, Viau M, Maalouf M, Thomas C, Colin C, Foray N. Radiation biology: major advances and perspectives for radiotherapy. Cancer Radiother. 2011; 15:348–354. [PubMed: 21683640]
12. Czarnota GJ, Karshafian R, Burns PN, Wong S, Al Mahrouki A, Lee JW, Caissie A, Tran W, Kim C, Furukawa M, Wong E, Giles A. Tumor radiation response enhancement by acoustical stimulation of the vasculature. Proc Natl Acad Sci U S A. 2012; 109:E2033–E2041. [PubMed: 22778441]
13. Begg AC, Stewart FA, Vens C. Strategies to improve radiotherapy with targeted drugs. Nat Rev Cancer. 2011; 11:239–253. [PubMed: 21430696]
14. Pickhard AC, Margraf J, Knopf A, Stark T, Piontek G, Beck C, Boulesteix AL, Scherer EQ, Pigorsch S, Schlegel J, Arnold W, Reiter R. Inhibition of radiation induced migration of human head and neck squamous cell carcinoma cells by blocking of EGF receptor pathways. BMC Cancer. 2011; 11:388. [PubMed: 21896192]
15. Matsumoto F, Valdecanas DN, Mason KA, Milas L, Ang KK, Raju U. The impact of timing of EGFR and IGF-1R inhibition for sensitizing head and neck cancer to radiation. Anticancer Res. 2012; 32:3029–3035. [PubMed: 22843870]
16. Wu L, Shao L, Li M, Zheng J, Wang J, Feng W, Chang J, Wang Y, Hauer-Jensen M, Zhou D. BMS-345541 Sensitizes MCF-7 Breast Cancer Cells to Ionizing Radiation by Selective Inhibition of Homologous Recombinational Repair of DNA Double-Strand Breaks. Radiat Res. 2012
17. Mladenov E, Iliakis G. Induction and repair of DNA double strand breaks: the increasing spectrum of non-homologousend joining pathways. Mutat Res. 2011; 711:61–72. [PubMed: 21329706]
18. Lord CJ, Ashworth A. The DNA damage response and cancer therapy. Nature. 2012; 481:287–294. [PubMed: 22258607]
19. Hazawa M, Hosokawa Y, Monzen S, Yoshino H, Kashiwakura I. Regulation of DNA damage response and cell cycle in radiation-resistant HL60 myeloid leukemia cells. Oncol Rep. 2012; 28:55–61. [PubMed: 22576796]
20. Mérino D, Strasser A, Bouillet P. Bim must be able to engage all pro-survival Bcl-2 family members for efficient tumor suppression. Oncogene. 2012; 31:3392–3396. [PubMed: 22081075]
21. Zhao L, Bode AM, Cao Y, Dong Z. Regulatory mechanisms and clinical perspectives of miRNA in tumor radiosensitivity. Carcinogenesis. 2012; 33:2220–2227. [PubMed: 22798379]
22. Liccardi G, Hartley JA, Hochhauser D. EGFR nuclear translocation modulates DNA repair following cisplatin and ionizing radiation treatment. Cancer Res. 2011; 71:1103–1114. [PubMed: 21266349]
23. Li P, Zhang Q, Torossian A, Li ZB, Xu WC, Lu B, Fu S. Simultaneous Inhibition of EGFR and PI3K Enhances Radiosensitivity In Human Breast Cancer. Int J Radiat Oncol Biol Phys. 2012; 83:e391–e397. [PubMed: 22414288]
24. Dittmann K, Mayer C, Kehlbach R, Rodemann HP. Radiation-induced caveolin-1 associated EGFR internalization is linked with nuclear EGFR transport and activation of DNA-PK. Mol Cancer. 2008; 7:69. [PubMed: 18789131]
25. Davis AJ, Lee KJ, Chen DJ. The amino-terminal region of the DNA-dependent protein kinase catalytic subunit (DNA-PKcs) is required for its DNA double-strand break-mediated activation. J Biol Chem. 2013
26. Mukherjee B, Choy H, Nirodi C, Burma S. Targeting nonhomologous end-joining through epidermal growth factor receptor inhibition: rationale and strategies for radiosensitization. Semin Radiat Oncol. 2010; 20:250–257. [PubMed: 20832017]
27. Kriegs M, Kasten-Pisula U, Rieckmann T, Holst K, Saker J, Dahm-Daphi J, Dikomey E. The epidermal growth factor receptor modulates DNA double-strand break repair by regulating non-homologous end-joining. DNA Repair (Amst). 2010; 9:889–897. [PubMed: 20615764]
28. Lee KM, Choi EJ, Kim IA. microRNA-7 increases radiosensitivity of human cancer cells with activated EGFR-associated signaling. Radiother Oncol. 2011; 101:171–176. [PubMed: 21676478]

29. Tao J, Wu D, Xu B, Qian W, Li P, Lu Q, Yin C, Zhang W. microRNA-133 inhibits cell proliferation migration and invasion in prostate cancer cells by targeting the epidermal growth factor receptor. Oncol Rep. 2012; 27:1967–1975. [PubMed: 22407299]
30. Xu B, Wang N, Wang X, Tong N, Shao N, Tao J, Li P, Niu X, Feng N, Zhang L, Hua L, Wang Z, Chen M. MiR-146a suppresses tumor growth and progression by targeting EGFR pathway and in a p-ERK-dependent manner in castration-resistant prostate cancer. Prostate. 2012; 72:1171–1178. [PubMed: 22161865]
31. Shi Y, Zhang X, Tang X, Wang P, Wang H, Wang Y. MiR-21 is continually elevated long-term in the brain after exposure to ionizing radiation. Radiat Res. 2012; 177:124–128. [PubMed: 22034847]
32. Yee D. Insulin-like growth factor receptor inhibitors: baby or the bathwater? J Natl Cancer Inst. 2012; 104:975–981. [PubMed: 22761272]
33. Bortvedt SF, Lund PK. Insulin-like growth factor 1: common mediator of multiple enterotrophic hormones and growth factors. Curr Opin Gastroenterol. 2012; 28:89–98. [PubMed: 22241077]
34. Tognon CE, Sorensen PH. Targeting the insulin-like growth factor 1 receptor (IGF1R) signaling pathway for cancer therapy. Expert Opin Ther Targets. 2012; 16:33–48. [PubMed: 22239439]
35. Xue M, Cao X, Zhong Y, Kuang D, Liu X, Zhao Z, Li H. Insulin-like growth factor-1 receptor (IGF-1R) kinase inhibitors in cancer therapy: advances and perspectives. Curr Pharm Des. 2012; 18:2901–2913. [PubMed: 22571659]
36. Valenciano A, Henríquez-Hernández LA, Moreno M, Lloret M, Lara PC. Role of IGF-1 receptor in radiation response. Transl Oncol. 2012; 5:1–9. [PubMed: 22348170]
37. Casa AJ, Dearth RK, Litzenburger BC, Lee AV, Cui X. The type I insulin-like growth factor receptor pathway: a key player in cancer therapeutic resistance. Front Biosci. 2008; 13:3273–3287. [PubMed: 18508432]
38. Arnaldez FI, Helman LJ. Targeting the insulin growth factor receptor 1. Hematol Oncol Clin North Am. 2012; 26:527–542. vii-viii. [PubMed: 22520978]
39. Zaidi SH, Huddart RA, Harrington KJ. Novel targeted radiosensitisers in cancer treatment. Curr Drug Discov Technol. 2009; 6:103–134. [PubMed: 19519337]
40. Jiang L, Liu X, Chen Z, Jin Y, Heidbreder CE, Kolokythas A, Wang A, Dai Y, Zhou X. MicroRNA-7 targets IGF1R (insulin-like growth factor 1 receptor) in tongue squamous cell carcinoma cells. Biochem J. 2010; 432:199–205. [PubMed: 20819078]
41. Sun Z, Shushanov S, LeRoith D, Wood TL. Decreased IGF type 1 receptor signaling in mammary epithelium during pregnancy leads to reduced proliferation, alveolar differentiation, and expression of insulin receptor substrate (IRS)-1 and IRS-2. Endocrinology. 2011; 152:3233–3245. [PubMed: 21628386]
42. Jia CY, Li HH, Zhu XC, Dong YW, Fu D, Zhao QL, W Wu, Wu XZ. MiR-223 suppresses cell proliferation by targeting IGF-1R. PLoS One. 2011; 6:e27008. [PubMed: 22073238]
43. Kong KL, Kwong DL, Chan TH, Law SY, Chen L, Li Y, Qin YR, Guan XY. MicroRNA-375 inhibits tumour growth and metastasis in oesophageal squamous cell carcinoma through repressing insulin-like growth factor 1 receptor. Gut. 2012; 61:33–42. [PubMed: 21813472]
44. Banno K, Kisu I, Yanokura M, Masuda K, Kobayashi Y, Ueki A, Tsuji K, Yamagami W, Nomura H, Susumu N, Aoki D. Endometrial Cancer and Hypermethylation: Regulation of DNA and MicroRNA by Epigenetics. Biochem Res Int. 2012; 2012:738–274.
45. Grunt TW, Mariani GL. Targeting the PI3K/AKT/mTOR Pathway in Breast Cancer. Curr Cancer Drug Targets. 2012
46. Martelli AM, Tabellini G, Bressanin D, Ognibene A, Goto K, Cocco L, Evangelisti C. The emerging multiple roles of nuclear Akt. Biochim Biophys Acta. 2012; 1823:2168–2178. [PubMed: 22960641]
47. Kim TR, Cho EW, Paik SG, Kim IG. Hypoxia-induced SM22α in A549 cells activates the IGF1R/PI3K/Akt pathway, conferring cellular resistance against chemo- and radiation therapy. FEBS Lett. 2012; 586:303–309. [PubMed: 22245152]
48. Zhan M, Han ZC. Phosphatidylinositide 3-kinase/AKT in radiation responses. Histol Histopathol. 2004; 19:915–923. [PubMed: 15168354]

49. Fokas E, Yoshimura M, Prevo R, Higgins G, Hackl W, Maira SM, Bernhard EJ, McKenna WG, Muschel RJ. NVP-BEZ235 and NVP-BGT226, dual phosphatidylinositol 3-kinase/mammalian target of rapamycin inhibitors, enhance tumor and endothelial cell radiosensitivity. Radiat Oncol. 2012; 7:48. [PubMed: 22452803]
50. Fang YX, Xue JL, Shen Q, Chen J, Tian L. miR-7 inhibits tumor growth and metastasis by targeting the PI3K/AKT pathway in hepatocellular carcinoma. Hepatology. 2012; 55:1852–1862. [PubMed: 22234835]
51. Rao E, Jiang C, Ji M, Huang X, Iqbal J, Lenz G, Wright G, Staudt LM, Zhao Y, McKeithan TW, Chan WC, Fu K. The miRNA-17~92 cluster mediates chemoresistance and enhances tumor growth in mantle cell lymphoma via PI3K/AKT pathway activation. Leukemia. 2012; 26:1064–1072. [PubMed: 22116552]
52. Guo C, Sah JF, Beard L, Willson JK, Markowitz SD, Guda K. The noncoding RNA, miR-126, suppresses the growth of neoplastic cells by targeting phosphatidylinositol 3-kinase signaling and is frequently lost in colon cancers. Genes Chromosomes Cancer. 2008; 47:939–946. [PubMed: 18663744]
53. Chun-Zhi Z, Lei H, An-Ling Z, Yan-Chao F, Xiao Y, Guang-Xiu W, Zhi-Fan J, Pei-Yu P, Qing-Yu Z, Chun-Sheng K. MicroRNA-221 and microRNA-222 regulate gastric carcinoma cell proliferation and radioresistance by targeting PTEN. BMC Cancer. 2010; 10:367. [PubMed: 20618998]
54. Small EM, O'Rourke JR, Moresi V, Sutherland LB, McAnally J, Gerard RD, Richardson JA, Olson EN. Regulation of PI3-kinase/Akt signaling by muscle-enriched microRNA-486. Proc Natl Acad Sci U S A. 2010; 107:4218–4223. [PubMed: 20142475]
55. Lyng FM, Maguire P, McClean B, Seymour C, Mothersill C. The involvement of calcium and MAP kinase signaling pathways in the production of radiation-induced bystander effects. Radiat Res. 2006; 165:400–409. [PubMed: 16579652]
56. Liang X, So YH, Cui J, Ma K, Xu X, Zhao Y, Cai L, Li W. The low-dose ionizing radiation stimulates cell proliferation via activation of the MAPK/ERK pathway in rat cultured mesenchymal stem cells. J Radiat Res. 2011; 52:380–386. [PubMed: 21436606]
57. Brzezianska E, Pastuszak-Lewandoska D. A minireview: the role of MAPK/ERK and PI3K/Akt pathways in thyroid follicular cell-derived neoplasm. Front Biosci. 2011; 16:422–439. [PubMed: 21196179]
58. Dent P, Yacoub A, Fisher PB, Hagan MP, Grant S. MAPK pathways in radiation responses. Oncogene. 2003; 22:5885–5896. [PubMed: 12947395]
59. Yacoub A, McKinstry R, Hinman D, Chung T, Dent P, Hagan MP. Epidermal growth factor and ionizing radiation up-regulate the DNA repair genes XRCC1 and ERCC1 in DU145 and LNCaP prostate carcinoma through MAPK signaling. Radiat Res. 2003; 159:439–452. [PubMed: 12643788]
60. Golding SE, Rosenberg E, Neill S, Dent P, Povirk LF, Valerie K. Extracellular signal-related kinase positively regulates ataxia telangiectasia mutated, homologous recombinationrepair, and the DNA damage response. Cancer Res. 2007; 67:1046–1053. [PubMed: 17283137]
61. Ikeda Y, Tanji E, Makino N, Kawata S, Furukawa T. MicroRNAs associated with mitogen-activated protein kinase in human pancreatic cancer. Mol Cancer Res. 2012; 10:259–269. [PubMed: 22188669]
62. Yang F, Yin Y, Wang F, Wang Y, Zhang L, Tang Y, Sun S. miR-17-5p Promotes migration of human hepatocellular carcinoma cells through the p38 mitogen-activated protein kinase-heat shock protein 27 pathway. Hepatology. 2010; 51:1614–1623. [PubMed: 20209605]
63. Ling M, Li Y, Xu Y, Pang Y, Shen L, Jiang R, Zhao Y, Yang X, Zhang J, Zhou J, Wang X, Liu Q. Regulation of miRNA-21 by reactive oxygen species-activated ERK/NF-κB in arsenite-induced cell transformation. Free Radic Biol Med. 2012; 52:1508–1518. [PubMed: 22387281]
64. Liu LZ, Li C, Chen Q, Jing Y, Carpenter R, Jiang Y, Kung HF, Lai L, Jiang BH. MiR-21 induced angiogenesis through AKT and ERK activation and HIF-1α expression. PLoS One. 2011; 6:e19139. [PubMed: 21544242]
65. Perkins ND. The diverse and complex roles of NF-κB subunits in cancer. Nat Rev Cancer. 2012; 12:121–132. [PubMed: 22257950]

66. Li F, Sethi G. Targeting transcription factor NF-kappaB to overcome chemoresistance and radioresistance in cancer therapy. Biochim Biophys Acta. 2010; 1805:167–180. [PubMed: 20079806]

67. Veuger SJ, Hunter JE, Durkacz BW. Ionizing radiation-induced NF-kappaB activation requires PARP-1 function to confer radioresistance. Oncogene. 2009; 28:832–842. [PubMed: 19060926]

68. Ahmed KM, Li JJ. ATM-NF-kappaB connection as a target for tumor radiosensitization. Curr Cancer Drug Targets. 2007; 7:335–342. [PubMed: 17979628]

69. Bensimon A, Aebersold R, Shiloh Y. Beyond ATM: the protein kinase landscape of the DNA damage response. FEBS Lett. 2011; 585:1625–1639. [PubMed: 21570395]

70. Veuger SJ, Durkacz BW. Persistence of unrepaired DNA double strand breaks caused by inhibition of ATM does not lead to radio-sensitisation in the absence of NF-κB activation. DNA Repair (Amst). 2011; 10:235–244. [PubMed: 21144805]

71. Vaz C, Mer AS, Bhattacharya A, Ramaswamy R. MicroRNAs modulate the dynamics of the NF-κB signaling pathway. PLoS One. 2011; 6:e27774. [PubMed: 22114691]

72. Ma X, Becker Buscaglia LE, Barker JR, Li Y. MicroRNAs in NF-kappaB signaling. J Mol Cell Biol. 2011; 3:159–166. [PubMed: 21502305]

73. Yamagishi M, Nakano K, Miyake A, Yamochi T, Kagami Y, Tsutsumi A, Matsuda Y, Sato-Otsubo A, Muto S, Utsunomiya A, Yamaguchi K, Uchimaru K, Ogawa S, Watanabe T. Polycomb-mediated loss of miR-31 activates NIK-dependent NF-κB pathway in adult T cell leukemia and other cancers. Cancer Cell. 2012; 21:121–135. [PubMed: 22264793]

74. Dyson HJ, Komives EA. Role of disorder in IκB-NFκB interaction. IUBMB Life. 2012; 64:499–505. [PubMed: 22573609]

75. Jiang L, Lin C, Song L, Wu J, Chen B, Ying Z, Fang L, Yan X, He M, Li J, Li M. MicroRNA-30e* promotes human glioma cell invasiveness in an orthotopic xenotransplantation model by disrupting the NF-κB/IκBα negative feedback loop. J Clin Invest. 2012; 122:33–47. [PubMed: 22156201]

76. Lu Z, Li Y, Takwi A, Li B, Zhang J, Conklin DJ, Young KH, Martin R, Li Y. miR-301a as an NF-κB activator in pancreatic cancer cells. EMBO J. 2011; 30:57–67. [PubMed: 21113131]

77. Kelly RJ, Morris JC. Transforming growth factor-beta: a target for cancer therapy. J Immunotoxicol. 2010; 7:15–26. [PubMed: 19916703]

78. Korpal M, Kang Y. Targeting the transforming growth factor-beta signalling pathway in metastatic cancer. Eur J Cancer. 2010; 46:1232–1240. [PubMed: 20307969]

79. Hneino M, François A, Buard V, Tarlet G, Abderrahmani R, Blirando K, Hoodless PA, Benderitter M, Milliat F. The TGF-β/Smad repressor TG-interacting factor 1 (TGIF1) plays a role in radiation-induced intestinal injury independently of a Smad signaling pathway. PLoS One. 2012; 7:e35672. [PubMed: 22567107]

80. Dancea HC, Shareef MM, Ahmed MM. Role of Radiation-induced TGF-beta Signaling in Cancer Therapy. Mol Cell Pharmacol. 2009; 1:44–56. [PubMed: 20336170]

81. Biswas S, Guix M, Rinehart C, Dugger TC, Chytil A, Moses HL, Freeman ML, Arteaga CL. Inhibition of TGF-beta with neutralizing antibodies prevents radiation-induced acceleration of metastatic cancer progression. J Clin Invest. 2007; 117:1305–1313. [PubMed: 17415413]

82. An YS, Kim MR, Lee SS, Lee YS, Chung E, Song JY, Lee J, Yi JY. TGF-β signaling plays an important role in resisting γ-irradiation. Exp Cell Res. 2013; 319:466–473. [PubMed: 23262026]

83. Zhang M, Kleber S, Röhrich M, Timke C, Han N, Tuettenberg J, Martin-Villalba A, Debus J, Peschke P, Wirkner U, Lahn M, Huber PE. Blockade of TGF-β signaling by the TGFβR-I kinase inhibitor LY2109761 enhances radiation response and prolongs survival in glioblastoma. Cancer Res. 2011; 71:7155–7167. [PubMed: 22006998]

84. Wang M, Saha J, Hada M, Anderson JA, Pluth JM, O'Neill P, Cucinotta FA. Novel Smad proteins localize to IR-induced double-strand breaks: interplay between TGFβ and ATM pathways. Nucleic Acids Res. 2013; 41:933–942. [PubMed: 23221633]

85. Keklikoglou I, Koerner C, Schmidt C, Zhang JD, Heckmann D, Shavinskaya A, Allgayer H, Gückel B, Fehm T, Schneeweiss A, Sahin O, Wiemann S, Tschulena U. MicroRNA-520/373 family functions as a tumor suppressor in estrogen receptor negativebreast cancer by targeting NF-κB and TGF-β signaling pathways. Oncogene. 2012; 31:4150–4163. [PubMed: 22158050]

86. Kumarswamy R, Volkmann I, Jazbutyte V, Dangwal S, Park DH, Thum T. Transforming growth factor-β-induced endothelial-to-mesenchymal transition is partly mediated by microRNA-21. Arterioscler Thromb Vasc Biol. 2012; 32:361–369. [PubMed: 22095988]
87. Wang T, Zhang L, Shi C, Sun H, Wang J, R Li, Zou Z, Ran X, Su Y. TGF-β-induced miR-21 negatively regulates the antiproliferative activity but has no effect on EMT of TGF-β in HaCaT cells. Int J Biochem Cell Biol. 2012; 44:366–376. [PubMed: 22119803]
88. Li L, Shi JY, Zhu GQ, Shi B. MiR-17–92 cluster regulates cell proliferation and collagen synthesis by targeting TGFB pathway in mouse palatal mesenchymal cells. J Cell Biochem. 2012; 113:1235–1244. [PubMed: 22095742]
89. Wang B, Hsu SH, Majumder S, Kutay H, Huang W, Jacob ST, Ghoshal K. TGFbeta-mediated upregulation of hepatic miR-181b promotes hepatocarcinogenesis by targeting TIMP3. Oncogene. 2010; 29:1787–1797. [PubMed: 20023698]
90. Wang Y, Yu Y, Tsuyada A, Ren X, Wu X, Stubblefield K, Rankin-Gee EK, Wang SE. Transforming growth factor-β regulates the sphere-initiating stem cell-like feature in breast cancer through miRNA-181 and ATM. Oncogene. 2011; 30:1470–1480. [PubMed: 21102523]
91. Smith AL, Iwanaga R, Drasin DJ, Micalizzi DS, Vartuli RL, Tan AC, Ford HL. The miR-106b-25 cluster targets Smad7, activates TGF-β signaling, and induces EMT and tumor initiating cell characteristics downstream of Six1 in human breast cancer. Oncogene. 2012; 31:5162–5171. [PubMed: 22286770]
92. Basu B, Yap TA, Molife LR, de Bono JS. Targeting the DNA damage response in oncology: pastpresent and future perspectives. Curr Opin Oncol. 2012; 24:316–324. [PubMed: 22476188]
93. Furgason JM, Bahassi EM. Targeting DNA repair mechanisms in cancer. Pharmacol Ther. 2012
94. Bolderson E, Richard DJ, Zhou BB, Khanna KK. Recent advances in cancer therapy targeting proteins involved in DNA double-strand break repair. Clin Cancer Res. 2009; 15:6314–6320. [PubMed: 19808869]
95. Gonfloni S. Targeting DNA damage response: Threshold, chromatin landscape and beyond. Pharmacol Ther. 2013
96. Ivashkevich A, Redon CE, Nakamura AJ, Martin RF, Martin OA. Use of the γ-H2AX assay to monitor DNA damage and repair in translational cancer research. Cancer Lett. 2012; 327:123–133. [PubMed: 22198208]
97. Riches LC, Lynch AM, Gooderham NJ. Early events in the mammalian response to DNA double-strand breaks. Mutagenesis. 2008; 23:331–339. [PubMed: 18644834]
98. López-Contreras AJ, Fernandez-Capetillo O. The ATR barrier to replication-born DNA damage. DNA Repair (Amst). 2010; 9:1249–1255. [PubMed: 21036674]
99. Flynn RL, Zou L. ATR: a master conductor of cellular responses to DNA replication stress. Trends Biochem Sci. 2011; 36:133–140. [PubMed: 20947357]
100. Escribano-Díaz C, Orthwein A, Fradet-Turcotte A, Xing M, Young JT, Tkáč J, Cook MA, Rosebrock AP, Munro M, Canny MD, Xu D, Durocher D. A Cell Cycle-Dependent Regulatory Circuit Composed of 53BP1-RIF1 and BRCA1-CtIP Controls DNA Repair Pathway Choice. Mol Cell. 2013
101. Bergs JW, Krawczyk PM, Borovski T, Ten Cate R, Rodermond HM, Stap J, Medema JP, Haveman J, Essers J, van Bree C, Stalpers LJ, Kanaar R, Aten JA, Franken NA. Inhibition of homologous recombination by hyperthermia shunts early double strand break repair to non-homologous end-joining. DNA Repair (Amst). 2013; 12:38–45. [PubMed: 23237939]
102. Chapman JR, Taylor MR, Boulton SJ. Playing the end game: DNA double-strand break repair pathway choice. Mol Cell. 2012; 47:497–510. [PubMed: 22920291]
103. Laulier C, Lopez BS. The secret life of Bcl-2: apoptosis-independent inhibition of DNA repair by Bcl-2 family members. Mutat Res. 2012; 751:247–257. [PubMed: 22677530]
104. Redwood AB, Gonzalez-Suarez I, Gonzalo S. Regulating the levels of key factors in cell cycle and DNA repair: new pathways revealed by lamins. Cell Cycle. 2011; 10:3652–3657. [PubMed: 22045204]
105. Sousa FG, Matuo R, Soares DG, Escargueil AE, Henriques JA, Larsen AK, Saffi J. PARPs and the DNA damage response. Carcinogenesis. 2012; 33:1433–1440. [PubMed: 22431722]

106. Langelier MF, Pascal JM. PARP-1 mechanism for coupling DNA damage detection to poly(ADP-ribose) synthesis. Curr Opin Struct Biol. 2013

107. Fong PC, Yap TA, Boss DS, Carden CP, Mergui-Roelvink M, Gourley C, De Greve J, Lubinski J, Shanley S, Messiou C, A'Hern R, Tutt A, Ashworth A, Stone J, Carmichael J, Schellens JH, de Bono JS, Kaye SB. Poly(ADP)-ribose polymerase inhibition: frequent durable responses in BRCA carrier ovarian cancer correlating with platinum-free interval. J Clin Oncol. 2010; 28:2512–2519. [PubMed: 20406929]

108. Do K, Chen A. Molecular Pathways: Targeting PARP in Cancer Treatment. Clin Cancer Res. 2012

109. Patel AG, Sarkaria JN, Kaufmann SH. Nonhomologous end joining drives poly (ADP-ribose) polymerase (PARP) inhibitor lethality in homologous recombination-deficient cells. Proc Natl Acad Sci U S A. 2011; 108:3406–3411. [PubMed: 21300883]

110. Sun X, Yang C, Liu H, Wang Q, Wu SX, Li X, Xie T, Brinkman KL, Teh BS, Butler EB, Xu B, Zheng S. Identification and characterization of a small inhibitory peptide that can target DNA-PKcs autophosphorylation and increase tumor radiosensitivity. Int J Radiat Oncol Biol Phys. 2012; 84:1212–1219. [PubMed: 22592045]

111. Song L, Lin C, Wu Z, Gong H, Zeng Y, Wu J, Li M, Li J. miR-18a impairs DNA damage response through downregulation of ataxia telangiectasia mutated (ATM) kinase. PLoS One. 2011; 6:e25454. [PubMed: 21980462]

112. Yan D, Ng WL, Zhang X, Wang P, Zhang Z, Mo YY, Mao H, Hao C, Olson JJ, Curran WJ, Wang Y. Targeting DNA-PKcs and ATM with miR-101 sensitizes tumors to radiation. PLoS One. 2010; 5:e11397. [PubMed: 20617180]

113. Chapman JR, Sossick AJ, Boulton SJ, Jackson SP. BRCA1-associated exclusion of 53BP1 from DNA damage sites underlies temporal control of DNA repair. J Cell Sci. 2012; 125:3529–3534. [PubMed: 22553214]

114. Chang S, Wang RH, Akagi K, Kim KA, Martin BK, Cavallone L, Haines DC, Basik M, Mai P, Poggi E, Isaacs C, Looi LM, Mun KS, Greene MH, Byers SW, Teo SH, Deng CX, Sharan SK. Kathleen Cuningham Foundation Consortium for Research into Familial Breast Cancer (kConFab). Tumor suppressor BRCA1 epigenetically controls oncogenic microRNA-155. Nat Med. 2011; 17:1275–1282. [PubMed: 21946536]

115. Wohlbold L, Merrick KA, De S, Amat R, Kim JH, Larochelle S, Allen JJ, Zhang C, Shokat KM, Petrini JH, Fisher RP. Chemical genetics reveals a specific requirement for Cdk2 activity in the DNA damage response and identifies Nbs1 as a Cdk2 substrate in human cells. PLoS Genet. 2012; 8 e1002935.

116. Gogineni VR, Nalla AK, Gupta R, Dinh DH, Klopfenstein JD, Rao JS. Chk2-mediated G2/M cell cycle arrest maintains radiation resistance in malignant meningioma cells. Cancer Lett. 2011; 313:64–75. [PubMed: 21945852]

117. Pabla N, Bhatt K, Dong Z. Checkpoint kinase 1 (Chk1)-short is a splice variant and endogenous inhibitor of Chk1 that regulates cell cycle and DNA damage checkpoints. Proc Natl Acad Sci U S A. 2012; 109:197–202. [PubMed: 22184239]

118. Reinhardt HC, Yaffe MB. Kinases that control the cell cycle in response to DNA damage: Chk1, Chk2, and MK2. Curr Opin Cell Biol. 2009; 21:245–255. [PubMed: 19230643]

119. Smith J, Tho LM, Xu N, Gillespie DA. The ATM-Chk2 and ATR-Chk1 pathways in DNA damage signaling and cancer. Adv Cancer Res. 2010; 108:73–112. [PubMed: 21034966]

120. Squatrito M, Brennan CW, Helmy K, Huse JT, Petrini JH, Holland EC. Loss of ATM/Chk2/p53 pathway components accelerates tumor development and contributes to radiation resistance in gliomas. Cancer Cell. 2010; 18:619–629. [PubMed: 21156285]

121. Thanasoula M, Escandell JM, Suwaki N, Tarsounas M. ATM/ATR checkpoint activation downregulates CDC25C to prevent mitotic entry with uncapped telomeres. EMBO J. 2012; 31:3398–3410. [PubMed: 22842784]

122. Chen Y, Poon RY. The multiple checkpoint functions of CHK1 and CHK2 in maintenance of genome stability. Front Biosci. 2008; 13:5016–5029. [PubMed: 18508566]

123. Wang WJ, Wu SP, Liu JB, Shi YS, Huang X, Zhang QB, Yao KT. MYC Regulation of CHK1 and CHK2 Promotes Radioresistance in a Stem Cell-like Population of Nasopharyngeal Carcinoma Cells. Cancer Res. 2013; 73:1219–1231. [PubMed: 23269272]

124. Zhang H, Zuo Z, Lu X, Wang L, Wang H, Zhu Z. MiR-25 regulates apoptosis by targeting Bim in human ovarian cancer. Oncol Rep. 2012; 27:594–598. [PubMed: 22076535]

125. Zhang YK, Wang H, Leng Y, Li ZL, Yang YF, Xiao FJ, Li QF, Chen XQ, Wang LS. Overexpression of microRNA-29b induces apoptosis of multiple myeloma cells through down regulating Mcl-1. Biochem Biophys Res Commun. 2011; 414:233–239. [PubMed: 21951844]

126. Desjobert C, Renalier MH, Bergalet J, Dejean E, Joseph N, Kruczynski A, Soulier J, Espinos E, Meggetto F, Cavaillé J, Delsol G, Lamant L. MiR-29a down-regulation in ALK-positive anaplastic large cell lymphomas contributes to apoptosis blockade through MCL-1 overexpression. Blood. 2011; 117:6627–6637. [PubMed: 21471522]

127. Chen J, Zhang X, Lentz C, Abi-Daoud M, Paré GC, Yang X, Feilotter HE, Tron VA. miR-193b Regulates Mcl-1 in Melanoma. Am J Pathol. 2011; 179:2162–2168. [PubMed: 21893020]

Highlights

- Tumor radiation response is a core determining factor of tumor radiosensitivity.
- miRNA plays a critical role in the modulation of tumor radiation response.
- miRNA interplays with the radiation related signal transduction pathways.
- The regulatory mechanism of these pathways will be elucidated from three aspects.

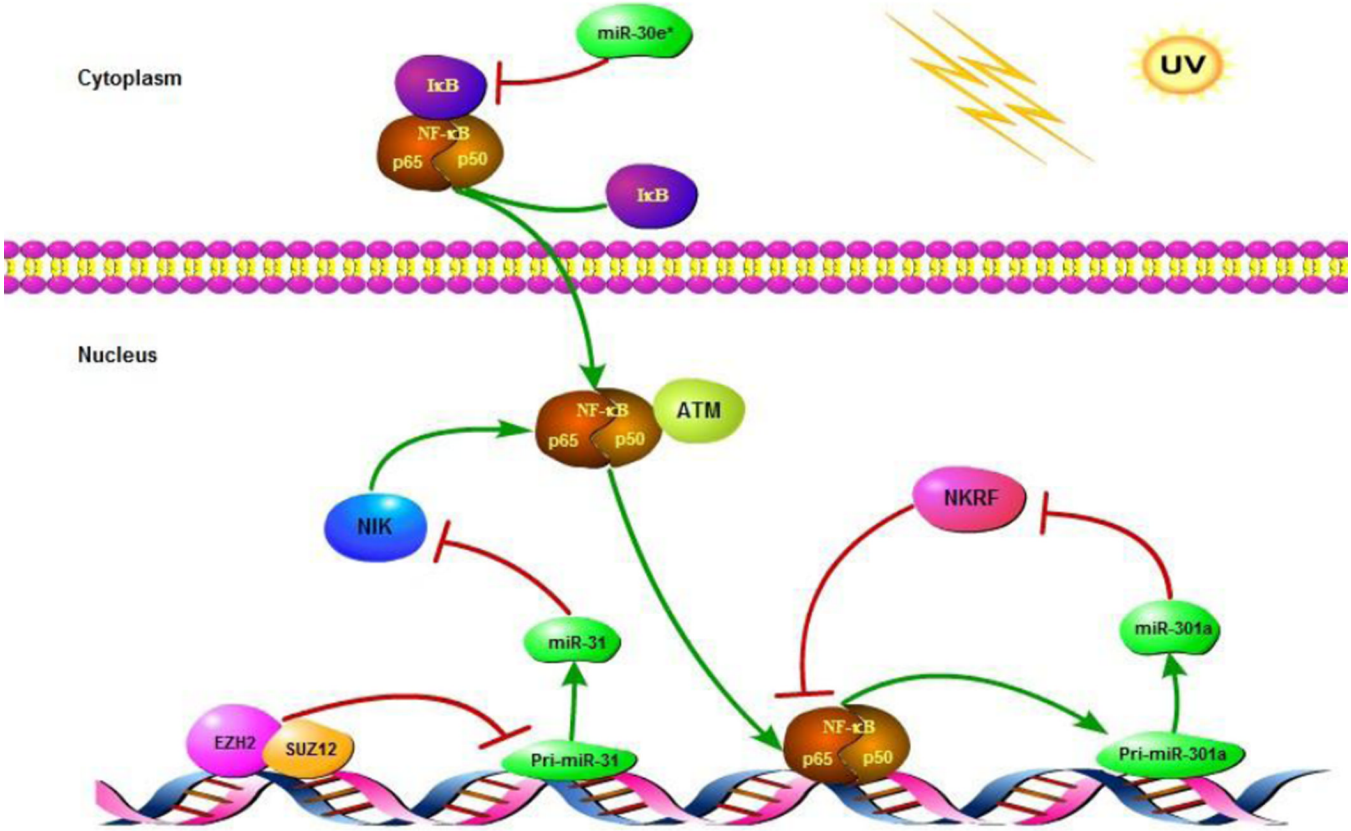

Figure 1. NF-κB and miRNA feedback regulatory loop

NF-κB consists of three subunits, p50, p65 and IκB. IκB is a negative regulator of NF-κB activity, as it can tightly bind to the other two subunits of NF-κB in the cytoplasm and prevent them from translocating into the nucleus. miR-30e* directly targets IκB and suppresses its expression, which leads to the hyper-activation of NF-κB and promotes its translocation into the nucleus to regulate the expression of downstream genes. So a NF-κB/IκB negative feedback loop is formed by miR-30e* to constitutively activate the NF-κB pathway. Besides, polycomb protein (EZH2, SUZ12) can epigenetically down-regulate the expression of miR-31 gene. Through targeting NF-κB inducing kinase (NIK), miR-31 negatively modulates the function of NF-κB signal pathway. On the other hand, NF-κB acts as a transcription factor, binding to the promoter region of miR-301a, and elevates its expression level accordingly. Moreover, miR-301a directly targets NF-κB repressing factor (NKRF) and further promotes the activation of NF-κB. Thus, a positive feedback loop of miR-301a, NKRF and NF-κB is formed to maintain the persistence of NF-κB activation. During the DNA damage response induced by ionizing radiation or UV lights, ATM interplays with NF-κB in the nucleus and plays a critical role in the activation of NF-κB signaling pathway.

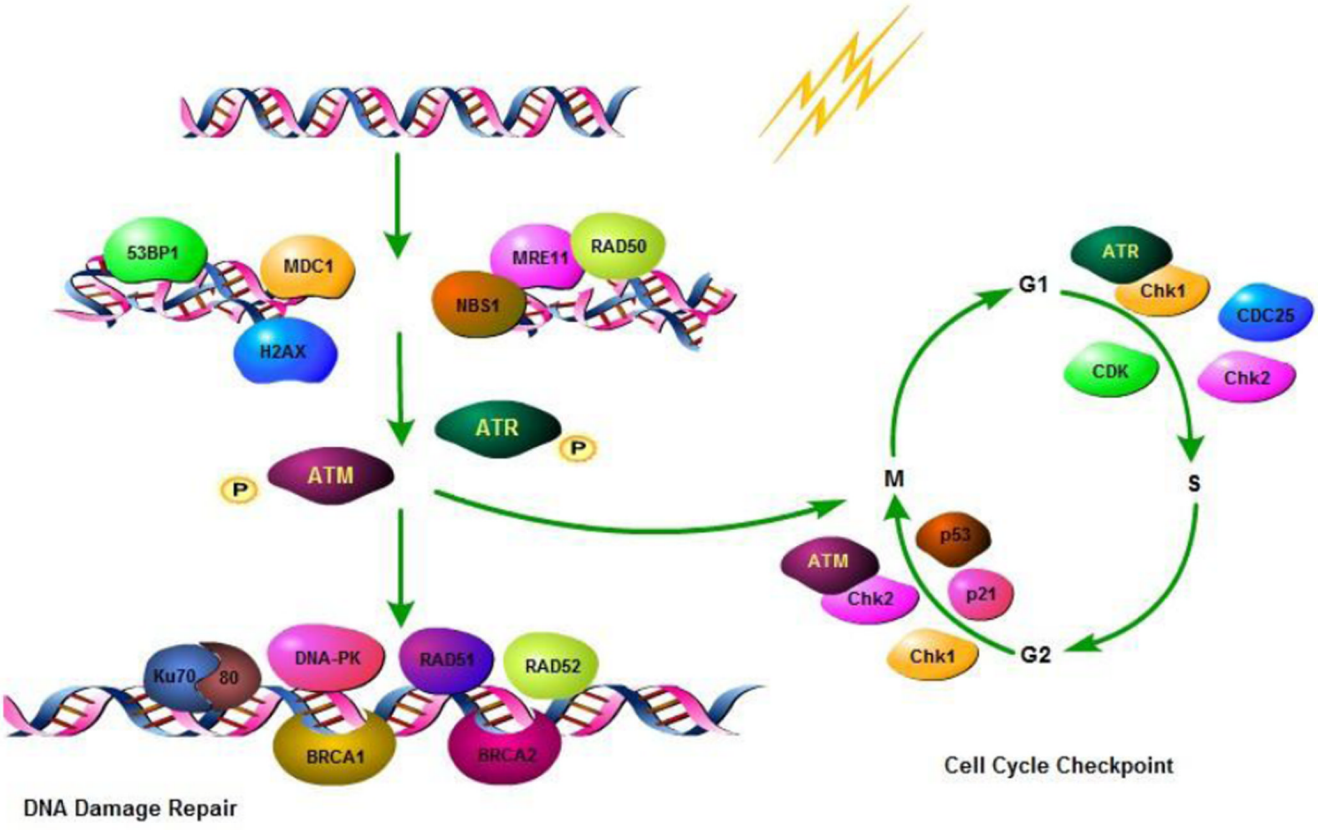

Figure 2. Overview of DNA damage response process in nucleus

When tumor cell is inflicted with ionizing radiation, the double strands of DNA are broken and the DNA damage response (DDR) pathway is further initiated. This pathway contains three major components: sensors, transducers and effectors. H2AX, MDC1, 53BP1 and MRE11-RAD50-NBS1 complex act as the core sensors and could directly bind to the damaged ends of DNA fragments to start the beginning of DDR. At the level of transducers, ATM and ATR are proximal kinases in the central of the entire DDR and could recruit multiple downstream effectors, including DNA-PK, Ku70/80, BRCA1, BRCA2, RAD51, RAD52, in order to complete two types of DNA damage repair (NHEJ and HR) processes. Meanwhile, ATM and ATR can separately interplay with Chk2 and Chk1, and activate other cell cycle regulators like CDC25, CDK, p53, p21, so as to disturb the function of cell cycle checkpoint and interfere with the G1/S and G2/M phase progression.

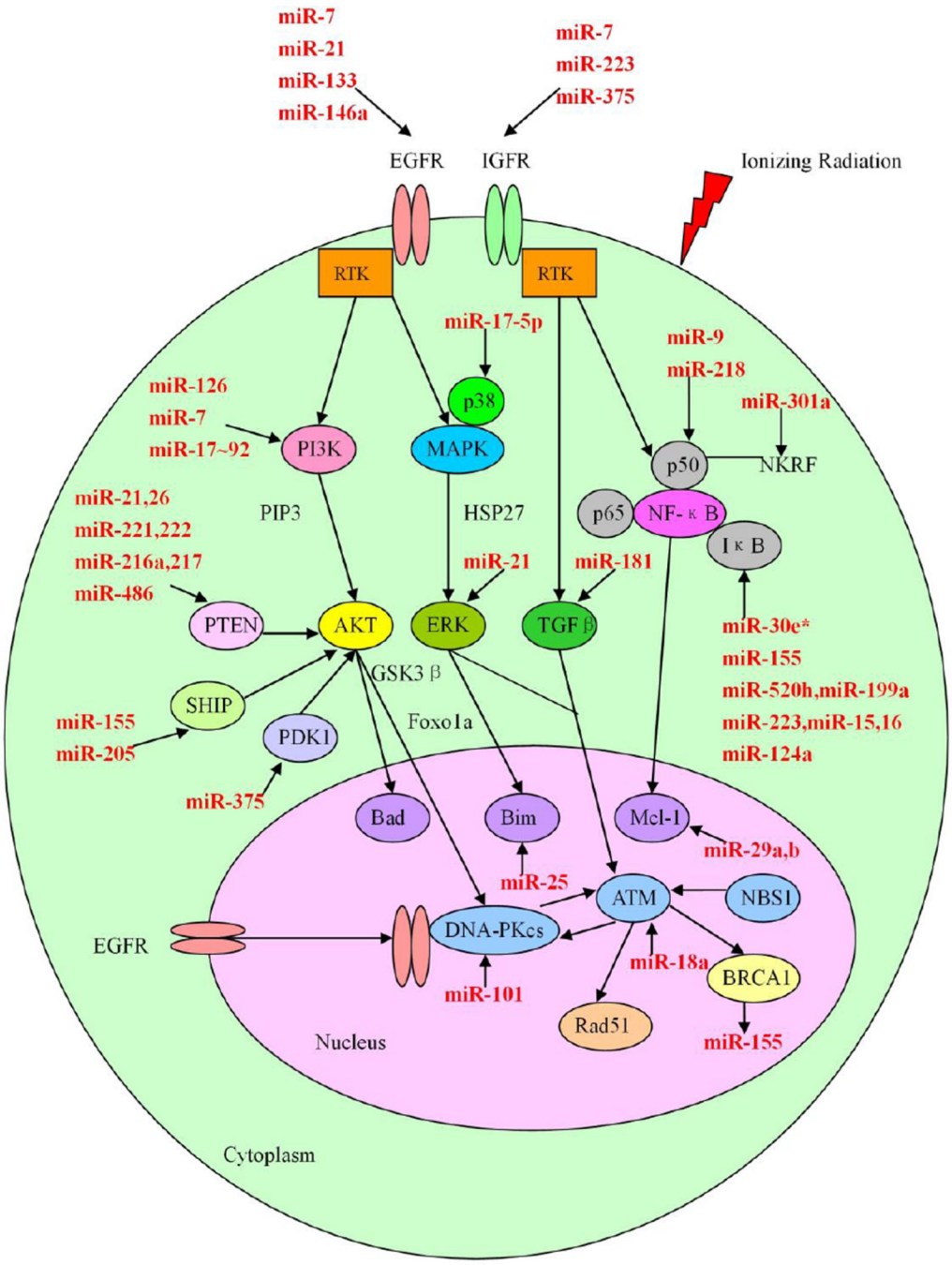

Figure 3. MiRNA is involved in the regulation of radiation related signal transduction pathways

When the tumor cell is inflicted with ionizing radiation or when the intracellular RTK is activated by EGFR or IGFR, the PI3K/AKT, MAPK/ERK, NF-κB and TGF-β pathways will be activated as cascades. Multiple miRNAs could modulate the expression of key components in these pathways, accompanied with the expression of their upstream molecules like PTEN, SHIP and PDK1, or their activated subunits like p38, p50, p65 and IκB, so as to further promote the activation of these signal transduction pathways. After the signals transfer into the nucleus, more downstream target genes will be activated, which include the apoptosis related genes like Bad, Bim and Mcl-1, and the DNA damage response genes like DNA-PKcs, ATM, NBS1, Rad51 and BRCA1. Also, EGFR can translocate into

the nucleus and form a complex with DNA-PKcs in order to initiate the NHEJ repair process. Consequently, miRNA is involved in the regulation of various key factors in these radiation related signal transduction pathways and could effectively affect the radiation response and radiosensitivity of tumor cells.

Table 1

MiRNA in the radiation related signal transduction pathways

Signal pathways	MiRNA	Targets	Biological effects	Refs
Upstream Receptors				
EGFR	miR-7	EGFR	overcome tumor radioresistance and improve radiotherapeutic effects	[28]
	miR-133	EGFR	inhibit tumor cell proliferation, invasion and migration	[29]
	miR-146a	MMP2, EGFR	suppress tumor growth and progression	[30]
	miR-21	EGFR	modulate radioresistance or radiosensitivity	[31]
IGFR	miR-7	IGFR, IRS1 and IRS2	regulate the survival and proliferation of tumor cells	[40]
	miR-223	IGFR, Rasa1	inhibit the proliferation and growth of tumor cells	[42]
	miR-375	IGFR	suppress tumor cell colony formation, migration and metastasis	[43]
Midstream Pathways				
PI3K/AKT pathway	miR-7	PIK3CD, mTOR and p70S6K	repress tumorigenesis and reverse the metastasis process of tumor cells	[50]
	miR-17–92	PHLPP2, PTEN and BIM	suppress tumor growth and affect chemoresistance	[51]
	miR-126	p85 beta	inhibit tumor cell growth	[52]
	miR-221 miR-222	PTEN	affect tumor cell growth, invasion and radiosensitivity	[53]
	miR-486	PTEN, Foxo1a	influence the activity of the PI3K/AKT pathway	[54]
MAPK/ERK pathway	miR-7-3 miR-34a miR-181d miR-193b	MAPK	inhibit tumor cell proliferation	[61]
	miR-17-5p	p38	regulate the proliferation and migration of tumors	[62]
	miR-21	ERK, PTEN	modulate the metabolic state of tumor cells and induce tumor angiogenesis and metastasis	[63, 64]
NF-κB pathway	miR-31	NIK	activate NF-κB pathway and influence tumor cell proliferation and apoptosis	[73]
	miR-30e*	IκB	elevate invasiveness and metastasis ability of tumor cells	[75]
	miR-301a	NKRF	promote the activation of NF-κB	[76]
TGF-β pathway	miR-520c miR-373	TGF-β, NF-κB	affect tumor metastasis, inflammation and progression	[85]

Signal pathways	MiRNA	Targets	Biological effects	Refs
	miR-21	TGF-β	regulate the EMT process	[86, 87]
	miR-17–92	TGFBR2, SMAD2 and SMAD4	inhibit cell proliferation and regulate collagen synthesis	[88]
	miR-181	ATM	regulate the properties of cancer stem cells	[89, 90]
	miR-106b-25	SMAD7	induce the EMT transition and tumor initiating-cell like phenotypes	[91]
Downstream Genes				
DNA damage response genes	miR-18a	ATM	affect DNA damage repair ability and HR efficiency	[111]
	miR-101	DNA-PK, ATM	influence the processes of NHEJ and HR in DNA damage repair	[112]
	miR-155	BRCA1	modulate DNA damage response and radiosensitivity of some specific tumors	[114]
Apoptosis related genes	miR-25	BIM, BAX and caspase-3	interfere apoptosis process and prevent cancer progression	[124]
	miR-29b miR-29a	MCL1	activate caspase-3 and induce apoptosis	[125, 126]
	miR-193b	MCL1	disturb apoptosis process and inhibit tumor cell proliferation	[127]

Abbreviation: EGFR: Epidermal growth factor receptor; MMP2: Matrix metalloproteinase-2; IGFR: Insulin-like growth factor receptor; IRS1: Insulin receptor substrate 1; IRS2: Insulin receptor substrate 2; mTOR: mammalian target of rapamycin; PHLPP2: PH domain and leucine rich repeat protein phosphatases; PTEN: Phosphatase and tensin homolog; MAPK: Mitogen-activated protein kinases; ERK: Extracellular signal-regulated kinases; NIK: NF-κB inducing kinase; NKRF: NF-κB repressing factor; EMT: Epithelial–mesenchymal transition; TGFBR2: Transforming growth factor, beta receptor 2; ATM: Ataxia telangiectasia mutated; DNA-PK: DNA-dependent protein kinase; HR: Homologous recombination; NHEJ: Non-homologous end joining; BRCA1: Breast cancer 1; MCL1: Myeloid cell leukemia sequence