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FIRST ATTEMPTS AT PREDICTION OF DNA STRAND-BREAK YIELDS USING NANODOSIMETRIC DATA

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We present the first results of our attempts to correlate yields of ionisation clusters in a gas model of DNA and corresponding double-strand break (DSB) yields in irradiated plasmids, using a simple statistical model of DNA lesion formation. Based on the same statistical model, we also provide a comparison of simulated nanodosimetric data for electrons and published DSB yields obtained with the PARTRAC code.

INTRODUCTION

The most sensitive target for radiation effects in tissue is the DNA; lesions in DNA interfere with its replication and transcription and, if left unrepaired, can cause mutation and cell death. It is the hallmark of ionising radiation that it forms lesion-clusters, which have similar dimensions as the DNA itself(1). Therefore, understanding the radiation track structure on nanometric scales is of crucial importance to the evaluation of radiation damage to biological systems in general and to DNA in particular. In recent years, we have been developing ion-counting nanodosimetry(2–4) as a novel technique for studying ionisation clusters formed by charged particles on the nanometer scale, by using low-pressure gas as an expanded tissue-model. The ion-counting technique is the only experimental technique that allows probing radiation track structure with sufficient resolution to attempt modelling the initiation of DNA damage. The initiation of DNA damage is typically modelled by overlaying track structures, simulated using measured cross sections in liquid water, onto DNA models of varying complexity. The probabilities of damage formation at a location where an energy deposit overlaps the DNA or where a radiation-induced radical diffuses to the DNA are typically adjusted by comparison with biological data.

Our work takes a more basic approach, replacing the *simulated* track segment with the *measured* number of ions induced by each track segment traversing a small DNA-like gas volume, providing an ion-cluster size distribution. This approach is the only one possible in case of unknown radiation fields. Using a rather straightforward model to correlate the ion-cluster size with DNA lesion probability, we have obtained a reasonably good agreement with experimental DNA damage yields(5) measured under equivalent conditions as well as with simulated ones(6,7). We summarise below our approach and results; a more detailed description is under preparation.

DESCRIPTION OF THE MODEL

The nanodosimetric data consist of ion-cluster size distributions [$f(n_{ion})$], providing the absolute frequency of ion-clusters of various sizes formed within the adjustable sensitive volume (SV) of the nanodosemeter (ND) by a single projectile particle. The SV can be approximated by a cylinder of 1.6 mm diam. (in 120 Pa propane gas) whose length can be selected offline by applying time cuts(8). In this work we used a SV length of 2.9 mm in gas. We assume that an identical ion-cluster size distribution exists in an equivalent, 16 base-pairs, DNA segment with a 1.3 nm thick hydration layer on all sides, namely a cylinder of 4.5 nm diam. and 8 nm length. The equivalence of the gaseous and condensed sensitive volumes is discussed in detail in Ref. (9). We then assume that each ionisation in tissue has a finite probability, p_{SB}, to be converted into a strand break. The distribution of strand-break clusters (i.e. the probability that n_{ion} ions are converted to

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n_{SB} strand breaks) is then given by:

$$P_{SB}(n_{SB}) = \sum_{n_{ion}} f(n_{ion}) \times \left[\binom{n_{ion}}{n_{SB}} (p_{SB})^{n_{SB}} (1-p_{SB})^{n_{ion}-n_{SB}} \right], \quad (1)$$

where the term in brackets is the binomial distribution. A similar calculation can be performed for other types of lesions (e.g. base lesions).

Using simple combinatorics, it is possible to obtain the probability per projectile of generating specific complex lesions. For example the probability for a double-strand break (DSB), i.e. not all breaks on the same strand, is

$$P_{DSB} = \sum_{n_{ion}} \sum_{n_{SB}} f(n_{ion}) \times \left[\binom{n_{ion}}{n_{SB}} (p_{SB})^{n_{SB}} (1-p_{SB})^{n_{ion}-n_{SB}} \right] \times \left(1 - \left(\frac{1}{2}\right)^{n_{SB}-1} \right) = \sum_{n_{ion}} f(n_{ion}) \times \left(1 - 2\left(1-\frac{p_{SB}}{2}\right)^{n_{ion}} + (1-p_{SB})^{n_{ion}} \right) \quad (2)$$

To convert this to a yield of lesions, which can be compared to existing biological data, we divide by the dose per projectile and by the target mass

$$G_{DSB}\left[\mathrm{Gy}^{-1}\mathrm{Da}^{-1}\right] = 9.6 \times 10^{-10} \times \frac{\rho\left[\frac{g}{\mathrm{cm}^3}\right] V_{SV}\left[\mathrm{nm}^3\right]}{w_i[eV]\langle n_{ion}\rangle} \times \sum_{n_{ion}} \left\{ f(n_{ion}) \left(1 - 2\left(1-\frac{p_{SB}}{2}\right)^{n_{ion}} + (1-p_{SB})^{n_{ion}} \right) \right\} \quad (3)$$

where $10^{-24}\rho V_{SV}$ is the target mass in kg, w_i is the mean energy to create an ion pair in gas and $\langle n_{ion}\rangle$ is the average ion-cluster size.

Similarly, the yield of single-strand breaks (SSBs) can be calculated as

$$G_{SSB}\left[\mathrm{Gy}^{-1}\mathrm{Da}^{-1}\right] = 9.6 \times 10^{-10} \times \frac{\rho\left[\frac{g}{\mathrm{cm}^3}\right] V_{SV}\left[\mathrm{nm}^3\right]}{w_i[eV]\langle n_{ion}\rangle} \times \sum_{n_{ion}} \left\{ f(n_{ion}) \times 2\left[\left(1-\frac{p_{SB}}{2}\right)^{n_{ion}} - (1-p_{SB})^{n_{ion}} \right] \right\} \quad (4)$$

When modelling initiation of DNA damage due to low-energy electrons, we could not use the w_i value, which is poorly defined. Instead we used an effective value based on the simulated energy deposit in the ND

$$\tilde{w}_i = \frac{\Delta E}{\langle n_{ion}\rangle} \quad (5)$$

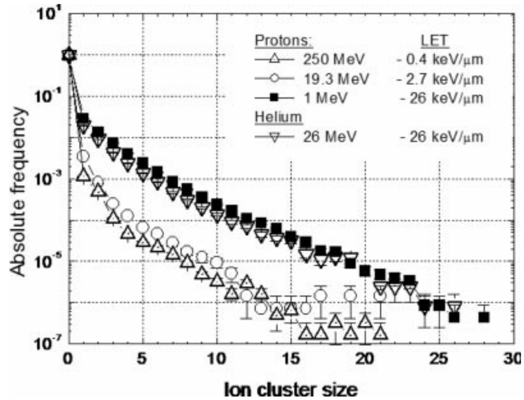

Figure 1. Examples of ion-cluster size distributions induced in the ion-counting nanodosemeter.

Alternatively, ΔE can be easily measured using standard microdosimetric devices. The need to know w_i is eliminated when looking at the ratio of DSBs and SSBs. As both are linearly dependant on w_i we can predict the complexity of DNA damage based only on parameters measured in the ND.

RESULTS AND DISCUSSION

The ion-cluster size distributions induced in gas by protons and Helium nuclei are shown in Figure 1. These data were obtained by irradiating the ND[3] with a broad particle beam, equivalent to the one used to obtain the biological data[4].

Similarly, ion-cluster size distributions, induced by a wider range of proton and Helium nuclei energies as well as by electrons, were simulated using the MC code described in appendix A of Ref. (3). We have previously shown the reasonably good agreement between the ion-cluster size distributions measured in the ND and those obtained using this code down to frequencies of 10^{-4} [3].

As biological data for comparison we used the SSB and DSB yields measured by us[5] in irradiated DNA; we further used data from[6,7], obtained using the much more detailed PARTRAC code and corroborated against measured yields in γ-irradiated cells.

Figure 2 shows the SSB and DSB yields, measured[5] and predicted from Equations 3 and 4 with $\rho V_{SV} = 1.5 \times 10^{-22}$ kg, $w_i = 28\ eV$ and $p_{SB} = 9.5\%$; the latter was fitted to obtain the best least-squares fit to the ion data. The agreement between the two is rather good.

The SSB yields of Ref. (5) are about a factor of two larger than those predicted from the nanodosimetric data. Such a discrepancy could be expected since the plasmids were irradiated in a partially scavenging solution (scavenging capacity: $3.8 \times 10^8\ \mathrm{s}^{-1}$)

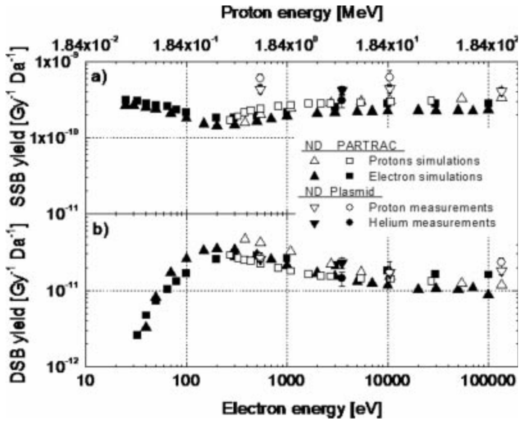

Figure 2. The predicted and measured[5] yields of (a) SSBs and (b) DSBs. The open and closed symbols refer to electrons and ions, respectively. The abscissa gives the particle velocity expressed as electron energy (bottom) and proton energy (top) (for helium ions, multiply top axis by four).

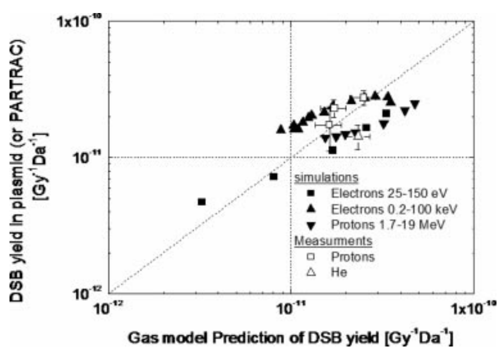

Figure 3. Correlation of the measured and modelled DSB breaks (in Gy^{-1} Da^{-1}). The dashed line denotes equality.

and were sensitive to radiation-induced radicals formed far from the DNA. Such an effect is not taken into account in the model (Equations 3 and 4) and is not present in Refs. (6) and (7).

The predictive power of our approach can be seen in Figure 3, depicting the measured yield of DSBs vs. the model-predicted ones. The agreement between the nanodosimetric prediction and the biological measurements is rather striking, considering the wide range of particles and energies.

Finally we present the ratio of DSBs to SSBs. This ratio characterises the 'complexity' of the damage, without being dependant on the deposited dose in the target. In Figure 4a, we depict this ratio as a function of DSB yield. It is clear that there is a good correlation between this ratio and the yield of DSBs, regardless of the type of radiation field. Figure 4b shows the good agreement between the biological data of Ref. (7) and the nanodosimetric data. The data of Ref. (5) is not shown.

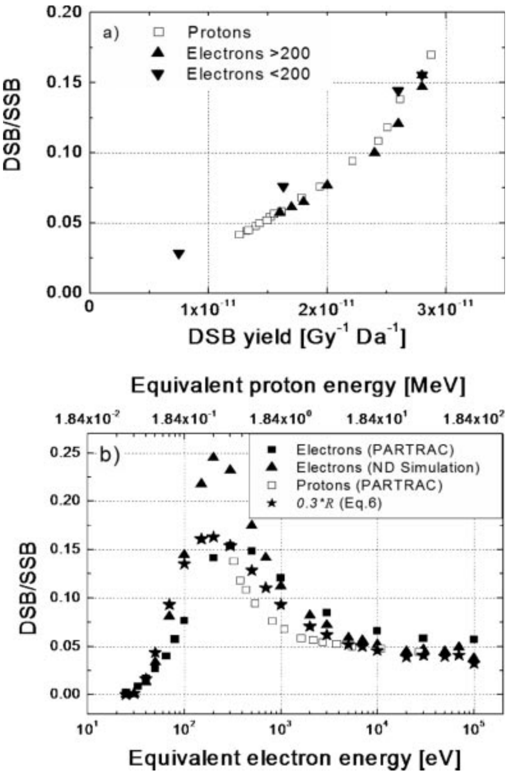

Figure 4. Ratio of yield of DSB to SSB for protons and electrons vs. (a) DSB yield and (b) energy. Electron and ion energies are scaled to have the same velocity.

It is interesting to look at the simple ratio of the frequency of ion clusters with two or more ions to the frequency of those with one or more ions

$$R = \frac{\sum_{n=2}^{\infty} f(n)}{\sum_{n=1}^{\infty} f(n)} \qquad (6)$$

This ratio is a rough approximation of the DSB/SSB ratio (see Figure 4b), where an empirical pre-factor (0.3) is required to account for the inefficiency to convert ionisations to strand breaks. This is a simplification of the more complex polynomial in p_{SB} in Equations 3 and 4. Although it follows the general trend of the DSB/SSB ratio rather well, it is not sensitive to the details of the cluster-size distribution and is only accurate at very low or very high track densities.

CONCLUSIONS

Nanodosimetry is an emerging technology for measuring ionisation clusters in small gas volumes modelling DNA-like sizes. We have demonstrated that, using ion-cluster size distributions measured in gas and a rather straightforward statistical model, it is possible to predict the complexity of DNA damage induced by ionising radiation with a similar degree of accuracy obtained with much more complex techniques.

Contrary to more complicated MC models as well as to direct biological measurements, nanodosimetry does not require any pre-existing knowledge of the radiation field. It is based only on directly measured parameters. As such it could become a tool of choice for characterisation of complex or unknown radiation fields, such as the ones encountered in space or in radiotherapeutic scenarios.

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