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DNA Methylation in Radiation-Induced Carcinogenesis: Experimental Evidence and Clinical Perspectives

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Abstract

Ionizing radiation is a valuable tool in many spheres of human life. At the same time, it is a genotoxic agent with a well-established carcinogenic potential. Progress achieved in the last two decades has demonstrated convincingly that ionizing radiation can also target the cellular epigenome. Epigenetics is defined as heritable changes in the expression of genes that are not due to alterations of DNA sequence but consist of specific covalent modifications of chromatin components, such as methylation of DNA, histone modifications, and control performed by non-coding RNAs. Accumulating evidence suggests that DNA methylation, a key epigenetic mechanism involved in the control of expression of genetic information, may serve as one of the driving mechanisms of radiation-induced carcinogenesis. Here, we review the literature on the effects of ionizing radiation on DNA methylation in various biological systems, discuss the role of DNA methylation in radiation carcinogenesis, and provide our opinion on the potential utilization of this knowledge in radiation oncology.

Keywords

DNA damage; epigenetics; LINE-1; methionine; mutation; one-carbon metabolism; repetitive elements

I. INTRODUCTION

Ionizing radiation (IR) remains a pillar of the care and cure of many cancer patients worldwide. At the same time, the carcinogenic potential of IR has been recognized for over a century since the first radiation-induced skin cancers and leukemia in occupationally exposed workers were reported.[1–4] These studies were followed by a plethora of

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epidemiological studies, including long-term follow-up studies on A-bomb survivors,[5–7] as well as reports on occupational, accidental, and environmental exposures[8–10] and on patients exposed for various diagnostic and treatment purposes.[11–14] Those studies, together with the number of studies that utilized cellular and animal models, highlighted the universal carcinogenic potential of IR, which was shown to cause cancers “in most tissues of most species and at all ages.”[15]

The major mechanisms of radiation carcinogenesis are linked to DNA damage associated with it misrepaired DNA lesions and the induction of mutations, our knowledge of which is summarized in excellent reviews elsewhere.[1,15] In addition to these mechanisms, the development of radiation-induced genomic instability, a multifactorial phenomenon exhibited as an increased frequency of mitotically heritable genetic alterations observed in the progeny of irradiated cells multiple generations after exposure,[16–18] suggests that there are other mechanisms that may be implicated in radiation carcinogenesis. The observed frequency of genetic instability induced by IR exposure is substantially higher than that observed for gene mutations at a similar dose; therefore, the latter is considered highly unlikely to be the initiating mechanism.[15,19] In this regard, epigenetic alterations are of particular interest.[18,20]

II. EPIGENETIC MECHANISMS THAT REGULATE THE EXPRESSION OF GENETIC INFORMATION

Epigenetics is the study of heritable changes in gene expression that are not associated with alterations in the underlying DNA sequence—in other words, changes in phenotype without actual changes in genotype. The epigenetic mechanisms that regulate the expression of genetic information include DNA methylation, post-translational histone modifications, and nucleosome positioning along DNA. These covalent marks that ensure the proper structure and function of the epigenome are applied by specific enzymes: DNA and histone methyltransferases, the so-called “writers.” The “readers,” proteins that recognize these marks, in turn modulate the gene expression at particular genomic loci. Conversely, the “erasers” are the enzymes that guarantee the reversibility of the previously applied covalent marks, underlining the plasticity of the epigenome.[21]

Epigenetic mechanisms are critical during development, as well as for the maintenance of cellular homeostasis. Regarding epigenetic mechanisms, the expression of genetic information is regulated in a cell-, tissue-, and sex-specific manner.[22,23] Epigenetics also plays a key role in controlling the expression of repetitive elements (REs), which occupy more than 50% of mammalian genomes.[24]

DNA methylation, the most studied and characterized epigenetic modification, is a covalent addition of a methyl group to the fifth position of carbon. This process is enabled by a complex interplay among the DNA methyltransferases, methyl-CpG-binding proteins, and the recently characterized protein ubiquitin-like with PHD and RING finger domains 1 (*UHRF1*). More than 50% of eukaryotic genes, as well as a mosaic palette of REs, contain CpGrich regions (also known as CpG islands, or CGIs). Depending on the location of CGIs, DNA methylation can have differential regulatory functions. For instance, methylation of

DNA at CGIs located within the gene promoter/transcription start site is usually associated with gene silencing.[25] However, it remains unknown whether this mechanism is repressive by nature or is simply a lack of activation.[26] Conversely, DNA methylation of gene bodies (introns and exons) is not associated with gene silencing and either leads to stimulated elongation and splicing or prevention of initiation of aberrant transcription from alternative transcription start sites.[22,27,28]

Alterations in DNA methylation may lead to cellular epigenetic-based reprogramming, resulting in altered gene and RE expressions, genomic instability, and the development of pathological states, including cancer. In the 1970s and early 1980s, epigenetic mechanisms were proposed to be involved in carcinogenesis, with the loss of global DNA methylation being reported as the first epigenetic alteration detected in several human cancers.[29,30] These studies were followed by the identification of DNA hypermethylation at the promoter regions of tumor-suppressor genes in cancerous tissue.[31–33] Further studies have demonstrated that these alterations in global and gene-specific DNA methylation can often be detected at early stages of carcinogenesis, leading to the development of the hypothesis that epigenetic mechanisms may serve as drivers of carcinogenesis.[34–38]

III. EPIGENETIC MECHANISMS OF CARCINOGENESIS

Today, several decades since the first report of global DNA hypomethylation in cancerous tissue,[29,30,39] virtually all human cancers have been characterized by this epigenetic alteration. Currently, it is generally accepted that global genomic hypomethylation is a recognized hallmark of cancer.[40,41] Loss of global genomic methylation is usually associated with the hypomethylation of REs, many of which, such as the LINE-1 and *Alu* elements, are retrotransposons by nature.[42] Retrotransposons are mobile DNA elements that, during evolution had heavily populated mammalian genomes due to their ability to propagate via the “copy-paste” mechanism.[43] DNA hypomethylation in retrotransposons may lead to the loss of epigenetic control over those REs, resulting in their reactivation and subsequent retrotransposition. The latter is usually exhibited as insertional mutagenesis, in which the retrotransposon randomly inserts its copy at a different genomic location, often leading to a shifted open reading frame of the protein-encoding genes.[24]

It remains unclear whether the retrotransposition is a carcinogenesis-driven event or merely the consequence of the overall genomic instability observed in cancer cells. However, LINE-1 retrotransposition has been reported recently in numerous human cancers.[44–46] Increased retrontransposition may also result in another unwanted effect, genomic amplification, which may further have a negative effect on genome stability.

Even without the subsequent reactivation of retrotransposons, DNA hypomethylation may have substantial influence on carcinogenesis. For instance, it has been shown that the *MET* oncogene has evolutionary acquired a LINE-1 insertion within its gene body together with the CGI containing an alternative transcription start site. Loss of DNA methylation from this CGI results in aberrant transcription and increased *MET* copy numbers and is usually associated with a poor prognosis and rapid development of distant metastasis.[47,48]

DNA hypermethylation, which is observed in a number of tumor-suppressor genes, is another frequently observed epigenetic alteration in cancer cells. Often, promoter DNA hypermethylation in such genes as cyclin-dependent kinase 2A (*CDKN2A*, also known as $P16^{INK4A}$), O^6-methylguanine-DNA methyltransferase (*MGMT*), and phosphatase and tensin homolog (*PTEN*), to name a few, is associated with their transcriptional silencing in the tumor. The latter effect, however, remains controversial because a number of studies reported on gene silencing preceding DNA hypermethylation, as well as a lack of influence of promoter DNA methylation on gene and RE expression.[22,28,49]

It is becoming increasingly recognized that both genetic and epigenetic alterations in concert contribute to the process of carcinogenesis. For instance, a number of recent studies have convincingly demonstrated that the vast majority of human cancers harbor mutations in the genes that belong to the epigenetic machinery (i.e., DNA methyltransferases).[50,51] Alterations in the expression of those genes may compromise the cellular epigenome, subsequently leading to altered gene expression and genomic instability.[21,25,52] Conversely, epigenetic alterations may further predispose to mutations because DNA hypermethylation-mediated silencing of the critical DNA repair gene *MLH1* has led to the development of new mutations due to inefficient DNA repair.[53]

IV. DNA methylation in radiationinduced cancers

In 2004, Belinsky et al. reported DNA hypermethylation of the $p16^{INK4A}$ gene in lung adenocarcinomas of plutonium-exposed workers at the Russian nuclear enterprise MAYAK.[54] Interestingly, the levels of $p16^{INK4A}$ DNA hypermethylation were 3.5-fold higher in the adenocarcinomas of exposed workers compared with non-IR worker controls (confidence interval = 1.5–8.5; $p = 0.001$). The investigators also reported that the increased probability for gene-specific methylation approximated a 4-fold increase in relative risk for adenocarcinoma in workers exposed to plutonium.[54] In another study, Su et al. detected DNA hypermethylation of the $P16^{INK4A}$ ($z = 2.844$, $p = 0.005$) and *MGMT* ($z = 3.034$, $p = 0.002$) genes in the sputum of uranium miners.[55] The degree of DNA hypermethylation in the promoter of p16 and *MGMT* significantly correlated with the cumulative doses of radon exposure, with the cumulative exposure dose range of 12 ± 6 to 294 ± 132 ($z = 3.859$, $p = 0.0001$). Interestingly, a recent study using a mouse model also identified DNA hypermethylation of $p16^{INK4A}$ and its transcriptional silencing in radiation-induced thymic lymphoma.[56]

Other studies performed with the cohort of MAYAK workers diagnosed with lung adenocarcinoma demonstrated hypermethylation of *GATA5*, a gene that plays a critical role in cellular differentiation.[57] The investigators have also acknowledged the higher incidence of DNA hypermethylation in adenocarcinomas from MAYAK workers, in whom at least one of five investigated genes was hypermethylated in 93% of cases, whereas in non-IR workers, this effect was observed in only 66% of cases. However, it must be emphasized that, to date, no radiation-specific DNA hypermethylation signatures were reported.

Results from epidemiological studies suggested that epigenetic alterations, aberrant DNA methylation in particular, may be involved in radiation carcinogenesis, which inspired the investigation of effects of IR on DNA methylation in experimental systems.

A number of studies showed that exposure to IR may substantially affect the cellular epigenome and result in the loss of global DNA methylation, especially in organs and systems known to be sensitive to radiation-induced carcinogenesis. For instance, Giotopoulos et al. reported loss of DNA methylation in bone marrow of mice exposed to 3 Gy of IR.[58] Similar losses of DNA methylation were observed in other cancer-prone tissues such as the thymus, mammary gland, and spleen,[59–61] but not in the lung or muscle tissues.[62] Importantly, those effects could be detected at extended time points after irradiation, even when the radiation-induced DNA damage was long since repaired.[59] These effects were also detectable in the organ and systems not directly exposed to IR.[63] A decrease in global DNA methylation was reported in a rat model shortly after irradiation,[61] as well as in rats with radiation-induced mammary tumors,[64] further establishing a link between radiation-induced global genomic hypomethylation and carcinogenesis. Another important finding from those studies was that the IR-induced changes in DNA methylation primarily stem from REs rather than from individual genes.[42,63,65,66]

Given the abundance of REs in mammalian genomes and their extensive DNA methylation, the effects of IR on DNA methylation on a global scale can be detected with a high degree of reproducibility. Conversely, studies investigating IR-induced gene-specific DNA methylation often present controversial results. The introduction of next-generation approaches into DNA methylation analysis promises to shed more light on the effects of IR at the level of single-gene resolution.

In one of the pioneering studies, Antwih et al., using the 450 K methylation array approach, observed substantial changes in gene-specific DNA methylation in human breast cancer cell lines.[67] Interestingly, gene ontology analysis revealed that a large fraction of affected genes belonged to radiation response pathways. Similarly, radiation-induced changes in DNA methylation were reported by Bae et al., who investigated the response in HCT116 human colorectal cells.[68] Conversely, two studies performed on normal human fibroblasts reported a lack of radiation-induced changes in DNA methylation.[69,70] The results of these studies clearly demonstrated that the IR-induced effects on DNA methylation critically depend on the cell/ tissue type. For example, even in one of the most sensitive organs to IR exposure, the bone marrow, clear patterns of cell specificity regarding the magnitude of response were observed.[71,72] The observed changes were primarily detected in genetically unstable cancer cells and cell lines with compromised DNA repair, which may explain the considerably higher degree of changes in DNA methylation after irradiation. Other contributing factors may also be associated with different doses, quality of radiation, time points after irradiation at which DNA methylation was evaluated, and differences in the approaches for data analysis.[66,73,74] Furthermore, *in vitro* systems do not represent the whole organismal response and lack the influence of other important factors that may predetermine the tissue/ cell epigenetic response. For instance, it has been shown that male C57BL/6J mice exhibiting a robust IR-induced epigenetic response have a clear lack of this response when a gonadectomy is performed.[75]

V. ARE THE OBSERVED EPIGENETIC EFFECTS DRIVERS OR PASSENGERS?: HIGH-LINEAR ENERGY TRANSFER (LET) RADIATION AS A MODEL TO STUDY THE EPIGENETIC MECHANISMS OF RADIATION CARCINOGENESIS

It is becoming increasingly recognized that alterations in DNA methylation are not just the passive bystanders in the process of carcinogenesis or consequences of neoplastic transformation and, very possibly, they are the active players that shape the tumor landscape. Indeed, as discussed above, altered DNA methylation can be detected very early during the process of carcinogenesis and may influence numerous biological processes. It is also becoming increasingly recognized that both genetic and epigenetic alterations in concert contribute to carcinogenesis. However, one of the most challenging aspects of investigating the role of epigenetic alterations in genotoxic carcinogenesis is determining the truly epigenetically driven mechanisms. For instance, the loss of DNA methylation that is observed in the vast majority of cases after exposure to doses of 1 Gy and above may be also mediated by the substantial damage to DNA or preoccupation of DNA methylatransferases (DNMT1) and DNA methylation accessory protein (*UHRF1*) in recruiting the repair complexes to the sites of damaged DNA instead of a direct response in the maintenance of DNA methylation. One could thus consider that the genotoxic effects of IR simply predetermine the epigenetic alterations.

In this regard, of particular interest are the model systems that utilize exposure to low mean absorbed doses of high-LET radiation, such as protons and heavy ions, types of IR that are dominant in the space environment. The necessity of understanding the effects of IR exposure during the space missions and the introduction of high-LET radiation into clinical practice have triggered the investigation of biological and molecular mechanisms of response to high-LET radiation.[76–78]

Studies in *in vitro* and animal experimental systems clearly indicate that exposure to heavy ions results in clustered DNA damage compared with low-LET terrestrial radiation.[76,79–81] This more complex DNA damage is frequently irreparable and usually promotes apoptosis via p53 at the S/G_2 checkpoint, leading to greater relative biological effectiveness.[82]

Cytogenetic studies report a much higher complexity of chromosomal rearrangements caused by exposure to heavy ions compared with sparsely ionizing IR.[83,84] The complexity of these rearrangements, however, determines the lethality of the vast majority of them. Both *in vitro* and *in vivo* studies have reported a very low number of complex rearrangements within a short time after irradiation.[79,85] Furthermore, it has been shown that the levels of chromosomal aberrations in astronauts with a total time of 2 years spent in space were not substantially higher than the background measurements taken before the first flight.[86]

Despite the lack of detectable DNA damage and chromosomal aberrations, studies in experimental animal models have shown that heavy ions are not only potent carcinogens, but can induce cancers at much lower doses and even in organs that are not known to be the classical organs for IR-induced carcinogenesis.[87–94] For instance, leukemogenesis studies in

mice demonstrate that exposures to as low as a 0.4 Gy mean absorbed dose of heavy iron ions (^{56}Fe) were enough to increase the levels of leukemia, whereas doses above 1 Gy of low-LET irradiation were needed to cause the same effect. Other studies also reported lung tumors in mice exposed to ^{56}Fe or protons,[89] as well as enhanced intestinal tumor multiplicity in APC^{min} mice.[90,91] In addition, high-LET radiation was shown to be a very potent inducer of liver tumors, a site that is not common for radiation carcinogenesis.

Studies using the exposures to low-mean absorbed doses of high-LET IR may aid in better understanding the driving potential of epigenetic mechanisms in radiation-induced carcinogenesis. For instance, in a study assessing the dose-dependent effects of total body irradiation to low-mean absorbed doses of ^{56}Fe, Miousse et al. demonstrated an absence of detectable DNA damage, as well as no increases in reactive oxygen species, senescent cells, or apoptotic events in the hematopoietic stem and progenitor cells 1 and 5 months after exposure.[71] At the same time, changes in DNA methylation of transposable elements LINE-1 and SINE B1 (corresponding to *Alu* elements in humans) and DNA methylation machinery were detected in the pool of hematopoietic stem and progenitor cells after exposure to leukemogenic (0.4 Gy, 1 A GeV), but not lower doses of ^{56}Fe. Importantly, those changes were still evident 5 months after exposure and also resulted in reactivation of LINE-1 elements that may further lead to LINE-1 insertional mutagenesis, genome amplification, and the development of genomic instability. The persistence of epigenetic alterations considered as a hallmark of cancer (loss of global and RE-associated DNA methylation paralleled by reactivation of the LINE-1 retrotransposon) in the absence of detectable DNA damage and other cellular and molecular alterations suggests that epigenetic reprogramming may serve as one of the driving forces in IR-induced carcinogenesis. Further studies are clearly needed to confirm this hypothesis and to prove the causative role of epigenetic alterations and DNA methylation related to radiation-induced carcinogenesis.

VI. TARGETING THE CANCER EPIGENOME FOR RADIOSENSITIZATION: CONSIDERATIONS FOR ONE-CARBON METABOLISM RELATED EFFECTS

Given that IR is capable of inducing stable alterations to DNA methylation and that there is a higher degree of IR-induced epigenetic responses in the cancerous cell, it seems reasonable to expect that tumor radiosensitization could be achieved by mod-ulation of the tumor cell epigenome. This notion is strengthened by the finding that DNA methylation levels can regulate the cancer cell response to radiotherapy. Kim et al., using the radiosensitive (H460) and radioresistant (H1299) human non-small-cell lung cancer cell lines, demonstrated the differential DNA methylation patterns in 747 genes.[95] The investigators have further shown that silencing of *SERPINB5* and *S100A6* can mediate radioresistance in H460 cells.

The DNA methylation status of LINE-1, the most abundant and usually heavily methylated RE, is becoming a valuable tool in the prognosis of tumor response to therapy. It is generally recognized that a lower degree of LINE-1 DNA methylation is associated with a poor prognosis, advanced metastasis, and weak tumor response to treatment.[42] Some studies indicate that the DNA methylation status of LINE-1 may also serve as a predictor of tumor

response to radiotherapy[96]; however, more basic and clinical research is needed to confirm these findings.

The potential of DNMT inhibitors such as nucleoside analogs (5-azacytidine, decitabine, and zebularine) in tumor radiosensitization is becoming increasingly recognized. Those drugs were first introduced into clinical practice several decades ago, but were only shown to improve the blood cell count and survival in patients with myelodysplastic syndrome and acute myeloid leukemia after dose optimization was achieved in recent years.[97,98] At the same time, the results of the clinical trials in patients with solid tumors were less promising.[99]

A series of *in vitro* studies demonstrated increased sensitivity to radiotherapy in gastric, glioblastoma, head and neck, colorectal, and nasopharyngeal cancer cell lines when treated with these nucleoside analogs.[100–104] Emerging evidence also exists on the success of combined nucleoside analog/ radiotherapy treatment in glioblastoma U251 and nasopharyngeal carcinoma xenograft models.[100,104]

Another promising and still unexplored avenue in tumor radiosensitization is via targeting one-carbon metabolism, one of the major biochemical pathways in living organisms that affects nearly all cellular functions and more than 100 specific biomethylation reactions. One-carbon metabolism involves the reactions that surround the transfer of the methyl group from S-adenosylmethionine (SAM) to acceptor molecules and the regeneration of SAM. The latter ties together gene regulation, amino acid synthesis, purine and pyrimidine synthesis, antioxidants, and four vitamins. One of the central molecules involved in one-carbon metabolism is the essential amino acid methionine. Methionine is critical for a number of vital processes, including the synthesis of SAM, a universal donor of methyl groups for DNA, RNA, protein, and lipid methylation. Methionine is also needed for the synthesis of glutathione and is indispensable for protein synthesis.

Interestingly, there is a remarkable difference in the needs for methionine between normal tumor cells; rapidly proliferating cancer cells require much higher levels of methionine to maintain function. Therefore, as would be expected, tumor cells are extremely sensitive to methionine restriction. Although the normal cell has a capacity for re-methylation and further utilization of methionine from homocysteine, the cancer cell is incapable of proper synthesis and utilization of endogenous methionine.[105] For instance, it has been shown that plating normal fibroblasts and tumor cells together in methionine-deficient homocysteine-supplemented medium results in cell cycle arrest and apoptosis of tumor cells, whereas normal fibroblasts grow abundantly.[106]

Although the potentiation of the chemotherapy effect of methionine deprivation has been investigated both *in vitro*[107] and *in vivo*[108,109] and even in clinical trials,[110,111] the potential combination of methionine dietary deprivation with radiotherapy has yet to be addressed. Accumulating evidence indicates that cancer cells are radiosensitized by the deprivation of other methyl group donors,[112,113] suggesting that methionine deprivation combined with radiotherapy may have beneficial effects for cancer treatment.

Whereas the potential of this approach seems clear, several critical issues need to be addressed due to the toxicity associated with long-term methionine restriction. Classical cancer therapy regimens, chemotherapy or radiotherapy, are usually a lengthy process over several months. Long-term methionine deficiency is associated with substantial weight loss both in rodent models and in clinical trials, as well as thrombocytopenia, neutropenia, and the development of hepatosteatosis.[110,111,114,115] Recent advances both in tumor imaging and radiation techniques, as well as the development and widespread implementation of stereotactic body radiation therapy regimens into clinical practice[116–118] may, along with improved local tumor control, significantly decrease the duration of treatment and thus allow for effective and safe methionine dietary interventions.

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ABBREVIATIONS:

CDKN2A	cyclin-dependent kinase 2A
CGI	CpG island
DNMT	DNA methyltransferase
IR	ionizing radiation
LET	linear energy transfer
LINE-1	Long Interspersed Nucleotide Element 1
MGMT	O^6-methylguanine-DNA methyltransferase
PTEN	phosphatase and tensin homolog
RE	repetitive element
SAM	S-adenosylmethionine
SINE B1	Short Interspersed Nucleotide Element B1
UHRF1	ubiquitin-like with PHD and RING finger domains 1

REFERENCES

1. Simic MG, Grossman L, Upton AC, Bergtold DS. Mechanisms of DNA damage and repair: Implications for carcinogenesis and risk assessment New York: Springer Science and Business Media; 2013.
2. Little MP, Tawn EJ, Tzoulaki I, Wakeford R, Hildebrandt G, Paris F, Tapio S, Elliott P. A systematic review of epidemiological associations between low and moderate doses of ionizing radiation and

late cardiovascular effects, and their possible mechanisms. Radiat Res 2008;169(1):99–109. [PubMed: 18159955]

3. de Gonzalez AB, Kutsenko A, Rajaraman P. Sarcoma risk after radiation exposure. Clinical Sarcoma Res 2012;2(1): 18. [PubMed: 23036235]
4. Council NR. Health risks from exposure to low levels of ionizing radiation: BEIR VII phase 2 Washington, DC: National Academies Press; 2006.
5. Preston DL, Ron E, Tokuoka S, Funamoto S, Nishi N, Soda M, Mabuchi K, Kodama K. Solid cancer incidence in atomic bomb survivors: 1958–1998. Radiat Res 2007; 168(1):1–64. [PubMed: 17722996]
6. Thompson DE, Mabuchi K, Ron E, Soda M, Tokunaga M, Ochikubo S. Cancer incidence in atomic-bomb survivors. 2. Solid Tumors, 1958–1987. Radiat Res 1994;137(2): S17–67. [PubMed: 8127952]
7. Wolff S More about cancer incidence in atomic-bomb survivors: Solid tumors, 1958–1987 [Reply]. Radiat Res 1995;141(2):230–1.
8. Robertson A, Allen J, Laney R, Curnow A. The cellular and molecular carcinogenic effects of radon exposure: A review. Int J Mol Sci 2013;14(7):14024–63. [PubMed: 23880854]
9. Johnson CJ. Cancer incidence in an area of radioactive fallout downwind from the Nevada test site. JAMA 1984;251(2):230–6. [PubMed: 6690781]
10. Krestinina LY, Davis FG, Schonfeld S, Preston DL, Degteva M, Epifanova S, Akleyev AV. Leukaemia incidence in the Techa River Cohort: 1953–2007. Br J Cancer 2013;109(11):2886–93. [PubMed: 24129230]
11. de Gonzalez AB, Gilbert E, Curtis R, Inskip P, Kleinerman R, Morton L, Rajaraman P, Little MP. Second solid cancers after radiation therapy: a systematic review of the epidemiologic studies of the radiation dose–response relationship. Int J Radiat Oncol 2013;86(2):224–33.
12. Travis LB, Ng AK, Allan JM, Pui CH, Kennedy AR, Xu XG, Purdy JA, Applegate K, Yahalom J, Constine LS, Gilbert ES, Boice JD, Jr. Second malignant neoplasms and cardiovascular disease following radiotherapy. J Natl Cancer Inst 2012;104(5):357–70. [PubMed: 22312134]
13. Tukenova M, Guibout C, Hawkins M, Quiniou E, Mousannif A, Pacquement H, Winter D, Bridier A, Lefkopoulos D, Oberlin O, Diallo I, de Vathaire F. Radiation therapy and late mortality from second sarcoma, carcinoma, and hematological malignancies after a solid cancer in childhood. Int J Radiat Oncol 2011;80(2):339–46.
14. Kamran SC, de Gonzalez AB, Ng A, Haas-Kogan D, Viswanathan AN. Therapeutic radiation and the potential risk of second malignancies. Cancer 2016;122(12): 1809–21. [PubMed: 26950597]
15. Little JB. Radiation carcinogenesis. Carcinog 2000;21(3): 397–404.
16. Morgan WF. Non-targeted and delayed effects of exposure to ionizing radiation: I. Radiation-induced genomic instability and bystander effects in vitro. Radiat Res 2003; 159(5):567–80. [PubMed: 12710868]
17. Morgan WF. Non-targeted and delayed effects of exposure to ionizing radiation: II. Radiation-induced genomic instability and bystander effects in vivo, clastogenic factors and transgenerational effects. Radiat Res 2003;159(5):581–96. [PubMed: 12710869]
18. Baulch JE, Aypar U, Waters KM, Yang AJ, Morgan WF. Genetic and epigenetic changes in chromosomally stable and unstable progeny of irradiated cells. PLoS One 2014;9(9):e107722. [PubMed: 25251398]
19. Morgan WF. Non-targeted and delayed effects of exposure to ionizing radiation: I. Radiation-induced genomic instability and bystander effects in vitro. Radiat Res 2012; 178(2):AV223–36. [PubMed: 22870973]
20. Aypar U, Morgan WF, Baulch JE. Radiation-induced epigenetic alterations after low and high LET irradiations. Mutat Res 2011;707(1–2):24–33. [PubMed: 21159317]
21. Jones PA, Issa JPJ, Baylin S. Targeting the cancer epigenome for therapy. Nat Rev Genet 2016;17(10):630–41. [PubMed: 27629931]
22. Jones PA. Functions of DNA methylation: islands, start sites, gene bodies and beyond. Nat Rev Genet 2012;13(7): 484–92. [PubMed: 22641018]
23. Espada J, Esteller M. Epigenetic control of nuclear architecture. Cell Mol Life Sci 2007;64(4):449–57. [PubMed: 17221334]

24. Miousse IR, Chalbot MCG, Lumen A, Ferguson A, Kavouras IG, Koturbash I. Response of transposable elements to environmental stressors. Mutat Res Rev Mutat Res 2015;765:19–39. [PubMed: 26281766]
25. Baylin SB, Jones PA. Epigenetic determinants of cancer. Cold Spring Harb Perspect Biol 2016;8(9):a019505. [PubMed: 27194046]
26. Baubec T, Schubeler D. Genomic patterns and context specific interpretation of DNA methylation. Curr Opin Genet Dev 2014;25:85–92. [PubMed: 24614011]
27. Ehrlich M, Ehrlich KC. DNA cytosine methylation and hydroxymethylation at the borders. Epigenom 2014;6(6): 563–6.
28. Portela A, Esteller M. Epigenetic modifications and human disease. Nat Biotechnol 2010;28(10): 1057–68. [PubMed: 20944598]
29. Gamasosa MA, Slagel VA, Trewyn RW, Oxenhandler R, Kuo KC, Gehrke CW, Ehrlich M. The 5-methylcytosine content of DNA from human tumors. Nucleic Acids Res 1983;11(19):6883–94. [PubMed: 6314264]
30. Feinberg AP, Vogelstein B. Hypomethylation distinguishes genes of some human cancers from their normal counterparts. Nature 1983;301(5895):89–92. [PubMed: 6185846]
31. Graff JR, Herman JG, Lapidus RG, Chopra H, Xu R, Jarrard DF, Isaacs WB, Pitha PM, Davidson NE, Baylin SB. E-cadherin expression is silenced by DNA hypermethylation in human breast and prostate carcinomas. Cancer Res 1995;55(22):5195–9. [PubMed: 7585573]
32. Melki JR, Vincent PC, Clark SJ. Concurrent DNA hypermethylation of multiple genes in acute myeloid leukemia. Cancer Res 1999;59(15):3730–40. [PubMed: 10446989]
33. Pfeifer GP, Rauch TA. DNA methylation patterns in lung carcinomas. Semin Cancer Biol 2009;19(3):181–7. [PubMed: 19429482]
34. Koturbash I, Beland FA, Pogribny IP. Role of epigenetic events in chemical carcinogenesis-a justification for incorporating epigenetic evaluations in cancer risk assessment. Toxicol Mech Method 2011;21(4):289–97.
35. Robertson KD, Jones PA. DNA methylation: past, present and future directions. Carcinog 2000;21(3):461–7.
36. Koturbash I, Simpson NE, Beland FA, Pogribny IP. Alterations in histone H4 lysine 20 methylation: Implications for cancer detection and prevention. Antioxid Redox Sign 2012; 17(2): 365–74.
37. Sandoval J, Esteller M. Cancer epigenomics: beyond genomics. Curr Opin Genet Dev 2012;22(1): 50–5. [PubMed: 22402447]
38. Rodriguez-Paredes M, Esteller M. Cancer epigenetics reaches mainstream oncology. Nat Med 2011;17(3):330–9. [PubMed: 21386836]
39. Feinberg AP, Gehrke CW, Kuo KC, Ehrlich M. Reduced genomic 5-methylcytosine content in human colonic neoplasia. Cancer Res 1988;48(5):1159–61. [PubMed: 3342396]
40. Hanahan D, Weinberg RA. The hallmarks of cancer. Cell 2000;100(1):57–70. [PubMed: 10647931]
41. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell 2011;144(5):646–74. [PubMed: 21376230]
42. Miousse IR, Koturbash I. The fine LINE: methylation drawing the cancer landscape. Biomed Res Int 2015; 2015:131547. [PubMed: 26448926]
43. Hancks DC, Kazazian HH. Active human retrotransposons: variation and disease. Curr Opin Genet Dev 2012; 22(3):191–203. [PubMed: 22406018]
44. Miki Y, Nishisho I, Horii A, Miyoshi Y, Utsunomiya J, Kinzler KW, Vogelstein B, Nakamura Y. Disruption of the Apc gene by a retrotransposal insertion of L1 sequence in a colon cancer. Cancer Res 1992;52(3):643–5. [PubMed: 1310068]
45. Iskow RC, McCabe MT, Mills RE, Torene S, Pittard WS, Neuwald AF, Van Meir EG, Vertino PM, Devine SE. Natural mutagenesis of human genomes by endogenous retrotransposons. Cell 2010;141(7):1253–61. [PubMed: 20603005]
46. Lee E, Iskow R, Yang LX, Gokcumen O, Haseley P, Luquette LJ, Luquette LJ, 3rd, Lohr JG, Harris CC, Ding L, Wilson RK, Wheeler DA, Gibbs RA, Kucherlapati R, Lee C, Kharchenko PV,

Park PJ; Cancer Genome Atlas Research Network. Landscape of somatic retrotransposition in human cancers. Science 2012;337(6097):967–71. [PubMed: 22745252]

47. Hur K, Cejas P, Feliu J, Moreno-Rubio J, Burgos E, Boland CR, Goel A. Hypomethylation of long interspersed nuclear element-1 (LINE-1) leads to activation of proto-oncogenes in human colorectal cancer metastasis. Gut 2014;63(4):635–46. [PubMed: 23704319]
48. Wolff EM, Byun HM, Han HF, Sharma S, Nichols PW, Siegmund KD, Yang AS, Jones PA, Liang G. Hypomethylation of a LINE-1 promoter activates an alternate transcript of the MET oncogene in bladders with cancer. PLoS Genet 2010;6(4):e1000917. [PubMed: 20421991]
49. Prior S, Miousse IR, Nzabarushimana E, Pathak R, Skinner C, Kutanzi KR, Allen AR, Raber J, Tackett AJ, Hauer-Jensen M, Nelson GA, Koturbash I. Densely ionizing radiation affects DNA methylation of selective LINE-1 elements. Environ Res 2016;150:470–81. [PubMed: 27419368]
50. Cancer Genome Atlas Research Network, Weinstein JN, Collisson EA, Mills GB, Shaw KR, Ozenberger BA, Ellrott K, Shmulevich I, Sander C, Stuart JM.. The Cancer Genome Atlas Pan-Cancer analysis project. Nat Genet 2013;45(10):1113–20. [PubMed: 24071849]
51. Kretzmer H, Bernhart SH, Wang W, Haake A, Weniger MA, Bergmann AK, Betts MJ, Carrillo-de-Santa-Pau E, Doose G, Gutwein J, Richter J, Hovestadt V, Huang B, Rico D, Jühling F, Kolarova J, Lu Q, Otto C, Wagener R, Arnolds J, Burkhardt B, Claviez A, Drexler HG, Eberth S, Eils R, Flicek P, Haas S, Humme M, Karsch D, Kerstens HHD, Klapper W, Kreuz M, Lawerenz C, Lenzek D, Loeffler M, López C, MacLeod RAF, Martens JHA, Kulis M, Martín-Subero JI, Möller P, Nage I, Picelli S, Vater I, Rohde M, Rosenstiel P, Rosolowski M, Russell RB, Schilhabel M, Schlesner M, Stadler PF, Szczepanowski M, Trümper L, Stunnenberg HG, Küppers R, Ammerpohl O, Lichter P, Siebert R, Hoffmann S, Radlwimmer B. DNA methylome analysis in Burkitt and follicular lymphomas identifies differentially methylated regions linked to somatic mutation and transcriptional control. Nat Genet 2015;47(11):1316–25. [PubMed: 26437030]
52. Baylin SB, Jones PA. A decade of exploring the cancer epigenome: Biological and translational implications. Nat Rev Cancer 2011;11(10):726–34. [PubMed: 21941284]
53. Springuel L, Losdyck E, Saussoy P, Turcq B, Mahon FX, Knoops L, Renauld JC. Loss of mutL homolog-1 (MLH1) expression promotes acquisition of oncogenic and inhibitor-resistant point mutations in tyrosine kinases. Cell Mol Life Sci 2016;73(24):4739–48. [PubMed: 27436342]
54. Belinsky SA, Klinge DM, Liechty KC, March TH, Kang T, Gilliland FD, Sotnic N, Adamova G, Rusinova G, Telnov V. Plutonium targets the p16 gene for inactivation by promoter hypermethylation in human lung adenocarcinoma. Carcinogen 2004;25(6):1063–7.
55. Su SB, Jin YL, Zhang W, Yang LJ, Shen YP, Cao Y, Tong J. Aberrant promoter methylation of p16(INK4a) and O-6-methylguanine-DNA methyltransferase genes in workers at a Chinese uranium mine. J Occup Health 2006;48(4):261–6. [PubMed: 16902270]
56. Song WG, Liu YZ, Liu Y, Zhang C, Yuan B, Zhang LB, Sun S. Increased P16 DNA methylation in mouse thymic lymphoma induced by irradiation. PLoS One 2014;9(4):e93850. [PubMed: 24747802]
57. Lyon CM, Klinge DM, Liechty KC, Gentry FD, March TH, Kang T, Gilliland FD, Adamova G, Rusinova G, Telnov V, Belinsky SA. Radiation-induced lung adenocarcinoma is associated with increased frequency of genes, inactivated by promoter hypermethylation. Radiat Res 2007;168(4): 409–14. [PubMed: 17903034]
58. Giotopoulos G, McCormick C, Cole C, Zanker A, Jawad M, Brown R, Plumb M. DNA methylation during mouse hemopoietic differentiation and radiation-induced leukemia. Exp Hematol 2006;34(11):1462–70. [PubMed: 17046565]
59. Koturbash I, Pogribny I, Kovalchuk O. Stable loss of global DNA methylation in the radiation-target tissue - A possible mechanism contributing to radiation carcinogenesis? Biochem Bioph Res Co 2005;337(2):526–33.
60. Pogrlbny I, Koturbash I, Tryndyak V, Hudson D, Stevenson SML, Sedelnikova O, Bonner W, Kovalchuk O. Fractionated low-dose radiation exposure leads to accumulation of DNA damage and profound alterations in DNA and histone methylation in the murine thymus. Mol Cancer Res 2005;3(10):553–61. [PubMed: 16254189]
61. Loree J, Koturbash I, Kutanzi K, Baker M, Pogribny I, Kovalchuk O. Radiation-induced molecular changes in rat mammary tissue: Possible implications for radiation-induced carcinogenesis. Int J Radiat Biol 2006;82(11):805–15. [PubMed: 17148264]

62. Kovalchuk O, Burke P, Besplug J, Slovack M, Filkowski J, Pogribny I. Methylation changes in muscle and liver tissues of male and female mice exposed to acute and chronic lowdose X-ray-irradiation. Mutat Res. 2004;548(1–2):75–84. [PubMed: 15063138]

63. Koturbash I, Boyko A, Rodriguez-Juarez R, McDonald RJ, Tryndyak VP, Kovalchuk I, Pogribny IP, Kovalchuk O. Role of epigenetic effectors in maintenance of the long-term persistent bystander effect in spleen in vivo. Carcinogen 2007;28(8):1831–8.

64. Takabatake M, Blyth BJ, Daino K, Imaoka T, Nishimura M, Fukushi M, Shimada Y. DNA methylation patterns in rat mammary carcinomas induced by pre- and post-pubertal irradiation. PLoS One 2016;11(10):e0164194. [PubMed: 27711132]

65. Nzabarushimana E, Miousse IR, Shao LJ, Chang JH, Allen AR, Turner J, Stewart B, Raber J, Koturbash I. Long-term epigenetic effects of exposure to low doses of Fe-56 in the mouse lung. J Radiat Res 2014;55(4):823–8. [PubMed: 24585548]

66. Miousse IR, Kutanzi KR, Koturbash I. Effects of ionizing radiation on DNA methylation: from experimental biology to clinical applications. Int J Radiat Biol 2017; 93(5):457–69. [PubMed: 28134023]

67. Antwih DA, Gabbara KM, Lancaster WD, Ruden DM, Zielske SP. Radiation-induced epigenetic DNA methylation modification of radiation-response pathways. Epigen 2013;8(8):839–48.

68. Bae JH, Kim JG, Heo K, Yang K, Kim TO, Yi JM. Identification of radiation-induced aberrant hypomethylation in colon cancer. BMC Genom 2015;16:56.

69. Lahtz C, Bates SE, Jiang Y, Li AX, Wu XW, Hahn MA, Pfeifer GP. Gamma irradiation does not induce detectable changes in DNA methylation directly following exposure of human cells. PLoS One 2012;7(9):e44858. [PubMed: 23024770]

70. Maierhofer A, Flunkert J, Dittrich M, Müller T, Schindler D, Nanda I, Haaf T. Analysis of global DNA methylation changes in primary human fibroblasts in the early phase following X-ray irradiation. PLoS One 2017; 12(5):e0177442. [PubMed: 28489894]

71. Miousse IR, Shao LJ, Chang JH, Feng W, Wang YY, Allen AR, Turner J, Stewart B, Raber J, Zhou D, Koturbash I. Exposure to low-dose Fe-56-ion radiation induces long-term epigenetic alterations in mouse bone marrow hematopoietic progenitor and stem cells. Radiat Res 2014; 182(1):92–101. [PubMed: 24960414]

72. Miousse IR, Chang J, Shao L, Pathak R, Nzabarushimana É, Kutanzi KR, Landes RD, Tackett AJ, Hauer-Jensen M, Zhou D, Koturbash I. Inter-strain differences in LINE-1 DNA methylation in the mouse hematopoietic system in response to exposure to ionizing radiation. Int J Mol Sci 2017;18(7):1430.

73. Umer M, Herceg Z. Deciphering the epigenetic code: An overview of DNA methylation analysis methods. Antioxid Redox Sign 2013;18(15):1972–86.

74. Wilhelm-Benartzi CS, Koestler DC, Karagas MR, Flanagan JM, Christensen BC, Kelsey KT, Marsit CJ, Houseman EA, Brown R Review of processing and analysis methods for DNA methylation array data. Br J Cancer 2013; 109(6):1394–402. [PubMed: 23982603]

75. Koturbash I, Kutanzi K, Hendrickson K, Rodriguez-Juarez R, Kogosov D, Kovalchuk O. Radiation-induced bystander effects in vivo are sex specific. Mutat Res 2008;642(1–2):28–36. [PubMed: 18508093]

76. Durante M, Cucinotta FA. Heavy ion carcinogenesis and human space exploration. Nat Rev Cancer 2008; 8(6):465–72. [PubMed: 18451812]

77. Durante M New challenges in high-energy particle radiobiology. Br J Radiol 2014;87(1035):

78. Tommasino F, Durante M. Proton radiobiology. Cancers 2015;7(1):353–81. [PubMed: 25686476]

79. Rithidech KN, Honikel L, Whorton EB. mFISH analysis of chromosomal damage in bone marrow cells collected from CBA/CaJ mice following whole body exposure to heavy ions (Fe-56 ions). Radiat Environ Bioph 2007;46(2):137–45.

80. Liu CH, Kawata T, Zhou GM, Furusawa Y, Kota R, Kumabe A, Sutani S, Fukada J, Mishima M, Shigematsu N, George K, Cucinotta F. Comparison of the repair of potentially lethal damage after low- and high-LET radiation exposure, assessed from the kinetics and fidelity of chromosome rejoining in normal human fibroblasts. J Radiat Res 2013;54(6):989–97. [PubMed: 23674607]

81. Kronenberg A, Gauny S, Kwoh E, Grossi G, Dan C, Grygoryev D, Lasarev M, Turker MS. Comparative analysis of cell killing and autosomal mutation in mouse kidney epithelium exposed to 1 GeV protons in vitro or in vivo. Radiat Res 2013;179(5):511–20. [PubMed: 23560634]

82. Blakely EA, Kronenberg A. Heavy-ion radiobiology: New approaches to delineate mechanisms underlying enhanced biological effectiveness. Radiat Res 1998;150(5):S126–45. [PubMed: 9806616]

83. Durante M, George K, Wu H, Cucinotta FA. Karyotypes of human lymphocytes exposed to high-energy iron ions. Radiat Res 2002;158(5):581–90. [PubMed: 12385635]

84. Hada M, Cucinotta FA, Gonda SR, Wu HL. mBAND analysis of chromosomal aberrations in human epithelial cells exposed to low- and high-LET radiation. Radiat Res 2007;168(1):98–105. [PubMed: 17722995]

85. Durante M, George K, Cucinotta EA. Chromosomes lacking telomeres are present in the progeny of human lymphocytes exposed to heavy ions. Radiat Res 2006; 165(1):51–8. [PubMed: 16392962]

86. Durante M, Snigiryova G, Akaeva E, Bogomazova A, Druzhinin S, Fedorenko B, Greco O, Novitskaya N, Rubanovich A, Shevchenko V, Von Recklinghausen U, Obe G. Chromosome aberration dosimetry in cosmonauts after single or multiple space flights. Cytogenet Genome Res 2003;103(1–2):40–6. [PubMed: 15004462]

87. Wang ML, Hada M, Huff J, Pluth JM, Anderson J, O'Neill P, Cucinotta FA. Heavy ions can enhance TGF beta mediated epithelial to mesenchymal transition. J Radiat Res 2012;53(1):51–7. [PubMed: 22302045]

88. Bielefeldt-Ohmann H, Genik PC, Fallgren CM, Ullrich RL, Weil MM. Animal studies of charged particle-induced carcinogenesis. Health Phys 2012;103(5):568–76. [PubMed: 23032886]

89. Christofidou-Solomidou M, Pietrofesa RA, Arguiri E, Schweitzer KS, Berdyshev EV, McCarthy M, Corbitt A, Alwood JS, Yu Y, Globus RK, Solomides CC, Ullrich RL, Petrache I. Space radiation-associated lung injury in a murine model. Am J Physiol Lung Cell Mol Physiol 2015;308(5):L416–28. [PubMed: 25526737]

90. Datta K, Suman S, Kallakury BV, Fornace AJ, Jr. Heavy ion radiation exposure triggered higher intestinal tumor frequency and greater β-catenin activation than γ radiation in APC(Min/+) mice. PLoS One 2013;8(3):e59295. doi: 10.1371/journal.pone.0059295. [PubMed: 23555653]

91. Trani D, Datta K, Doiron K, Kallakury B, Fornace AJ. Enhanced intestinal tumor multiplicity and grade in vivo after HZE exposure: mouse models for space radiation risk estimates. Radiat Environ Biophys 2010;49(3):389–96. [PubMed: 20490531]

92. Weil MM, Ray FA, Genik PC, Yu Y, McCarthy M, Fallgren CM, Ullrich RL. Effects of 28Si ions, 56Fe ions, and protons on the induction of murine acute myeloid leukemia and hepatocellular carcinoma. PLoS One 2014 8 15; 9(7):e104819. doi: 10.1371/journal.pone.0104819. [PubMed: 25126721]

93. Weil MM, Bedford JS, Bielefeldt-Ohmann H, Ray FA, Genik PC, Ehrhart EJ, Fallgren CM, Hailu F, Battaglia CL, Charles B, Callan MA, Ullrich RL. Incidence of acute myeloid leukemia and hepatocellular carcinoma in mice irradiated with 1 GeV/nucleon Fe-56 ions. Radiat Res 2009;172(2):213–9. [PubMed: 19630525]

94. Yamamoto ML, Hafer K, Reliene R, Fleming S, Kelly O, Hacke K, Schiestl RH. Effects of 1 GeV/nucleon Fe-56 particles on longevity, carcinogenesis and neuromotor ability in Atm-deficient mice. Radiat Res 2011;175(2):231–9. [PubMed: 21268717]

95. Kim EH, Park AK, Dong SM, Ahn JH, Park WY. Global analysis of CpG methylation reveals epigenetic control of the radiosensitivity in lung cancer cell lines. Oncogene 2010;29(33):4725–31. [PubMed: 20531302]

96. Iwagami S, Baba Y, Watanabe M, Shigaki H, Miyake K, Ishimoto T, Iwatsuki M, Sakamaki K, Ohashi Y, Baba H. LINE-1 hypomethylation is associated with a poor prognosis among patients with curatively resected esophageal squamous cell carcinoma. Ann Surg 2013;257(3):449–55. [PubMed: 23023202]

97. Fenaux P, Mufti GJ, Hellstrom-Lindberg E, Santini V, Finelli C, Giagounidis A, Schoch R, Gattermann N, Sanz G, List A, Gore SD, Seymour JF, Bennett JM, Byrd J, Backstrom J, Zimmerman L, McKenzie D, Beach C, Silverman LR; International Vidaza High-Risk MDS

Survival Study Group. Efficacy of azacitidine compared with that of conventional care regimens in the treatment of higher-risk myelodysplastic syndromes: a randomised, open-label, phase III study. Lancet Oncol 2009;10(3):223–32. [PubMed: 19230772]

98. Lübbert M, Suciu S, Hagemeijer A, Rüter B, Platzbecker U, Giagounidis A, Selleslag D, Labar B, Germing U, Salih HR, Muus P, Pflüger KH, Schaefer HE, Bogatyreva L, Aul C, de Witte T, Ganser A, Becker H, Huls G, van der Helm L, Vellenga E, Baron F, Marie JP, Wijermans PW; EORTC Leukemia Group and the German MDS Study Group. Decitabine improves progression-free survival in older high-risk MDS patients with multiple autosomal monosomies: results of a subgroup analysis of the randomized phase III study 06011 of the EORTC Leukemia Cooperative Group and German MDS Study Group. Ann Hematol 2016;95(2):191–9. [PubMed: 26596971]
99. Stewart DJ, Issa JP, Kurzrock R, Nunez MI, Jelinek J, Hong D, Oki Y, Guo Z, Gupta S, Wistuba II.. Decitabine effect on tumor global DNA methylation and other parameters in a phase i trial in refractory solid tumors and lymphomas. Clin Cancer Res 2009;15(11):3881–8. [PubMed: 19470736]
100. Dote H, Cerna D, Burgan WE, Carter DJ, Cerra MA, Hollingshead MG, Camphausen K, Tofilon PJ. Enhancement of in vitro and in vivo tumor cell radiosensitivity by the DNA methylation inhibitor zebularine. Clin Cancer Res 2005; 11(12):4571–9. [PubMed: 15958643]
101. Qiu H, Yashiro M, Shinto O, Matsuzaki T, Hirakawa K. DNA methyltransferase inhibitor 5-aza-CdR enhances the radiosensitivity of gastric cancer cells. Cancer Sci 2009; 100(1):181–8. [PubMed: 19037991]
102. Hofstetter B, Niemierko A, Forrer C, Benhattar J, Albertini V, Pruschy M, Bosman FT, Catapano CV, Ciernik IF. Impact of genomic methylation on radiation sensitivity of colorectal carcinoma. Int J Radiat Oncol 2010;76(5):1512–9.
103. Brieger J, Mann SA, Pongsapich W, Koutsimpelas D, Fruth K, Mann WJ. Pharmacological genome demethylation increases radiosensitivity of head and neck squamous carcinoma cells. Int J Mol Med 2012;29(3):505–9. [PubMed: 22109647]
104. Jiang W, Li YQ, Liu N, Sun Y, He QM, Jiang N, Xu YF, Chen L, Ma J. 5-Azacytidine enhances the radiosensitivity of CNE2 and SUNE1 cells in vitro and in vivo possibly by altering DNA methylation. PLoS One 2014;9(4): e93273. [PubMed: 24691157]
105. Agrawal V, Alpini SEJ, Stone EM, Frenkel EP, Frankel AE. Targeting methionine auxotrophy in cancer: Discovery and exploration. Expert Opin Biol Ther 2012;12(1):53–61. [PubMed: 22171665]
106. Hoffman RM. Development of recombinant methioninase to target the general cancer-specific metabolic defect of methionine dependence: a 40-year odyssey. Expert Opin Biol Ther 2015;15(1):21–31. [PubMed: 25439528]
107. Kokkinakis DM, Brickner AG, Kirkwood JM, Liu XY, Goldwasser JE, Kastrama A, Sander C, Bocangel D, Chada S. Mitotic arrest, apoptosis, and sensitization to chemotherapy of melanomas by methionine deprivation stress. Mol Cancer Res 2006;4(8):575–89. [PubMed: 16908595]
108. Strekalova E, Malin D, Good DM, Cryns VL. Methionine deprivation induces a targetable vulnerability in triplenegative breast cancer cells by enhancing TRAIL receptor-2 expression. Clin Cancer Res 2015;21(12):2780–91. [PubMed: 25724522]
109. Guenin S, Morvan D, Thivat E, Stepien G, Demidem A. Combined methionine deprivation and chloroethylnitrosourea have time-dependent therapeutic synergy on melanoma tumors that NMR spectroscopy-based metabolomics explains by methionine and phospholipid metabolism reprogramming. Nutr Cancer 2009;61(4):518–29. [PubMed: 19838924]
110. Durando X, Thivat E, Farges MC, Cellarier E, D'Incan M, Demidem A, Vasson MP, Barthomeuf C, Chollet P. Optimal methionine-free diet duration for nitrourea treatment: A phase I clinical trial. Nutr Cancer 2008;60(1):23–30. [PubMed: 18444132]
111. Thivat E, Durando X, Demidem A, Farges MC, Rapp M, Cellarier E, Guenin S, D'Incan M, Vasson MP, Chollet P. A methionine-free diet associated with nitrosourea treatment down-regulates methylguanine-DNA methyl transferase activity in patients with metastatic cancer. Anticancer Res 2007;27(4C):2779–83. [PubMed: 17695447]
112. Beetstra S, Thomas P, Salisbury C, Turner J, Fenech M. Folic acid deficiency increases chromosomal instability, chromosome 21 aneuploidy and sensitivity to radiation-induced micronuclei. Mutat Res 2005;578(1–2):317–26. [PubMed: 16005909]

113. Leopardi P, Marcon F, Caiola S, Cafolla A, Siniscalchi E, Zijno A, Crebelli R. Effects of folic acid deficiency and MTHFR C677T polymorphism on spontaneous and radiation-induced micronuclei in human lymphocytes. Mutagen 2006;21(5):327–33.

114. Epner DE, Morrow S, Wilcox M, Houghton JL. Nutrient intake and nutritional indexes in adults with metastatic cancer on a phase I clinical trial of dietary methionine restriction. Nutr Cancer 2002;42(2):158–66. [PubMed: 12416254]

115. Koteish A, Diehl AM. Animal models of steatosis. Semin Liver Dis 2001;21(1):89–104. [PubMed: 11296700]

116. Brown JM, Brenner DJ, Carlson DJ. Dose escalation, not new biology, can account for the efficacy of stereotactic body radiation therapy with non-small cell lung cancer. Int J Radiat Oncol 2013;85(5):1159–60.

117. Song CW, Lee YJ, Griffin RJ, Park I, Koonce NA, Hui S, Kim MS, Dusenbery KE, Sperduto PW, Cho LC. Indirect tumor cell death after high-dose hypofractionated irradiation: implications for stereotactic body radiation therapy and stereotactic radiation surgery. Int J Radiat Oncol 2015;93(1):166–72.

118. Song CW, Cho LC, Yuan JL, Dusenbery KE, Griffin RJ, Levitt SH. Radiobiology of stereotactic body radiation therapy/stereotactic radiosurgery and the linear-quadratic model. Int J Radiat Oncol 2013;87(1):18–9.