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Review

Polymerase-tautomeric cancer risk model: the formation of 100% mutations is due to exposure to mutagens

Helen A. Grebneva*

Donetsk Physical and Technical Institute NAS of Ukraine, Kiev, Ukraine.

ABSTRACT

Typically, mutations that cause cancer are divided into mutations caused by hereditary factors and those caused by environmental factors. Recently, it has been hypothesized that there is a third source of mutations; these mutations appear as a result of random errors that occur during normal replication of stem cell DNA. Based on this hypothesis, it was concluded that the formation of about 67% of all mutations is not caused by exposure to any mutagens. In this paper, investigation is conducted to determine which part of mutations is due to the action of mutagens. The mechanisms of the formation of targeted base substitution mutations, targeted insertions, targeted deletions and targeted complex mutations are analyzed. The mechanisms of formation of untargeted, targeted delayed and untargeted delayed base substitution mutations are analyzed. The analysis is based on polymerase-tautomeric models for formation of various types of mutations. It was shown that all analyzed types of mutations are caused by mutagens. It is concluded that the prevention and treatment of cancer must necessarily include the removal of these mutagens from organisms. This will be the most effective, safe and cheapest way to prevent and possibly treat cancer.

KEYWORDS: cancer, targeted mutations, untargeted mutations, targeted delayed mutations, untargeted delayed mutations, cancer prevention, cancer treatment.

*Email id: grebneva@gmail.com

1. Introduction

1.1. The importance of mutations in tumor formation

Mutations, errors of DNA text, which are inherited, result in aging [1], genetic diseases [2] and cancer [3, 4]. Over the past decade, comprehensive sequencing efforts have revealed the genomic landscapes of common forms of human cancer. To date, these studies have revealed ~140 genes that, when altered by intragenic mutations can promote or "drive" tumorigenesis. A typical tumor contains two to eight of these "driver gene" mutations; the remaining mutations are passengers that confer no selective growth advantage. Mutation of the driver gene in physiological terms is defined as a factor providing a selective advantage in cell growth [5]. The development of solid tumors usually requires five to eight mutations in the driver genes [6]. The remaining mutations are called passenger mutations; they do not give a selective advantage in growth. The rest of the mutations are called passenger mutations and do not provide a selective growth advantage. Typically, driver genes are responsible for maintaining the genome [3, 4].

Somatic mutations (mutations in tissues) that can cause tumor growth are detected in tumors. In common tumors an average of 33 to 66 genes displays subtle somatic mutations [7-10]. About 95% of these mutations are single base substitution mutations, whereas other mutations are deletions or insertions of one or a few bases [3]. Tumors in children and leukemia contain an average of 9.6 point mutations per tumor cell [3]. Melanomas

and lung tumors contain much more mutations, about 200 mutations per tumor cell [11]. Tumor cells with DNA repair defects may contain thousands of mutations [12]. The frequency of formation of point mutations in tumors is similar to the frequency of mutations in normal cells [13]. In normal cells, the mutation rate in the genome changes by more than 100 times [14]. In tumor cells, differences in mutation rates in the genome may be even higher [15].

Groups of investigators analyzed the coding sequences of more than 20,000 genes from 24 advanced pancreatic adenocarcinomas [14] and glioblastomas [15, 16]. From the initial discovery screen, in the case of pancreatic cancer, more than 1327 genes were mutated in at least one sample [14]. In the case of glioblastomas, more than 685 genes were mutated in at least one sample [15, 16]. In primary lung adenocarcinomas, out of 623 cancer-related genes, more than half of genes (356 genes) were mutated at least once [17]. Only a few genes harbored point mutations, amplifications, or deletions with reasonable high frequencies [14, 15, 17-20].

1.2. Cancer models

Currently, the somatic mutation theory is generally accepted [21-26]. It has been confirmed by genome analysis [3, 4, 22, 27, 28]. The discovery of DNA as the genetic material and the observation that cancerous changes are transmitted from one generation of cells to the next pointed to DNA as the critical target of carcinogens [5]. According to the epigenetic theory, hereditary changes in genes contribute to carcinogenesis by increasing chromosomal instability [29]. According to the chromosomal theory, a carcinogen induces random aneuploidy [30, 31]. Cancer stem-cell theory suggests that carcinogens cause cancer by altering normal stem cells [32, 33]. Under the tissue organization field theory, cancer arises from disruption of tissue microarchitecture [18, 19, 34]. A by-product of a sufficiently long disruption of the morphostat gradient is genetic instability. The authors of this theory suggest that such a collapse is not necessarily related to an effect of mutations [18, 19, 34]. Genomic instability is a fundamental process of almost all human cancers [4].

"The mutator hypothesis" suggests that genomic instability present in precancerous cells acts as a driving force for tumor development by gradually elevating the rate of spontaneous mutation [35-37]. The oncogene-induced DNA replication stress model has been established and now is widely accepted as a leading hypothesis for sporadic cancer development [38-42].

"Oncogene-induced DNA replication stress model" [38] is based on the hypothesis that activated oncogenes induce genomic instability by causing DNA replication stress and associated DSBs. DNA replication stress happens when cell replication is significantly promoted during tumorigenesis, resulting in mutations in specific genomic sites, which was described as "common fragile sites" [43]. It is unclear how oncogene-induced replication stress causes various genomic instabilities particularly for tumor suppressor or the oncogenic pathway itself [44, 45]. At present, it is not possible to explain the instability of the genome by mutations in auxiliary genes. Therefore, it has not yet been possible to develop molecular mechanisms behind human carcinogenesis [20, 46-50]. Mutation analyses [51, 52] support the theory of "a few oncogenes induced genomic instabilities" as a reasonable theory for underlying mechanism of the development of sporadic cancer.

1.3. Cancer risk factors

Usually, mutations that arise from cancer are classified into mutations caused by hereditary factors and environmental factors. Risk factors caused by environmental factors include ultraviolet radiation, smoking, alcohol use, or human papilloma virus [53]. Risk factors of cancer include free radicals formed in the processes of metabolism; free radicals are the main cause of spontaneous mutagenesis [54]. In addition, they include heavy metals and other chemicals that can cause mutations and damage the DNA molecule. Such substances were found in patients with cardiovascular and cancer diseases [55]. It is believed that 5-10% of cancers are caused by hereditary diseases [56, 57].

Nearly 48 chemical agents were already known to induce somatic-cell mutations [58]. The initiating mutations of a tumor are present in each of the cancerous cells comprising the tumor [59].

Of 48 agents evaluated for induction of germ-cell mutations in rodents, 39 of them are positive [60-68]. The published rodent germ-cell mutagens include ionizing radiation; cancer chemotherapy agents; food components (acrylamide); environmental contaminants (benzo[*a*]pyrene); and common complex mixtures to which nearly everyone is exposed, such as main- and side-stream tobacco smoke, diesel exhaust, and the particulate fraction of air pollution [69-72]. Compounds capable of alkylating DNA cause germ cell mutations and subsequent mutations in the offspring of exposed males [73]. It is demonstrated that *de novo* mutations underlie many diseases such as schizophrenia, autism, epilepsy, and intellectual disability [74-77]. DeMarini [78] showed that different mutagens induce the same primary class of base substitution mutations, insertions and deletions in most organisms, thus reflecting the conserved nature of DNA replication and repair processes. They lead to aging, cancer, hereditary diseases, diabetes, and other chronic diseases [1-4, 72].

The authors of ref. [79] suggested that the fact that some tissues cause cancer in humans much more often than other tissues [80] can be explained by the number of stem cell divisions. It turned out that the average life expectancy of patients with various types of cancer is strongly correlated with the estimated number of normal stem cell divisions in the corresponding tissues, that occur throughout the patient's life [79]. On this basis, the authors of ref. [79] hypothesized that random errors arising from DNA replication in normal stem cells are the main factor contributing to the development of cancer. In other words, they suggested that there is a third source of mutations; these mutations appear as a result of random errors that occur during normal DNA replication. In other words, according to the cancer risk model [79], the formation of about 67% of all mutations is not caused by exposure to any mutagens. The authors conclude that no cancer prevention measures can affect this part of mutagenesis [79].

However, there are facts that contradict this hypothesis. Experiments show that with the combined action of psoralen and irradiation of a DNA molecule with ultraviolet light, 90% of all mutations are untargeted delayed base substitution mutations [81]. A polymerase-tautomeric model for untargeted delayed base substitution mutations formation shows [82] that all these mutations are formed opposite DNA bases in rare tautomeric forms and only when there are many other DNA damages in small neighborhoods from these bases. On this basis, it was concluded that the hypothesis that in 67% of cases the risk of malignant tumors is due to random mutations that occur during normal DNA replication [79] is erroneous.

Therefore, it is currently relevant and of great practical importance to study the question of what contribution various factors provide to the probability of cancer risk. In order to solve this problem, it is necessary to analyze the formation mechanisms of all types of mutations. First of all, it is necessary to compare the currently existing models of mutagenesis.

2. Features of the formation of mutations

2.1. Types of mutations

The mutations which occur at the same position as the photoproduct are targeted [83, 84]. Cyclobutane pyrimidine dimers and (6-4) photoproducts cause targeted substitution mutations (transitions and transversions) [85, 86], targeted deletions and targeted insertions [87], targeted complex frameshift mutations [88]. Sometimes mutations are formed in the vicinity of damage, a process that is termed untargeted mutagenesis [89]. In the case of ultraviolet mutagenesis, untargeted mutations appear in a small vicinity of photodimers [89]. Untargeted base substitution mutations, untargeted insertions, and untargeted deletions can occur [90, 91].

The conventional view of radiation mutagenesis is that radiation induces most mutations in cells shortly after irradiation [92]. Delayed mutations are mutations that occur in the progeny of the irradiated cell after many generations of cell division [93]. The delayed mutations are usually point mutations and more than half of them are base substitution mutations [94]. Delayed mutations can form opposite to lesions that can stop DNA synthesis. Accordingly, delayed targeted mutations can be formed [81]. Targeted delayed base substitution mutations, targeted delayed insertions,

and targeted delayed deletions can be formed [95]. Delayed mutations can form in so-called undamaged DNA sites. Accordingly, untargeted delayed mutations can be formed [81]. Untargeted delayed base substitution mutations, untargeted delayed insertions, and untargeted delayed deletions can be formed [95]. Mutations occur during error-prone and SOS replication, repair or transcription [96-99]. Targeted, untargeted and targeted delayed and untargeted delayed mutations can significantly contribute to genomic instability, cancer and genetic diseases [100, 101].

2.2. Features of DNA synthesis, which can lead to mutations

As a rule, DNA synthesis is a highly accurate process. If during DNA synthesis an error-free DNA polymerase, such as DNA polymerase III *E coli* or DNA polymerase ε of mammals, encounters DNA damage such as a pyrimidine cyclobutane dimer, the synthesis will stop, a gap will form opposite the dimer, which can later be fixed by repair. If there are lesions that can stop DNA synthesis, such as pyrimidine cyclobutane dimers, then SOS or an error-prone system is induced. DNA synthesis begins to happen in a new way. The so-called translesion synthesis begins to act. Specialized DNA polymerases that are capable of incorporating bases opposite to cyclobutane pyrimidine dimers are involved in translesion synthesis [102]. Replication on the damaged DNA template, however, leads to mutations.

If an erroneous base is inserted during the replication process, it is usually removed by $3' \rightarrow 5'$ exonuclease activity [103, 104]. In addition, the sliding clamp mechanism may be activated; sliding clamp presses the DNA polymerase onto the matrix and prevents the removal of the "wrong base". As a result, mutations may appear. Mutations are always formed during DNA synthesis in processes prone to errors or SOS replication, repair, or transcription [96-99].

3. Early models for mutagenesis

The entire genetic program for the development, functioning, growth and reproduction of all organisms is written in the DNA molecule. Both strands of double-stranded DNA store the same biological information. This information is written using four letters. The role of letters is played by molecules called DNA bases. The canonical bases of DNA include guanine, cytosine, adenine and thymine. In a DNA double helix, bases on one strand form hydrogen bonds with bases on the other strand. Adenine binds only to thymine, and cytosine only to guanine. This is called complementary base pairing or Watson-Crick base pairs. This is the basis of accurate DNA synthesis [1].

In 1953, Watson and Crick proposed a DNA double helix structure model and suggested that spontaneous mutagenesis may be caused by the ability of DNA bases to be in rare tautomeric forms. Bases in rare tautomeric forms are DNA bases to which one or more hydrogen atoms have been added or which have lost one or more hydrogen atoms. Watson and Crick suggested that DNA bases can change their tautomeric state due to interactions with water molecules [105].

Currently, the polymerase model of mutagenesis is generally accepted. The polymerase model is based on the idea that the cause of mutations is random DNA polymerase errors. It is assumed that polymerases are inserted opposite damage (for example, cyclobutane pyrimidine dimers) to non-complementary bases [106]. In other words, it is believed that DNA polymerases incorporate such canonical bases opposite matrix bases that cannot form hydrogen bonds with the matrix bases.

It is assumed that deamination of cytosine is the main cause of the formation of base substitution mutations. One hypothesis is that UV-induced mutations occur only after deamination of the cytosine or 5-methylcytosine within the pyrimidine dimer [107].

Untargeted mutations are mutations that appear on the so-called undamaged sites of DNA. In recent decades untargeted and untargeted delayed mutations are considered exclusively in terms of bystander effects [108]. Bystander effects are defined as the induction of cellular damage in unirradiated cells, induced by irradiated cells in the surrounding area [108].

The conventional view of radiation mutagenesis is that radiation induces most mutations in cells

shortly after irradiation. Radiation, including ionizing radiation such X-rays and charged particles (heavy ion radiation), as well as nonionizing radiation (UV light) and the DNA alkylating agent ethyl methane sulphonate, chemotherapeutic drugs, and photodynamic treatment induce genome instability many cell generations after the exposure. These delayed effects are observed after high (1-10 Gy) and very low (0.01-0.1 Gy) doses of ionizing radiation [92].

Experimental data suggest a specific, and perhaps unique, role for radiation-induced genome instability as a critical early event associated with initiation of the carcinogenic process. In other words, radiation-induced genome instability is a critical early event in the multi-step sequence leading to radiation-induced cancer [100]. Radiation-induced genome instability and radiation-induced bystander effects have been described in ref. [93]. Radiation-induced genome instability refers to biological effects that occur in the descendants of irradiated cells through many generations of cell division. Delayed mutations are mutations that occur in the progeny of the irradiated cells after many generations of cell division. Thus, at present the mechanism of delayed mutation formation and genome instability is not clear [93].

4. Polymerase-tautomeric models for mutagenesis

The polymerase-tautomeric models for targeted ultraviolet mutagenesis [109-120], radiation-induced bystander effects [117-120, 121-125], radiation-induced genomic instability [115, 117-120, 125-127] and *A-rule* [128] have been proposed. A mechanism for the formation of rare tautomeric forms of DNA bases [128-130] is proposed and it has been shown that the formation of five rare tautomeric forms of thymine and adenine [129, 130] and seven of guanine and cytosine [115, 131] is possible. Mechanisms for the formation of targeted base substitution mutations [123, 132], targeted insertions [110, 111], targeted deletions [112, 113], targeted delayed base substitution mutations [115, 126, 127] and targeted complex insertions [114] under error-prone and SOS synthesis of DNA containing *cis-syn* cyclobutane pyrimidine dimers have been developed. In addition, a mechanism for formation of the hot and cold spots of ultraviolet mutagenesis has been proposed [133].

Mechanisms for the formation of untargeted base substitution mutations [118, 121, 122, 124] and untargeted insertions [125] have been developed. Their source is DNA bases in certain rare tautomeric forms located in small neighborhoods of cyclobutane dimers [123]. A detailed substantiation of the polymerase-tautomeric models of radiation-induced bystander effects is given in ref. [124]. A mechanism for the formation of targeted delayed base substitution mutations caused by *cis-syn* cyclobutane thymine dimers [115, 118, 126] and cytosine dimers [127] have been developed. A mechanism for the formation of untargeted delayed base substitution mutations [82] has been developed. It has been shown [82] that the conclusions drawn from the cancer risk model [79] that 67% of all mutations are not caused by exposure to any mutagens are erroneous. This is true, at least with respect to untargeted delayed base substitution mutations [82].

Polymerase-tautomeric models for targeted ultraviolet mutagenesis, radiation-induced bystander effects and radiation-induced genomic instability [82, 109-133], are based on the idea by Watson and Crick [105] that one of the causes of mutagenesis is the ability of DNA bases to exist in various tautomeric forms. Experimental studies in which noncanonical base pairs of guanine – thymine [134] and cytosine – adenine [135] with one of the bases in rare tautomeric forms were found in the active centers of DNA polymerases unambiguously demonstrate that tautomeric base pairs can form in active sites of polymerase [134, 135]. This provides strong support for the ideas of Watson and Crick [105] and the polymerase-tautomeric models for mutagenesis [109-130] through direct structural evidence [134, 135].

5. Analysis of the mechanisms of the formation of targeted mutations

Targeted mutations are mutations that are formed opposite to damage that can stop DNA synthesis [85, 86]. Such damage is always caused by radiation, ultraviolet radiation, free radicals, other chemicals or

other external influences. Therefore, the question of whether the source of the targeted mutations is the effect of mutagens or some other reason never arose. Nevertheless, in order to compare the details and extent of the effect of mutagens on DNA during the formation of targeted mutations on the one hand and untargeted and delayed mutations on the other hand, it is useful to analyze the details of the mechanisms of the formation of targeted mutations.

5.1. Analysis of the mechanisms of the formation of targeted base substitution mutations

Base substitution mutations are called mutations when one base is replaced by another [85, 86]. Targeted base substitution mutations are formed opposite to lesions that can stop DNA synthesis [136].

The rare T^*_1, T^*_4 and T^*_5 thymine tautomers cannot form hydrogen bonds with canonical tautomers of adenine for steric reasons [109]. The structural analysis indicates that the C_1^*, C_2^*, C_5^* and C^*_6 cytosine tautomers cannot form hydrogen bonds with canonical tautomers of guanine for steric reasons [116]. But canonical tautomeric forms of other DNA bases can be incorporated opposite T^*_1, T^*_4, T^*_5, C_1^*, C_2^*, C_5^* and C^*_6. As a result, cyclobutane dimers with bases in rare tautomeric forms C_1^*, C_2^*, C_5^* and C^*_6, T^*_1, T^*_4 and T^*_5 will lead to targeted base substitution mutations [109, 116, 129].

5.2. Analysis of the mechanisms of formation of targeted insertions and targeted deletions

Insertions are the structural DNA changes wherein one DNA strand becomes longer than the other as a result of an insertion of a number of nucleotides [137]. Insertions may be targeted and untargeted types [138]. Frameshift mutations often account for approximately one-third of all mutations [139]. Frameshift mutations most commonly arise in DNA sites with a homogenous nucleotide composition, such as monotonous runs of G-C or A-T pairs or sequences with alternating A-T and T-A pairs. Now it is still unclear how frameshift mutations arise at cyclobutane pyrimidine dimers. The Streisinger model [140] is now the best-grounded model of frameshift mutations [141-145] suggesting gaps and DNA strand slippage during synthesis as the causes of mutations.

Structural analysis of the incorporation of canonical DNA bases opposite the *cis-syn* cyclobutane thymine dimer TT_2^* [112] and *cis-syn* cyclobutane cytosine dimers CC_3^* and CC_7^* [110] showed that it is impossible to insert any canonical base opposite them so that hydrogen bonds are formed between the C_3^* or C_7^* or T_2^* bases and the canonical DNA bases. A one-nucleotide gap arises opposite to a *cis-syn* cyclobutane dimer TT_2^* as a result of translesion synthesis driven by modified *E. coli* DNA polymerase III or mammalian DNA polymerase δ or ε or specialized (mammalian Pol η or Pol ζ or *E. coli* DNA polymerase IV or V) DNA polymerases. As was demonstrated experimentally, such a gap arises during DNA synthesis when the template contains an abasic site, leading to a one-nucleotide deletion [144]. The site in a nascent DNA strand may be lost because a bend forms in the site containing cyclobutane pyrimidine dimers and the hydrogen bonds between the bases are broken [146, 147]. A DNA site containing the *cis-syn* cyclobutane dimers TT_2^*, may form a loop. The resulting smaller gap is usually filled in by constitutive DNA polymerases, leading to the precipitation of several bases (deletion formation) [112].

In particular, *cis-syn* cyclobutane pyrimidine dimers, one or both bases of which are in such rare tautomeric forms that cannot form hydrogen bonds with canonical DNA bases, are the source of insertions or deletions [110-112, 115].

5.3. Analysis of the mechanisms of the formation of targeted complex mutations

Complex frameshifts are frameshifts with an adjacent base substitution [78, 148, 149]. If targeted complex mutations are formed, then they are caused by several DNA damage, such as cyclobutane pyrimidine dimers. At least one lesion must contain T_2^*, C_3^* or C_7^* bases that may lead to insertions or deletions, and one or more lesions must contain bases in rare tautomeric forms C_1^*, C_2^*, C_5^* and C^*_6, T^*_1, T^*_4 and T^*_5 which can lead to base substitution mutations [114].

6. Analysis of the mechanisms of the formation of untargeted mutations

If DNA bases in rare tautomeric forms are formed in small (3-5 bases) neighborhoods of cyclobutane

dimers, then these rare tautomeric states will be stable. They are preserved due to the fact that the DNA strand is bent opposite to cyclobutane dimers, as a result of which hydrogen bonds are lengthened or even broken [146, 147]. When prone to errors or SOS DNA synthesis, they can lead to untargeted mutations. Bases in rare tautomeric forms that are near to lesions such as cyclobutane dimers can lead to untargeted mutations. A detailed substantiation of the mechanism of formation of untargeted base substitution mutations is presented in ref. [124].

Let us consider a site of DNA, on which there are adenine-thymine base pairs in rare tautomeric forms in a small neighborhood of canonical cyclobutane thymine dimers. These sites are synthesized as a result of error-prone or SOS synthesis. Structural analysis indicates that canonical tautomeric forms of thymine cannot be incorporated opposite A_1*. But canonical tautomeric forms of cytosine or adenine can be incorporated opposite A_1*. Rare A_1* tautomer of adenine may result in untargeted transition A-T→G-C or untargeted homologous transversion A-T→T-A [122, 124]. A molecule of thymine can be inserted opposite A_2* and A_4* [122, 124]; a molecule of adenine can be inserted opposite T_3* [126]; it is likely they will not result in mutations. The rare A_3*, A_5* [122, 124] and T_2* [112] tautomers do not form hydrogen bonds with any canonical tautomers of DNA bases. So they cannot result in the base substitution mutations.

Let the DNA bases in rare tautomeric forms be formed in a small vicinity of cyclobutane dimers. Rare T_1* tautomer of thymine may result in A-T→G-C untargeted transition or A-T→T-A untargeted homologous transversion [122, 124]. Molecules of the thymine in rare tautomeric form T_4* may result in transversion A-T→C-G only [122, 124]. Rare T_5* tautomer can result in transversion A-T→C-G or homologous transversions A-T→T-A [122, 124].

The term untargeted mutations suggest that these mutations are formed on undamaged DNA sites. This is not true, as has been shown. It has been shown that untargeted mutations are mutations appearing on DNA damages unable to stop the synthesis of DNA. The hypothesis that untargeted mutations are formed on undamaged DNA sites was tested by biological methods only. Firmly established facts show that the so-called untargeted mutations appear on DNA sites, in which, using biological methods, no DNA damages were found. This does not mean that using other methods such as thermally stimulated luminescence, such DNA damages cannot be found. The nature of untargeted base substitution mutations cannot be explained in terms of radiation-induced bystander effects. However, this problem is easily solved within the framework of polymerase-tautomeric models. They are easily and naturally explained by polymerase-tautomeric models [122, 124].

Thus, we see that, firstly, untargeted mutations are formed as a result of the action of mutagens. Secondly, if untargeted mutations are formed, this means that either the tautomeric states in the DNA bases included in cyclobutane dimers have changed, or DNA bases in rare tautomeric forms have formed in small neighborhoods from cyclobutane dimers. And thirdly, in addition to these DNA damage, other damage is likely to have formed that can stop DNA synthesis and lead to the induction of an error-prone or SOS system.

7. Analysis of the mechanisms of formation of targeted delayed mutations

A polymerase-tautomeric model for radiation induced genomic instability: targeted delayed base substitution mutations during error-prone and SOS synthesis of double-stranded DNA, containing *cis-syn* cyclobutane cytosine [127] and thymine [126] dimers have been proposed. In order to determine which of the canonical bases will be inserted by the SOS-modified DNA-polymerase opposite *cis-syn* TT_3* cyclobutane thymine dimers and the *cis-syn* cyclobutane cytosine dimer CC_4*, consider the constraints on the formation of hydrogen bonds between the bases of the template DNA and the inserted bases. During SOS synthesis of DNA containing dimers, nucleotide bases are inserted opposite the dimers without the removal of the dimer-containing sites. This is only possible when the DNA-polymerase is pressed on the DNA by the “sliding clamp”, obstructing the operation of exonucleases, or

when the synthesis involves specialized DNA polymerases, such as *E. coli* polymerase V or IV, or when the specialized DNA-polymerase is pressed on the DNA by the "sliding clamp".

The rare T_3* thymine tautomer is capable of forming one H-bond with canonical adenine. But T_3* can form two H-bonds with canonical guanine and one H-bond with canonical cytosine and one H-bond with canonical thymine [126]. The rare C_4* cytosine tautomer is capable of forming two hydrogen bonds with canonical guanine. But C_4* can form one hydrogen bond with canonical adenine, one hydrogen bond with canonical cytosine and one H-bond with canonical thymine [127].

Consider a DNA site with a *cis-syn* TT_3* cyclobutane thymine dimer and a *cis-syn* CC_4* cyclobutane cytosine dimer. Suppose that other cyclobutane pyrimidine dimmers have formed; then they are quite far from *cis-syn* TT_3* cyclobutane thymine dimer and a *cis-syn* CC_4* cyclobutane cytosine dimer. Since the damage is only one, the synthesis through the damage will go quite quickly and with high accuracy. For example, the synthesis will be carried out using Pol III DNA polymerase of *Escherichia coli* or eukaryotic DNA polymerase δ. If a wrong nucleotide is inserted opposite the *cis-sin* cyclobutane thymine dimer TT_3* or the *cis-syn* CC_4* cyclobutane cytosine dimer, the erroneous nucleotides can be removed by 3'→5'-exonucleases. Therefore, there is a high probability that adenine will be inserted opposite thymine T_3* and guanine opposite cytosine C_4*. In this case the strand of DNA, containing *cis-sin* cyclobutane thymine dimers TT_3* or *cis-syn* CC_4* cyclobutane cytosine dimers, does not result in mutations. So many cycles of DNA replication can continue.

However, if further DNA synthesis will involve DNA polymerases having a low fidelity of synthesis, there may be base substitution mutations. Moreover, they may be formed through many cycles of replication after DNA has been damaged. Consequently, these are the delayed mutations. *Cis-syn* cyclobutane thymine dimers TT_3* may result in targeted delayed transitions T-A→C-G, targeted delayed transversions T-A→G-C and T-A→A-T [126]. *Cis-syn* cyclobutane CC_4* cytosine dimers may result in targeted delayed C-G→T-A transition, G-C→T-A transversion and homologous C-G→G-C transversion [127].

Opposite canonical thymine only cytosine can be incorporated. Canonical *cis-syn* cyclobutane thymine dimers TT may result in only targeted delayed transversions T-A→G-C [126]. Canonical tautomeric form of thymine may be formed opposite canonic tautomeric form of cytosine. Canonical *cis-syn* cyclobutane cytosine dimers CC may result in only targeted delayed transversions C-G→A-T [127].

Whether a delayed mutation will appear or not depends entirely on the presence or absence of other DNA damages near this damage. If there are no other DNA lesions or there are very few of them, then translesion synthesis will go quite accurately and mutations are not formed. If other lesions, capable of stopping the synthesis of DNA, are located next to the *cis-syn* cyclobutane cytosine dimer CC_4* or the *cis-syn* cyclobutane thymine dimers TT_3* then the synthesis will involve other specialized DNA polymerases with a lower accuracy of synthesis. As a result, transitions may appear. If there are a lot of damages, capable of stopping the synthesis of DNA, located next to the *cis-syn* cyclobutane cytosine dimer CC_4* or CC, the *cis-syn* cyclobutane thymine dimers TT_3* or TT then the synthesis will involve specialized DNA polymerases with very low synthesis accuracy; in addition, their accuracy can be further reduced by the operation of the sliding clamp. The *cis-syn* cyclobutane cytosine dimers CC_4* or TT_3* can result in targeted delayed T-A→G-C transversions and delayed homologous T-A→A-T transversions only when there are many other lesions not far from them that can stop DNA synthesis. But the canonical *cis-syn* cyclobutane cytosine dimer CC can result in the targeted delayed transversion of C-G→A-T only [127]. Canonical *cis-syn* cyclobutane thymine dimers TT may result in only targeted delayed transversions T-A→G-C [126]. New lesions may appear sometime after dimer formation. Only then will the conditions described above arise. And the *cis-syn* cyclobutane CC_4*, TT_3*, TT or CC pyrimidine

dimers can result in mutations through many cycles of replication after DNA has been damaged. Consequently, delayed mutations only occur when a lot of DNA damage is produced by the mutagens.

8. Analysis of the mechanisms of the formation of untargeted delayed mutations

Untargeted delayed mutations are mutations that can appear after several cycles of replication after exposure to the mutagen on the so-called not damaged DNA sites. In ref. [82], a polymerase-tautomeric model was proposed for a mechanism of the formation of untargeted delayed base substitution mutations caused by thymine and adenine molecules. Let's consider an error-prone and SOS synthesis of a DNA site, one strand of which contains one canonical *cis-syn* cyclobutane thymine dimer TT, and in the small vicinity of it there is thymine in the T_3* rare tautomeric form, adenine molecules in A_2* and A_4* rare tautomeric forms, as well as canonical thymine and canonical adenine. As the structural analysis of base insertion shows, adenine can be incorporated opposite thymine T_3*, but cytosine, thymine or guanine may be inserted opposite thymine T_3*. Opposite adenine in rare tautomeric form of A_2*, thymine can be incorporated, but guanine or adenine may be inserted. Opposite adenine A_4* thymine can be incorporated, but guanine may be inserted.

If next to thymine T_3*, adenine A_2* or A_4* there are no other DNA damages or there are a few of them, then synthesis through the damage will proceed quite accurately and mutations will not form.

If in the small neighborhood of the thymine in the rare tautomeric form T_3* or the adenine in the rare tautomeric form A_2* or A_4* there are other damages that can stop DNA synthesis, then the synthesis will be carried out using specialized DNA polymerases with low synthesis accuracy. DNA synthesis can also occur with the help of constitutive DNA polymerases, provided that they are pressed with a sliding clamp. As a result, the thymine in the rare tautomeric form T_3* can cause untargeted delayed T-A→C-G transition, and the adenine molecules A_2* or A_4* will not lead to a mutation [82].

If in the small neighborhood of the thymine in the rare tautomeric form T_3* or the adenine in the rare tautomeric form A_2* or A_4*, specialized DNA polymerases with very low accuracy of synthesis will be involved in the synthesis through damage. Moreover, their accuracy may be reduced by the operation of a sliding clamp. In this case, the thymine in the rare tautomeric form T_3* can cause T-A→C-G untargeted delayed transition, and can lead to T-A→G-C untargeted delayed transversion or T-A→A-T untargeted delayed homologous transversion. The adenine in the rare tautomeric form of A_2* can lead to the formation of untargeted delayed A-T→C-G transversion and untargeted delayed A-T→T-A homologous transversion. The adenine A_4* can lead to the formation of untargeted delayed A-T→C-G transversion [82].

The thymine in canonical tautomeric form can lead to untargeted delayed T-A→G-C transversion only, and the adenine in canonical tautomeric form can lead to untargeted delayed A-T→C-G transversion only [82].

It is concluded that thymine in the T_3* rare tautomeric form, which can form hydrogen bonds with both adenine and other canonical DNA bases, can be the source of untargeted delayed base substitution mutations. In addition, adenine molecules in the rare tautomeric forms A_2* and A_4*, which can form hydrogen bonds with thymine and other canonical DNA bases, can also be a source of untargeted delayed base substitution mutations. In addition, thymine and adenine in canonical tautomeric forms can also lead to untargeted delayed base substitution mutations. Whether or not untargeted delayed base substitution mutation appears, is completely dependent on the neighboring environment. Not all of these damages must be mutagenic. If these lesions are able to stop DNA synthesis, then, they can lead to synthesis through damage, cause DNA polymerase with low synthesis accuracy and, therefore, contribute to mutagenesis.

As shown earlier, the formation of five rare tautomeric forms of thymines or adenines is possible. If they are located in a small vicinity of the cyclobutane pyrimidine dimer or other damage causing the DNA strand to bend, then

these rare tautomeric states will be stable. Each of these bases in rare tautomeric forms can lead to certain types of untargeted mutations. Thus, thymine T_1*, T_4* and T_5* and adenine in the rare tautomeric form A_1* can cause only untargeted base substitution mutations [109]. Thymine T_2* can lead to untargeted frameshift mutations only, for example, to untargeted insertions [120]. Thymine in the T_3* rare tautomeric form can cause untargeted delayed base substitution mutations only. The thymine in the T_3* rare tautomeric form can cause T-A→C-G untargeted delayed transition, T-A→G-C untargeted delayed transversion or T-A→A-T untargeted delayed homologous transversion. The adenine in the A_2* rare tautomeric form can lead to the formation of untargeted delayed A-T→C-G transversion and untargeted delayed A-T→T-A homologous transversion. The adenine in the A_4* rare tautomeric form can lead to the untargeted delayed A-T→C-G transversion [82].

If there is even more damage on a DNA site, then even canonical bases can lead to untargeted delayed base substitution mutations. The canonical thymine can lead to untargeted delayed T-A→G-C transversion only, and the adenine in canonical tautomeric form can lead to untargeted delayed A-T→C-G transversion only. The canonical cytosine can lead to untargeted delayed C-G→A-T transversion, and the guanine in canonical tautomeric form can lead to untargeted delayed G-C→T-A transversion.

The source of untargeted delayed base substitution mutations is thymine in the T_3* rare tautomeric form and adenine in the A_2* and A_4* rare tautomeric forms. But even if such DNA damage appears, in most cases they will not lead to the appearance of mutations. In order for untargeted delayed mutations to form, it is necessary that there be other DNA damage. Opposite some lesions, the DNA strand must be bent, while other lesions should be able to stop DNA synthesis.

Since, under the combined action of 8-methoxy-psoralen and long-wave ultraviolet light, about 90% of the induced mutations were untargeted delayed mutations [81]; in this case, with the onset of cancer, at least 90% of the mutations were formed as a result of DNA damage. Long-wave ultraviolet caused the appearance of bases in rare tautomeric forms, and 8-methoxy-psoralen led to a curvature of the DNA strand and, as a result, stabilization of these rare tautomeric forms of DNA bases. In addition, 8-methoxy-psoralen led to induction of error-prone or SOS system.

Therefore, the conclusion drawn from the cancer risk model [79] that the formation of about 67% of all mutations is not caused by exposure to any mutagens, but is the result of normal replication, is erroneous. As we can see from the example of the formation of untargeted delayed mutations, all these mutations can appear during the induction of error prone or SOS systems only. Moreover, the synthesis should take place with the involvement of specialized DNA polymerases and even the work of a sliding clamp. This is only possible when the synthesis of DNA containing a lot of damage occurs. It contradicts experimental facts. Naturally, the conclusion of the cancer risk model [79] that no cancer prevention methods can prevent 67% of all mutations is certainly wrong.

The strategy is pretty obvious. It is necessary to find out in what form heavy metals and other substances that we received with air, water and food are. A method must be developed for their removal. As soon as we reduce the mutagenic and damaging load on DNA molecules, it is quite possible the body will cope with the tumor. Maybe, you need help to ensure that all body systems work. A deeper understanding of the mechanisms of mutation formation, and, consequently, a deeper understanding of the mechanisms of cancer formation, will allow us to develop more effective methods for the prevention and treatment of cancer.

Thus, we see that untargeted delayed mutations are generated by mutagens. Untargeted delayed mutations can only be formed when certain conditions are met. First, rare tautomeric forms T_3* of thymine and rare tautomeric forms A_2* and A_4* of adenine for thymines and adenines should be formed. Moreover, such bases in rare tautomeric forms, as a rule, should be located in the near vicinity of damages that bend the DNA strand, such as cyclobutane dimers. Second, a lot of damage must necessarily be formed, leading to the induction of an error-prone or SOS system

so that synthesis occurs with the involvement of specialized DNA polymerases, and even with the involvement of a sliding clamp. These conditions can be realized only under the influence of a large number of mutagens.

9. Analysis of large-scale mutations

Large-scale mutations are called mutations when changes occur at the chromosome level. Large-scale mutations include gene duplications, deletions and insertions of large chromosomal regions, large scale changes to the structure of chromosomes and other damage to the structure of chromosomes. The sources of such damage to the DNA molecule associated with damage to the chromosome, when breaks in the sugar-phosphate backbones occur, are undoubtedly mutagens. Here it is enough to take into account the law of conservation of energy.

10. Conclusion

Under certain conditions, mutations can result in cancer [3, 4]. It turned out that somatic mutations (mutations in tissues) can lead to cancer. The mutation frequency changes tens of times even in normal cells [14]; in tumor cells the mutation frequency changes hundreds of times [15]. Genome analysis shows [3, 4, 22, 27, 28] that somatic mutations are the main cause of cancer [21-26].

At present, it is not entirely clear what factors lead to mutations. It is believed that about 10% of cancers are caused by hereditary diseases [56, 57]. Until recently, it was assumed that most mutations are caused by environmental factors. Recently, the cancer risk model was proposed [79], in which it was concluded that the formation of about 67% of all mutations occurs in normal cells during normal DNA replication and is simply bad luck. However, it was shown in [82] that this conclusion is erroneous.

Analysis of the mechanisms of formation of targeted base substitution mutations [109, 116, 129], targeted insertions, targeted deletions [110-112, 115], targeted complex mutations [114], untargeted base substation mutations [124], targeted delayed base substitution mutations [126, 127] and untargeted delayed base substitution mutations [82] was done. It was concluded that targeted, untargeted, targeted delayed and untargeted delayed mutations are formed as a result of the action of mutagens.

The authors of the cancer risk model [79] concluded that no cancer prevention measures can affect this part of mutagenesis. Since the model [79] is wrong, this conclusion is also not true. It can be concluded that the prospects for cancer patients are not as hopeless as the authors of the work [79] try to suggest.

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CONFLICT OF INTEREST STATEMENT

There are no conflicts of interest.

REFERENCES

1. Lopez-Otin, C., Blasco, M. A., Partridge, L., Serrano, M. and Kroemer, G. 2013, Cell, 153, 1194.
2. Bauer-Mehren, A., Bundschus, M., Rautschka, M., Mayer, M. A., Sanz, F. and Furlong, L. I. 2011, PLoS One, 6, 1.
3. Vogelstein, B., Papadopoulos, N., Velculescu, V. E., Zhou, S., Diaz, Jr. L. A. and Kinzler, K. W. 2013, Science, 339, 1546.
4. Moon, J. J., Lu, A. and Moon, C. 2019, Experimental Biology and Medicine, 244, 227.
5. Thiagalingam, S., Lengauer, Ch., Leach, F. S., Schutte, M., Hahn, S. A., Overhauser, J., Willson, J. K. V., Markowitz, S., Hamilton, S. R., Kern, S. E., Kinzler, K. W. and Vogelstein, B. 1996, Nat. Genet., 13, 343.
6. Heller, R. C., Kang, S., Lam, W. M., Chen, S., Chan, C. S. and Bell, S. P. 2011, Cell, 146, 80.
7. Laurenti, E. and Dick, J. E. 2012, Ann. N. Y. Acad. Sci., 1266, 68.
8. Welch, J. S., Ley, T. J., Link, D. C., Ding, L., Mardis, E. R. and Wilson, R. K. 2012, Cell, 150, 264.

9. Yachida, S., Jones, S., Bozic, I., Antal, T., Leary, R., Fu, B., Kamiyama, M., Hruban, R. H., Eshleman, J. R., Nowak, M. A., Velculescu, V. E., Kinzler, K. W., Vogelstein, B. and Iacobuzio-Donahue, Ch. A. 2010, Nature, 467, 1114.
10. Forbes, S. A., Bindal, N., Bamford, S., Cole, Ch., Kok, Ch. Y., Beare, D., Jia, M., Shepherd, R., Leung, K., Menzies, A., Teague, J. W., Campbell, P. J., Stratton, M. R. and Futreal, P. A. 2011, Nucleic Acids Res., 39, D945.
11. Govindan, R., Ding, L., Griffith, M., Subramanian, J., Dees, N. D., Kanchi, K. L., Maher, Ch. A., Fulton, R., Fulton, L., Wallis, J., Chen, K., Walker, J., McDonald, S., Bose, R., Ornitz, D., Xiong, D., You, M., Dooling, D. J., Watson, M., Mardis, E. R. and Wilson, R. K. 2012, Cell, 150, 1121.
12. Gryfe, R. and Gallinger, S. 2001, Surgery, 130, 17.
13. Pelizzola, M. and Ecker, J. R. 2011, FEBS Lett., 585, 1994.
14. Michaelson, J. J., Shi, Y., Gujral, M., Zheng, H., Malhotra, D., Jin, X., Jian, M., Liu, G., Greer, D., Bhandari, A., Wu, W., Corominas, R., Peoples, A., Koren, A., Gore, A., Kang, Sh., Lin, G. N., Estabillo, J., Gadomski, T. H., Singh, B., Zhang, K., Akshoomoff, N., Corsello, Ch., McCarroll, S., Iakoucheva, L. M., Li, Y., Wang, J. and Sebat, J. 2012, Cell, 151, 1431.
15. Nik-Zainal, S., Alexandrov, L. B., Wedge, D. C., Loo, P. V., Greenman, Ch. D., Raine, K., Jones, D., Hinton, J., Marshall, J., Stebbings, L. A., Menzies, A., Martin, S., Leung, K., Chen, L., Leoroy, C., Ramakrishna, M., Rance, R., Lau, K. W., Mudie, L. J., Varela, I., McBride, D. J., Bignell, G. R., Cooke, S. L., Shlien, A., Gamble, J., Whitmore, I., Maddison, M., Tarpey, P. S., Davies, H. R., Papaemmanuil, E., Stephens, Ph. J., McLaren, S., Butler, A. P., Teague, J. W., Jonsoon, G., Garber, J. E., Silver, D., Miron, P., Fatima, A., Boyault, S., Langerod, A., Tutt, A., Martens, J. W. M., Aparicio, S. A. J. R., Borg, A., Salomon, A. V., Thomas, G., Borresen-Dale, A-L., Richardson, A. L., Neuberger, M. S., Futreal, P. A., Campbell, P. J. and Stratton, M. R. 2012, Cell, 149, 979.
16. Maffini, M. V., Soto, A. M., Calabro, J. M., Ucci, A. A. and Sonnenschein, C. 2004, J. Cell Sci., 117, 1495.
17. Ding, L., Getz, G., Wheeler, D. A., Mardis, E. R., McLellan, M. D., Cibulskis, K., Sougnez, C., Greulech, H., Muzny, D. M., Morgan, M. M., Fulton, L., Fulton, R. S., Zhang, Q., Wendl, M. C., Lawrence, M. S., Larson, D. E., Chen, K., Dooling, D. J., Sabo, A., Hawes, A. C., Shen, H., Jhangiani, Sh. N., Lewies, L. R., Hall, O., Zhu, Y., Mathew, T., Ren, Yalf, G. A., Yao, J., Scherer, S. E., Clerc, K., Ng, B., Milosavljevic, A., Gonzalez-Garay, Osborne, J. R., Meyer, R., Shi, X., Tang, Y., Koboldt, D. C., Lin, L., Abbott, R., Miner, T. L., Pohl, C., Fewell, G., Haipek, C., Schmidt, H., Dunford-Shore, B. H., Kraja, A., Crosby, S. D., Sawyer, Ch. S., Vickery, T., Sander, S., Robinson, J., Winckler, W., Baldwin, J., Cherieac, L. R., Dutt, A., Fennell, T., Hanna, M., Johnson, B. E., Onjfrio, R. C., Thomas, R. C., Tonon, G., Weir, B. A., Zhao, X., Ziaugra, L., Zody, M. C., Giordano, Th., Orringer, M. B., Roth, J. A., Spitz, M. R., Wistuba, I. I., Ozenberger, B., Good, P. J., Chang, A. C., Beer, D. G., Watson, M. A., Ladanyi, M., Broderick, S., Yoshizawa, A., Travis, W. D., Pao, W., Province, M. A., Weinstock, G. M., Varmus, H. E., Gabriel, S. B., Lander, E. S., Gibbs, R. A., Meyerson, M. and Wilson, R. K. 2008, Nature, 455, 1069.
18. Soto, A. M. and Sonnenschein, C. 2004, BioEssays, 26, 1097.
19. Sonnenschein, C. and Soto, A. M. 2008, Semin. Can. Biol., 18, 372.
20. Cancer Genome Atlas Research Network. 2008, Nature, 45, 1061.
21. Boveri, T. 1914, Zur Frage der Entstehung Maligner Tumoren, G. Fischer, Jena, Germany.
22. Nordling, C. O. 1953, Br. J. Cancer, 7, 68.
23. Armitage, P. and Doll, R. 1954, Br. J. Cancer, 8, 1.
24. Armitage, P. and Doll, R. 1957, Br. J. Cancer, 11, 161.

25. Knudson, A. G. Jr. 1971, Proc. Natl. Acad. Sci. USA, 68, 820.
26. Fearon, E. R. and Vogelstein, B. 1990, Cell, 61, 759.
27. Garraway, L. A. and Lander, E. S. 2013, Cell, 153, 17.
28. Stratton, M. R., Campbell, P. J. and Futreal, P. A. 2009, Nature, 458, 719.
29. Esteller, M. 2008, N. Engl. J. Med., 359, 1148.
30. Duesberg, P., Li, R., Fabarius, A. and Hehlmann, R. 2005, Cell Oncol., 27, 293.
31. Rowley, J. D. 2009, Annu. Rev. Genom. Hum. Genet., 10, 1.
32. Bruce, W. R. and van Der Gaag, H. 1963, Nature, 199, 79.
33. Reya, T., Morrison, S. J., Clarke, M. F. and Weissman, I. L. 2001, Nature, 414, 105.
34. Potter, J. D. 2007, Nat. Rev. Can., 7, 464.
35. Nowell, P. C. 1976, Science, 194, 23.
36. Loeb, L. A. 1991, Can. Res., 51, 3075.
37. Loeb, L. A. and Harris, C. C. 2008, Can. Res., 6, 6863.
38. Halazonetis, T. D., Gorgoulis, V. G. and Bartek, J. 2008, Science, 319, 13525.
39. Bartkova, J., Horejsi, Z., Koed, K., Kramer, A., Tort, F., Zieger, K., Guldberg, P., Sehested, M., Nesland, J. M., Lukas, C., Ørntoft, T., Lukas, J. and Bartek, J. 2005, Nature, 43, 864.
40. Bartkova, J., Rezaei, N., Liontos, M., Karakaidos, P., Kletsas, D., Issaeva, N., Vassiliou, L. V., Kolettas, E., Niforou, K., Zoumpourlis. V. C., Takaoka, M., Nakagawa, H., Tort, F., Fugger, K., Johansson, F., Sehested, M., Andersen, C. L., Dyrskjot, L., Ørntoft, T., Lukas, J., Kittas, C., Helleday, T., Halazonetis, T. D., Bartek, J. and Gorgoulis, V. G. 2006, Nature, 444, 633.
41. Di Micco, R., Fumagalli, M., Cicalese, A., Piccinin, S., Gasparini, P., Luise, C., Schurra, C., Garre', M., Nuciforo, P. G., Bensimon, A., Maestro, R., Pelicci, P. G. and d'Adda di Fagagna, F. 2006, Nature, 444, 638.
42. Gorgoulis, V. G., Vassiliou, L. V., Karakaidos, P., Zacharatos, P., Kotsinas, A., Liloglou, T., Venere, M., Ditullio, R. A. Jr., Kastrinakis, N. G., Levy, B., Kletsas, D., Yoneta, A., Herlyn, M., Kittas, C. and Halazonetis, T. D. 2005, Nature, 434, 907.
43. Durkin, S. G. and Glover, T. W. 2007, Annu. Rev. Genet., 41, 169.
44. Brady, N., Gaymes, T. J., Cheung, M., Mufti, G. J. and Rassool, F. V. 2003, Cancer Res., 63, 1798.
45. Evangelou, K., Bartkova, J., Kotsinas, A., Pateras, I. S., Liontos, M., Velimezi, G., Kosar, M., Liloglou, T., Trougakos, I. P., Dyrskjot, L., Andersen, C. L., Papaioannou, M., Drosos, Y., Papafotiou, G., Hodny, Z., Sosa-Pineda, B., Wu, X. R., Klinakis, A., Ørntoft, T., Lukas, J., Bartek, J. and Gorgoulis, V. G. 2013, Cell Death Differ, 20, 1485.
46. Sjoblom, T., Jones, S., Wood, L. D., Parsons, D. W., Lin, J., Barber, T. D., Mandelker, D., Leary, R. J., Ptak, J., Silliman, N., Szabo, S., Buckhaults, P., Farrell, C., Meeh, P., Markowitz, S. D., Willis, J., Dawson, D., Willson, J. K., Gazdar, A. F., Hartigan, J., Wu, L., Liu, C., Parmigiani, G., Park, B. H,, Bachman, K. E., Papadopoulos, N., Vogelstein, B., Kinzler, K. W. and Velculescu, V. E. 2006, Science, 314, 268.
47. Wood, L. D., Parsons, D. W., Jones, S., Lin, J., Sjoblom, T., Leary, R. J., Ptak, J., Silliman, N., Szabo, S., Dezso, Z., Ustyanksky, V., Nikolskaya, T., Nikolsky, Y., Karchin, R., Wilson, P. A., Kaminker, J. S., Zhang, Z., Croshaw, R., Willis, J., Dawson, D., Shipitsin, M., Willson, J. K., Sukumar, S., Polyak, K., Park, B. H., Pethiyagoda, C. L., Pant, P. V., Ballinger, D. G., Sparks, A. B., Hartigan, J., Smith, D. R., Suh, E., Papadopoulos, N., Buckhaults, P., Markowitz, S. D., Parmigiani, G., Kinzler, K. W., Velculescu, V. E. and Vogelstein, B. 2007, Science, 318, 1108.
48. Jones, S., Zhang, X., Parsons, D. W., Lin, J. C., Leary, R. J., Angenendt, P., Mankoo, P., Carter, H., Kamiyama, H., Jimeno, A., Hong, S. M., Fu, B., Lin, M. T., Calhoun, E. S., Kamiyama, M., Walter, K., Nikolskaya, T., Nikolsky, Y., Hartigan, J., Smith, D. R., Hidalgo, M., Leach, S. D.,

Klein, A. P., Jaffee, E. M., Goggins, M., Maitra, A., Iacobuzio-Donahue, C., Eshleman, J. R., Kern, S. E., Hruban, R. H., Karchin, R., Papadopoulos, N., Parmigiani, G., Vogelstein, B., Velculescu, V. E. and Kinzler, K. W. 2008, Science, 321, 1801.

49. Parsons, D. W., Jones, S., Zhang, X., Lin, J. C., Leary, R. J., Angenendt, P., Mankoo, P., Carter, H., Siu, I. M., Gallia, G. L., Olivi, A., McLendon, R., Rasheed, B. A., Keir, S., Nikolskaya, T., Nikolsky, Y., Busam, D. A., Tekleab, H., Diaz, L. A. Jr., Hartigan, J., Smith, D. R., Strausberg, R. L., Marie, S. K., Shinjo, S. M., Yan, H., Riggins, G. J., Bigner, D. D., Karchin, R., Papadopoulos, N., Parmigiani, G., Vogelstein, B., Velculescu, V. E. and Kinzler, K. W. 2008, Science, 321, 1807.
50. Lengauer, C., Kinzler, K. W. and Vogelstein, B. 1997, Nature, 386, 623.
51. Parmigiani, G., Boca, S., Lin, J., Kinzler, K. W., Velculescu, V. and Vogelstein, B. 2009, Genomics, 93, 17.
52. O'Driscoll, M. 2008, Curr. Genom., 9, 137.
53. Danaei, G., Vander Hoorn, S., Lopez, A. D., Murray, C. J. and Ezzati, M. 2005, Lancet., 366, 1784.
54. Fielden, E. M. and O'Neill, P. 1991, The Early Effects of Radiation on DNA. Berlin: Springer, 230 p.
55. Khlifi, R. and Hamza-Chaffai, A. 2010, Toxicol. Appl. Pharmacol., 248, 71.
56. Fearon, E. R. 1997, Science, 278, 1043.
57. Lichtenstein, P., Holm, N. V., Verkasalo, P. K., Iliadou, A., Kaprio, J., Koskenvuo, M., Pukkala, E., Skytthe, A. and Hemminki, K. 2000, N. Engl. J. Med., 343, 78.
58. Singer, T. M. and Yauk, C. L. 2010, Environ. Mol. Mutagen., 51, 919.
59. Parsons, B. L., Culp, S. J., Manjatha, M. G. and Heflich, R. H. 2002, Carcinogenesis, 23, 943.
60. Da Costa, G. G., McDaniel-Hamilton, L. P., Heflich, R. H., Matques, M, M. and Beland, F. A. 2001, Carcinogenesis, 22, 1307.
61. Mei, N., Heflich, R. H, Choi, M. W. and Chen, T. 2004, Chem. Res. Toxicol., 17, 814.
62. Marchetti, F. and Wyrobek, A. J. 2005, Birth. Defects Res. C Embryo Today, 75, 112.
63. Glen, C. D., Smith, A. G. and Dubrova, Y. E. 2008, Can. Res., 63, 3630.
64. Von Tungeln, L. S., Churchwell, M. I., Doerge, D. R., Shaddock, J. G., McGarrity, L. J., Heflich, R. H., da Costa, G. G., Marques, M. M. and Beland, F. A. 2009, Int. J. Can., 124, 2006.
65. Marchetti, F., Rowan-Carroll, A., Williams, A., Polyzos, A., Berndt-Weis, M. L. and Yauk, C. L. 2011, Proc. Natl. Acad. Sci. USA, 10, 12811.
66. Somers, C. M. 2010, Syst. Biol. Reprod. Med., 57, 63.
67. Ding, W., Petibone, D. M., Latendresse, J. R., Pearce, M. G., Muskhelishvili, L., White, G. A., Chang, C.-W., Mittelstaedt, R. A., Shaddock, J. G., McDaniel, L. P., Doerge, D. R., Morris, S. M., Bishop, M. E., Manjanatha, G. M., Aidoo, A. and Heflich, R. H. 2012, Toxicol. Appl. Pharmacol., 261, 164.
68. McDaniel, L. P., Ding, W., Dobrovolsky, V. N., Shaddock, J. G. Jr., Mittelstaedt, R. A., Doerge, D. R. and Heflich, R. H. 2012, Mutat. Res., 742, 72.
69. DeMarini, D. M. 2004, Mutat. Res., 567, 447.
70. Yauk, C. L., Berndt, M. L., Williams, A., Rowan-Carroll, A., Douglas, G. R. and Stämpfli, M. R. 2007, Can. Res., 67, 5103.
71. Ritz, C., Ruminski, W., Hougaard, K. S., Wallin, H. and Vogel, U. 2011, Mutat. Res., 712, 55.
72. DeMarini, D. M. 2012, Environ. Mol. Mutag., 53, 166.
73. Yauk, C. L., Lambert, I. B., Meek, M. E., Douglas, G. R. and Marchetti, F. 2015, Environ. Mol. Mutagen., 56, 724.
74. Lupski, J. R. 2010, Nat. Genet., 42, 1036.
75. Morrow, E. M. 2010, J. Am. Acad. Child Adolesc. Psychiat., 49, 1091.
76. Vissers, L. E., de Ligt, J., Gilissen, C., Janssen, I., Steehouwer, M., de Vries, P., van Lier, B., Arts, P., Wieskamp, N., del Rosario, M., van Bon, B. W., Hoischen, A., de Vries, B. B., Brunner, H. G. and Veltman, J. A. 2010, Nat. Genet., 42, 1109.

77. Hehir-Kwa, J. Y., Rodríguez-Santiago, B., Vissers, L. E., de Leeuw, N., Pfundt, R., Buitelaar, J. K., Pérez-Jurado, L. A. and Veltman, J. A. 2011, J. Med. Genet., 48, 776.
78. DeMarini, D. M. 2000, Mutat. Res., 450, 5.
79. Tomasetti, C. and Vogelstein, B. 2015, Science, 347, 78.
80. Dubal, P. M., Svider, P. F., Kanumuri, V. V., Patel, A. A., Baredes, S. and Eloy, J. A. 2014, Laryngoscope, 124, 1877.
81. Boesen, J. J., Stuivenberg, S., Thyssens, C. H., Panneman, H., Darroudi, F., Lohman, P. H. and Simons, J. W. 1992, Mol. Gen. Genet., 234, 217.
82. Grebneva, H. A. 2019, J. Oncol. Res., 1, 24.
83. Lawrence, C. W., Banerjee, S. K., Borden, A. and LeClerc, J. E. 1990, Mol. Gen. Genet., 222, 166.
84. Lawrence, C. W., Gibbs, P. E., Borden, A., Horsfall, M. J. and Kilbey, B. J. 1993, Mutat. Res., 299, 157.
85. Ikehata, H., Kudo, H., Masuda, T. and Ono, T. 2003, Mutagenesis, 18, 511.
86. Santiago, M. J., Alejandre-Durán, A. and Ruiz-Rubio, M. 2006, Mutat. Res., 601, 51.
87. Heidenreich, E., Eisler, H. and Steinboeck, F. 2006, Mutat. Res., 593, 187.
88. Levine, J. G., Schaaper, R. M. and DeMarini, D. M. 1994, Genetics, 136, 731.
89. Brotcorne-Lannoye, A. and Maenhaut-Michel, G. 1986, Proc. Natl. Acad. Sci. USA, 83, 3904.
90. Huo, L., Nagasawa, H. and Little, J. B. 2001, Radiat. Res., 156, 521.
91. Kinashi, Y., Suzuki, M., Masunaga, S. and Ono, K. 2009, Appl. Radiat. Isotop., 67, S325.
92. Allen, C. P., Fujimori, A., Okayasu, R. and Nickoloff, J. A. 2013, D. Mittelman (Eds) Stress-induced mutagenesis. Springer, New York, 183.
93. Little, J. B. 2003, Oncogene, 22, 6978.
94. Little, J. B., Nagasawa, H., Pfenning, T. and Vetrovs, H. 1997, Radiat. Res., 148, 299.
95. Sargentini, N. J. and Smith, K. C. 1987, Mutat. Res., 179, 55.
96. Wang, H., Hoffman, P. D., Lawrence, C. and Hays, J. B. 2006, Environ. Mol. Mutagen., 47, 296.
97. Krutyakov, V. M. 2006, Mol. Biol. (Mosk), 40, 3.
98. Hendel, A., Ziv, O., Gueranger, Q., Geacintov, N. and Livneh, Z. 2008, DNA Repair (Amst.), 7, 1636.
99. Vasquez-Del Carpio, R., Silverstein, T. D., Lone, S., Johnson, R. E. and Prakash, L. 2011, J. Mol. Biol., 408, 252.
100. Ullrich, R. L. and Ponnaiya, B. 1998, Int. J. Radiat. Biol. 74, 747.
101. Niwa, O. 2006, J. Radiat. Res., 47(B), 25.
102. Yarosh, D. B. 2002, Mutat. Res., 509, 221.
103. Jonczyk, P., Fijalkowska, I. and Ciesla, Z. 1988, Proc. Natl. Acad. Sci. USA, 85, 2124.
104. Khare, V. and Eckert, K. A. 2002, Mutat. Res., 510, 45.
105. Watson, J. D. and Crick, F. H. C. 1953, Cold Spring Harbor Symp. Quant. Biol., 18, 123.
106. Taylor, J.-S. 2002, Mutat. Res., 510, 55.
107. Ikehata, H. and Ono, T. 2011, J. Radiat. Res., 52, 115.
108. Whiteside, J. R., Allinson, S. L., Trevor, J. and McMillan, T. J. 2011, Photochem. Photobiol., 87, 435.
109. Grebneva, H. A. 2006, Environ. Mol. Mutagen., 47, 733.
110. Grebneva, H. A. 2014, Dopovidi NAN Ukraine, 11, 156.
111. Grebneva, H. A. 2014, Mol. Biol. (Mosk), 48, 457.
112. Grebneva, H. A. 2015, J. Phot. Mat. Technol., 1, 19.
113. Grebneva, H. A. 2015, Dopovidi NAN Ukraine, 4, 124.
114. Grebneva, H. A. 2015, Dopovidi NAN Ukraine, 5, 145.
115. Grebneva, H. A. 2016, Dopovidi NAN Ukraine, 5, 101.
116. Grebneva, H. A. 2016, Int. J. Molec. Biol.: Open Access, 1, 1.
117. Grebneva, H. A. 2017, Polymerase-tautomeric model for ultraviolet mutagenesis: targeted base substitution and frameshift mutations caused by *cis-syn* cyclobutane

thymine dimers, LAP LAMBERT Academic Publishing, Saarbrucken, Germany.
118. Grebneva, H. A. 2018, Fizika Tecknika Visokih Davleniy, 28, 98.
119. Grebneva, H. A. 2019, The theory of thermal relaxation of the excitation energy of hydrogen bonds in a DNA molecule. Its contribution to ultraviolet mutagenesis, LAP LAMBERT Academic Publishing, Saarbrucken, Germany (in Russian).
120. Grebneva, H. A. 2019, Int. J. Molec. Biol.: Open Access, 4, 1.
121. Grebneva, H. A. and Ivanov, M. O. 2001, Biopol. Kletka (Ukr.), 17, 388.
122. Grebneva, H. A. 2002, Biopol. Kletka (Ukr.), 18, 394.
123. Grebneva, H. A. 2013, Dopovidi NAN Ukraine, 1, 143.
124. Grebneva, H. A. 2017, Int. J. Molec. Biol.: Open Access, 2, 1.
125. Grebneva, H. A. 2017, Vestnik Luganskogo Natl. Univ., 2, 157.
126. Grebneva, H. A. 2017, Fizika Tecknika Visokih Davleniy, 27, 131.
127. Grebneva, H. A. 2018, Int. J. Molec. Biol.: Open Access, 3, 125.
128. Grebneva, H. A. 2019, Tren. Cell Mol. Biol., 14, 51.
129. Grebneva, H. A. 2002, Biopol. Kletka (Ukr.), 18, 205.
130. Grebneva, H. A. 2003, J. Molec. Struct., 645, 133.
131. Grebneva, H. A. 2001, Dopovidi of NAN of Ukraine, 7, 165.
132. Grebneva, H. A. 2001, Biopol. Kletka (Ukr.), 17, 487.
133. Grebneva, H. A. 2012, Dopovidi NAN Ukraine, 10, 181.
134. Bebenek, K., Pedersen, L. C. and Kunkel, T. A. 2011, Proc. Natl. Acad. Sci. USA, 108, 1862.
135. Wang, W., Hellinga, H. W. and Beese, L. S. 2011, Proc. Natl. Acad. Sci. USA, 108, 17644.
136. LeClerc, J. E., Borden, A. and Lawrence, C. W. 1991, Proc. Natl. Acad. Sci. USA, 88. 9685.
137. Shibutani, S., Takeshita, M. and Grollman, A. P. 1991, Nature, 349, 431.
138. Kim, S. R., Matsui, K., Yamada, M., Gruz, P. and Nohmi, T. 2001, Mol. Genet. Genom., 266, 207.
139. Wang, C.-I. and Taylor, J.-S. 1992, Biochemistry, 31, 3671.
140. Streisinger, G., Okada, J., Emerich, J., Newrich, J., Tsugita, A., Terraghi, E. and Inouye, M. 1966, Cold Spring Harbor Symposia Quant. Biol., 31, 77.
141. Strand, M., Prolla, T. A., Liskay, R. M. and Petes, T. D. 1993, Nature, 365, 274.
142. Bzymek, M., Saveson, C. J., Feschenko, V. V. and Lovett, S. T. 1999, J. Bacteriol., 181, 477.
143. Baase, W. A., Jose, D., Ponedel, B. C., von Hippel, P. H. and Johnson, N. P. 2009, Nucleic Acids Res., 37, 1682.
144. Kobayashi, S., Valentine, M. R., Pham, P., O'Donnell, M. and Goodman, M. F. 2002, J. Biol. Chem., 277. 34198.
145. Papanicolaou, C. and Ripley, L. S. 1989, J. Molec. Biol., 207, 335.
146. Park, H. J., Zang, K., Ren, Y., Nadji, S., Sinha, N., Taylor, J.-S. and Kang, C. H. 2002, Proc. Natl. Acad. Sci. USA, 99, 15965.
147. Bdour, H, M., Kao, J. L. and Taylor, J.-S. 2006, J. Org. Chem., 71, 1640.
148. DeMarini, D. M., Shelton, M. L., Abu-Shakra, A., Szakmary, A. and Levine, J. G. 1998, Genetics, 149, 17.
149. Papanicolaou, C. and Ripley, L. S. 1989, J. Molec. Biol., 207, 335.