Mutation Research 704 (2010) 190–199

Contents lists available at ScienceDirect

Mutation Research/Reviews in Mutation Research

journal homepage: www.elsevier.com/locate/reviewsmr
Community address: www.elsevier.com/locate/mutres

Review

Effects and applications of ultrashort-lived prehydrated electrons in radiation biology and radiotherapy of cancer

Qing-Bin Lu *

Departments of Physics, Biology and Chemistry, University of Waterloo, 200 University Avenue West, Waterloo, Ontario N2L 3G1, Canada

ARTICLE INFO

Article history:
Received 26 September 2009
Received in revised form 28 December 2009
Accepted 19 January 2010
Available online 28 January 2010

Keywords:
Prehydrated electron
Radiation-induced DNA damage
Radiotherapy
Anticancer drugs
BrdU and IdU
Cisplatin

ABSTRACT

The subpicosecond-lived prehydrated electron (e_{pre}^-) is a fascinating species in radiation biology and radiotherapy of cancer. Using femtosecond time-resolved laser spectroscopy, we have recently resolved that e_{pre}^- states are electronically excited states and have lifetimes of ~180 fs and ~550 fs, after the identification and removal of a coherence spike, respectively. Notably, the weakly bound e_{pre}^- (<0 eV) has the highest yield among all the radicals generated in the cell during ionizing radiation. Recently, it has been demonstrated that dissociative electron transfer (DET) reactions of e_{pre}^- can lead to important biological effects. By direct observation of the transition states of the DET reactions, we have showed that DET reactions of e_{pre}^- play key roles in bond breakage of nucleotides and in activations of halopyrimidines as potential hypoxic radiosensitizers and of the chemotherapeutic drug cisplatin in combination with radiotherapy. This review discusses all of these findings, which may lead to improved strategies in radiotherapy of cancer, radioprotection of humans and in discovery of new anticancer drugs.

Contents

1. Introduction

Femtobiology and femtomedicine. Direct, real-time observation of molecular reactions is of significant importance in diverse fields from chemistry and biology, environmental sciences to medicine. *Femtosecond time-resolved laser spectroscopy* (fs-TRLS) is the most powerful, direct technique for real-time observation of molecular reactions. Its key strength lies in short duration laser flashes of a time scale at which reactions actually happen—femtoseconds (fs) (1 fs = 10^{-15} s). The application of fs-TRLS to chemistry led to the birth of a new field in chemistry, *femtochemistry* [1]. Although many biological effects, such as DNA damage and cell death, are rather slow processes that could be in the time scales of microseconds or longer, chemical reactions that initiate the biological effects often occur in the time scales from fs to picosecond (ps) [1]. Thus, biological events occurring at longer times are often triggered by fast processes at early times. It is therefore important to apply advanced physical methods to study biochemical reactions occurring in the ultrafast time domain in order to get a complete picture of relevant biological events. *Femtobiology* and *femtomedicine*, which involve a fusion of ultrafast laser techniques with biology and medicine, are new exciting transdisciplinary frontiers. Femtomedicine addresses important biological processes with close relevance to medical sciences by application of ultrafast laser techniques such as imaging and spectroscopy. It holds the promise of frontier advances in therapy.

* Tel.: +1 519 888 4567x33503; fax: +1 519 746 8115.
E-mail address: qblu@uwaterloo.ca.

1383-5742/$ – see front matter © 2010 Elsevier B.V. All rights reserved.
doi:10.1016/j.mrrev.2010.01.012

This is the remarkable opportunity afforded through real-time observation of biochemical reactions at the molecular level.

Role of water in radiotherapy. Radiotherapy is a major curative therapy for cancer. Exposure of living cells to ionizing radiation, such as hard X-rays and γ rays, leads to biological damage by both direct and indirect interactions with the cell components. The majority (over 66%) of the radiation energy deposited in the cell is absorbed initially by water (the cell contains 70–80% water) [2,3]. It is known that the contribution of free radicals via radiolysis of water to biological damage far exceeds that of direct action in ionizing radiation [3]. Indeed, Ito et al. [4] have observed that the yields of single-strand breaks (SSBs) and double-strand breaks (DSBs) of DNA by γ-ray radiation in an aqueous solution are three orders of magnitude higher than those for dry DNA.

It is well known that the absorption of radiation energy by water leads to ionization or excitation of H_2O molecules, generating an oxidizing $OH^•$ ($H^•$) radical and an electron in isolated volume elements called spurs. A fraction of the species react together while the remainder escape into the bulk solution. The great advantage of the radiolysis method over other methods for generating reactive intermediates and observing their reactions lies in the fact that the amount of energy absorbed by any component of the system is proportional to its electron fraction. This means that in moderately dilute (≤0.1 M) aqueous solution essentially all the energy is absorbed by the water so that the yields of the primary radicals (e^-, $OH^•$ and $H^•$) are always well known [3,5,6]. In the current context of radiobiology, the electron is believed to be rapidly solvated by surrounding H_2O molecules to form the well-known hydrated electron (e_{hyd}^-). Thus, the main free radicals are long known to be e_{hyd}^-, $OH^•$ and $H^•$ [2,3,5,6], and their quantum yields (*G* values) per 100 eV energy deposited are 2.8, 2.4 and 0.6 at 10^{-6} s after irradiation, respectively [6], as shown in Fig. 1. Despite its high yield, e_{hyd}^- is trapped in a deep potential well (at ∼−3.2 eV) and therefore ineffective at inducing biological damage [2,3,6]. Almost all of the indirect damage to DNA has long been thought to be due to attack by the *oxidizing* hydroxyl radical ($OH^•$) [2,3]. However, it was also pointed out that lesions produced in DNA by an $OH^•$ radical acting alone are unimportant and ineffective in cell killing, as they can be efficiently repaired [3,6]. In fact, there is a long-standing mystery as to how water enhances DNA damage under ionizing radiation [7,8].

Prehydrated electron in ionizing radiation. The advent of fs-TRLS has provided an unprecedented-level understanding of electron hydration dynamics. Following the first direct observation of the prehydrated electron (e_{pre}^-) by Migus et al. [9], researchers have

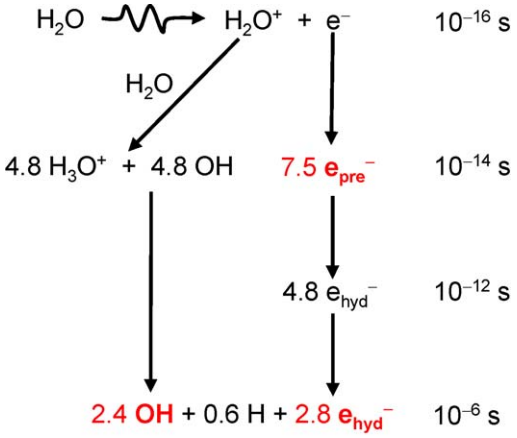

Fig. 1. Scheme for the radiolysis of water. *G*-values (molecules/100 eV) for radicals in neutral water at 10^{-16}–10^{-6} s after irradiation are also indicated [5,6]. The *G*-value of the prehydrated electron (e_{pre}^-) is obtained by assuming that about 64% of e_{pre}^- becomes the long-lived hydrated electron (e_{hyd}^-) [14].

intensely studied this ultrashort-lived species [10–19]. It is generally agreed that prior to the formation of e_{hyd}^-, the excess electron in water is located at precursor states with finite lifetimes < 1 ps (1 ps = 10^{-12} s), denoted as e_{pre}^- hereafter. However, these studies reported quite diverse lifetimes and physical properties of e_{pre}^- in water over the past two decades; the reported lifetimes ranged from 10 fs, 50 fs, 110 fs, ∼200 fs, ∼300 fs, 540 fs to ∼1 ps [9–19]. The quantum yield of e_{pre}^- is nearly double that of its ending product (e_{hyd}^-) [14] or the $OH^•$ radical. Thus, there has been significant interest in studying the reactivity of e_{pre}^- with various molecules [20–28]. In particular, there is evidence that e_{pre}^- can be attached to amino acids and nucleotides [20,21]. For example, Gauduel et al. applied fs-TRLS to observe the ultrafast one-electron reduction of oxidized pyridine nucleotides in micelles ($e_{pre}^- + Pyr^+ \rightarrow Pyr^•$), forming a pyridinyl ($Pyr^•$) radical [21], and the ultrafast low-energy electron attachment to a disulfide biomolecule (cystamine), forming a radical anion that may be relevant to the functions of enzymes, proteins or macromolecular complexes [22]. A recent review was given by Gauduel et al. [24], who emphasized that highly reactive water bridged three-body complexes [$OH^•\cdots e^-\cdots H_3O^+$] may potentially lead to ultrafast biomolecular damages triggered by ionizing radiations within an ultrashort time scale (less than 5 ps). Nevertheless, other researchers believe that the excess electron is rapidly transferred to a preexisting trap in water, forming a prehydrated electron [13,15].

Scope of this mini review. I will first briefly discuss our recent resolution of the controversies about the lifetimes and physical natures of e_{pre}^- [29]. I will then give a review of our recent findings about *dissociative electron transfer* (DET) reactions of e_{pre}^- with DNA [30] and anticancer drugs [31–34]. Specifically, we applied fs-TRLS to obtain direct observations of the intermediate states (AB^{*-}) of the DET reactions [29–34]:

$$e_{pre}^-(<0\,\text{eV}, <1\,\text{ps}) + AB \rightarrow AB^{*-} \rightarrow A^•(+B^-) \rightarrow \text{DNA damage}. \quad (1)$$

Here, AB is either a DNA base/nucleotides or an extra compound (e.g., a drug), and $A^•$ is the key dissociation product, a reactive radical that causes DNA damage. Our findings have demonstrated that DETs of e_{pre}^- may play key roles in causing damage to aqueous DNA under ionizing radiation and in activating halopyrimidines (CldU, BrdU and IdU) as potential radiosensitizers and the chemotherapeutic drug cisplatin (CDDP) in combination with radiotherapy. These findings may lead to new molecular-level understanding of the biological actions of ionizing radiation and the development of more effective drugs for therapies of cancer.

2. Resolution of the controversies about lifetimes and physical natures of e_{pre}^-

With our careful fs-TRLS measurements [29], we observed that the rise and decay kinetic traces of e_{pre}^- probed at an IR wavelength (1200–1300 nm) are strongly dependent on the experimental conditions, particularly on the *pump photon flux density*. With a low photon flux density, we observed a rise time of ∼180 fs and a decay time of ∼550 fs of e_{pre}^- (Fig. 2), independent of the pump power. With a photon flux density of 3–4 times higher, in striking contrast, there is clearly no rise time of e_{pre}^- after correction by the total instrument response function, while the decay kinetic trace of e_{pre}^- shows a strong dependence on the pump power, as shown in Fig. 3. At higher pump power, there are two decay times for e_{pre}^-, a dominant ultrashort decay of 20–30 fs and a slower decay of ∼550 fs; at low pump power, the slower decay completely disappears and only a *symmetric* narrow peak exhibits at delay (time) zero. If the sharp peak was incorrectly taken as the kinetic trace for e_{pre}^-, one could deduce a nearly zero