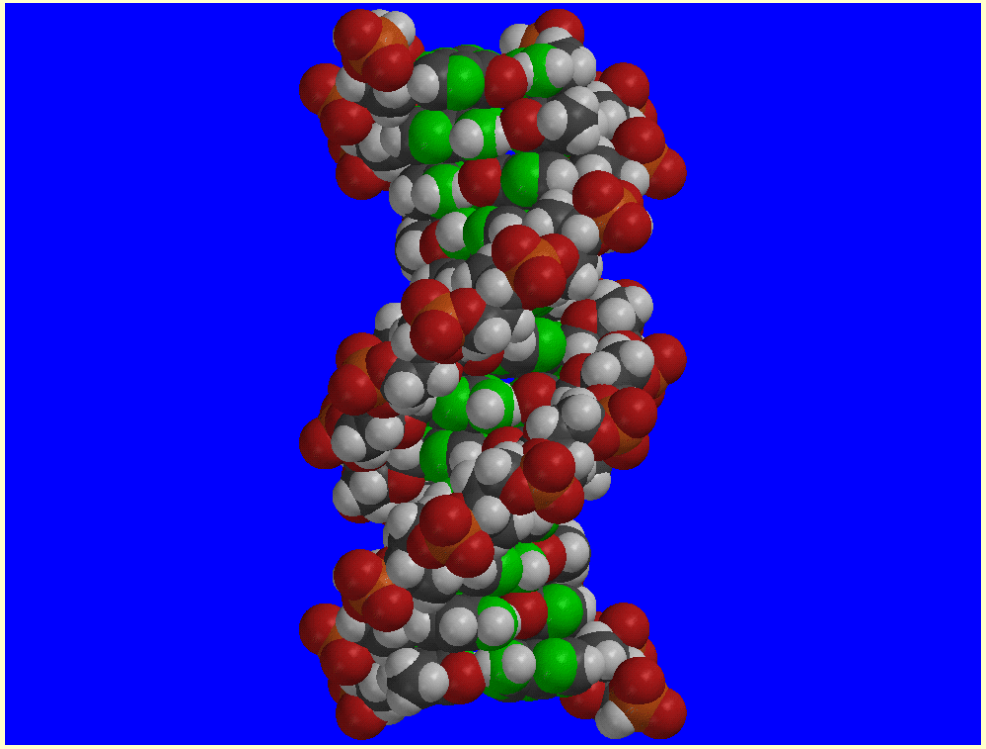
From Hydroxyl Radicals to Clustered DNA Lesions: Integrating Track-Structure Physics with Quantitative Radiation Damage Measurements



Amitava Adhikary

FRRBP, Radiation Oncology, Holden Comprehensive Cancer Center, Carver College of Medicine,

Iowa City Veterans Affairs Health Care System

The University of Iowa

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Motivation: Why does this study matter?

Biodosimetry = measuring radiation via biological damage

- Radiation dose (Gy) ≠ biological outcome
  (Joey Robertson's presentation)
- Clinical, space, and nuclear scenarios require: Biological measurement of radiation exposure

Gap:
Current biodosimetry is empirical, not mechanistic (see next slide + Joey Robertson and David Bolduc presentations)

Gap

- Initial physical-chemical damage:
  - lesions produced immediately by radiation
  - depends on radiation quality and local milieu
  - should be measured rapidly if the goal is exposure dosimetry

- Biological response to damage:
  - repair, signaling, bypass, cell-cycle effects
  - mutations, chromosome aberrations, cell death
  - depends on cell type, genotype, and post-irradiation processing

Radiation Protection Dosimetry, Vol. 97, No. 1, pp. 33–38 (2001)

Goal

Objective: A Mechanistic Framework

Bridge:

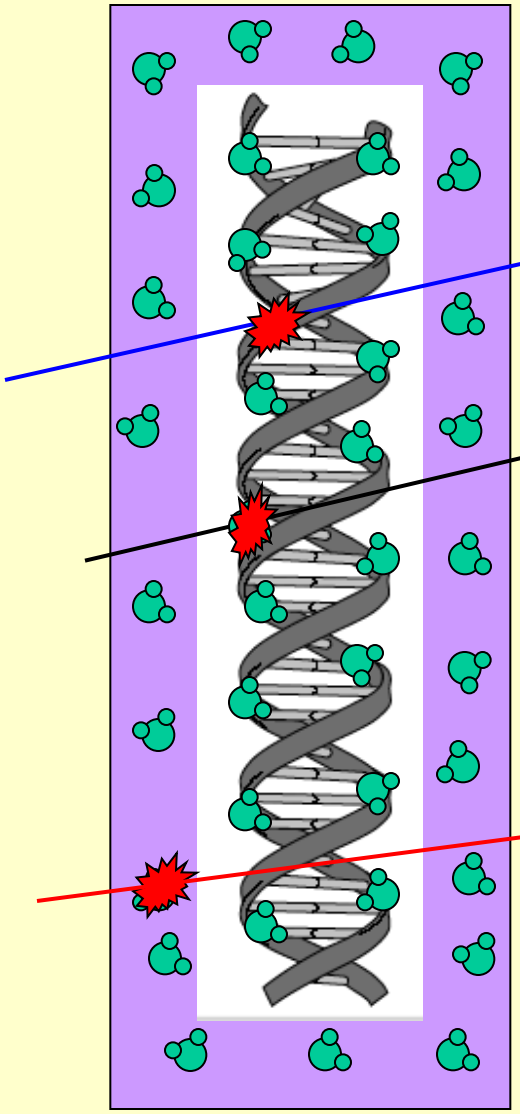
Physics → Chemistry → Biology

Develop **multi-scale framework**

Link:

- Track-structure physics
- CIRMS alignment: Quantitative, traceable radiation measurement science

Direct, Quasi-direct, Indirect effect of radiation



Direct ionization of bases, sugar-phosphate (Direct effect)

- Ionization of adjacent solvation water: fast hole/electron transfer. (Quasi-direct effect)

- Ionization of bulk water: diffusion of reactive species (•OH, e_{aq}^- + others) to DNA. (Indirect effect)

Also, Steven Swarts presentation

Methodological Approaches

Quantifying Radical and Molecular Damage

- **Free Radical Detection:** Utilizing ESR (Electron Spin Resonance) spectroscopy at low temperatures (77 K) in frozen aqueous solutions (direct and quasi-direct effect) and Pulse Radiolysis at room temperature (indirect effect) in aqueous solution to trap and quantify early-stage radicals.

- **Chemical Product Analysis:** Using **GC–MS/MS and LC–MS/MS** to quantify specific base damage, sugar lesions, and strand breaks at room temperature. (Miral Dizdaroglu and Steven Swarts)

- **Computational Modeling: Monte Carlo nanodosimetric simulations** to predict clustered DNA damage based on Linear Energy Transfer (LET) and particle fluence.

- **Dosimetry Tools:** Integrating **Bragg peak dosimetry** and **TRIM modeling** to link physical energy deposition (LET) to biological outcomes.

The Need for High-LET Precision

Understanding Complex Damage Patterns

- **LET-Resolved Analysis:** Radiation effectiveness varies along the ion track; damage products increase until just before the **Bragg Peak**, where ion-radical recombination occurs.
- **Clustered Lesions:** High-LET radiation promotes "clustered DNA damage" (multiple lesions in proximity), which is harder for cells to repair and governs biological effectiveness (RBE).
- **Spatio-Temporal Insights:** Simulations show that as LET increases, the complexity of these clusters increases, even if the total number of isolated damage sites decreases.

DNA **2024**, 4, 34–51. https://doi.org/10.3390/dna4010002

Direct-type effect
(Direct ionization and quasi-direct effect)

- Mechanisms are now quite clear
- The cell nucleus has a high global concentration of macromolecules (DNA, RNA, proteins etc.) which range from 65 to 220 mg/ml.
- From cellular perspective (equivalent to concentrated solution), direct effect and indirect effect are equally important.

Difference between dilute and concentrated solution

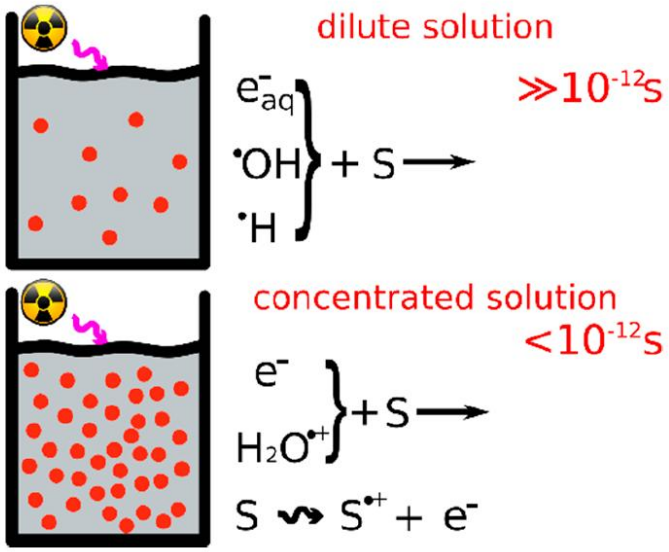
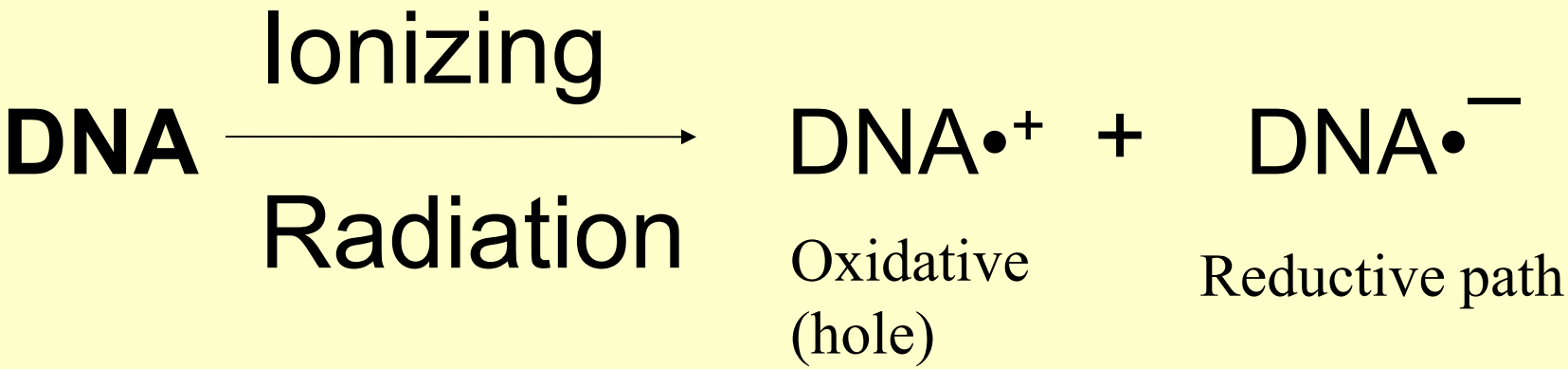


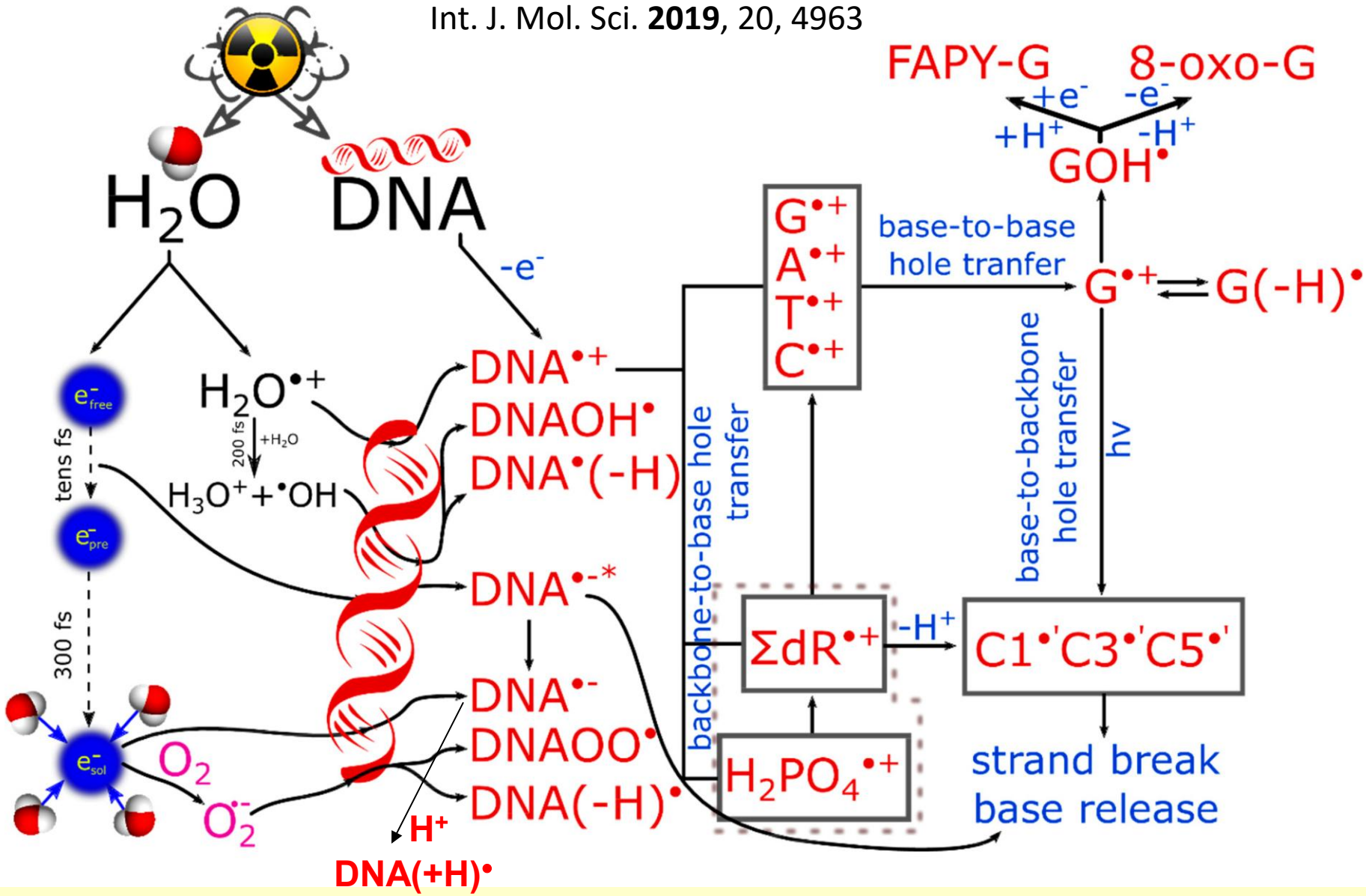
Figure 1. Ultrafast charge transfer from the water cation radical, $H_2O^{\bullet+}$, and excess electron attachment to the solute occurring in less than 1 ps in concentrated solutions; these processes do not occur in dilute solutions and reactions due to indirect effect of radiation are predominant in dilute solutions.

Int. J. Mol. Sci. **2019**, *20*, 4963

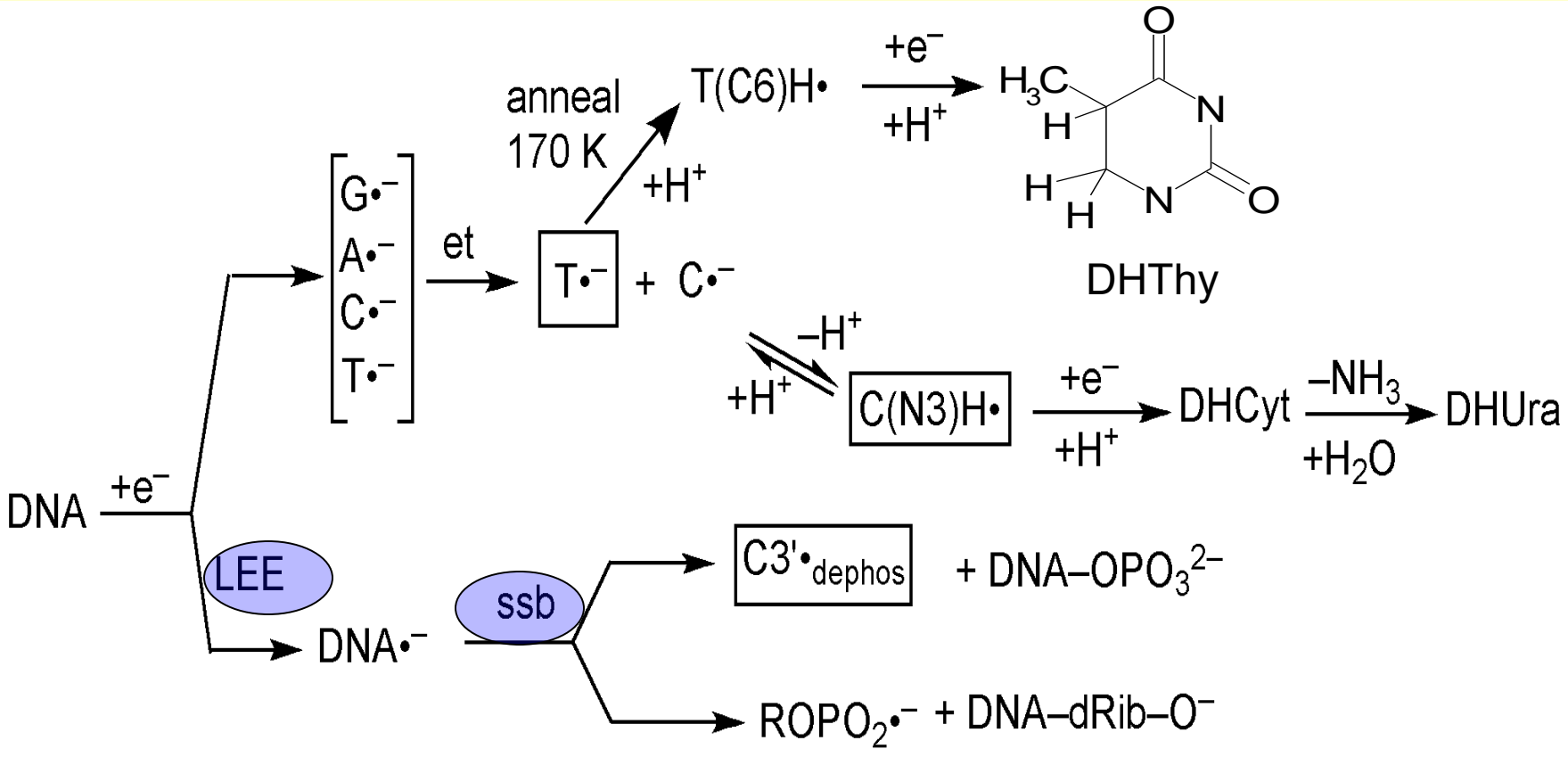


Our model includes contributions of excitations and Low energy electrons (LEE)

DNA-radicals formed via oxidative (i.e., electron-loss) pathway also the anion radical route shown



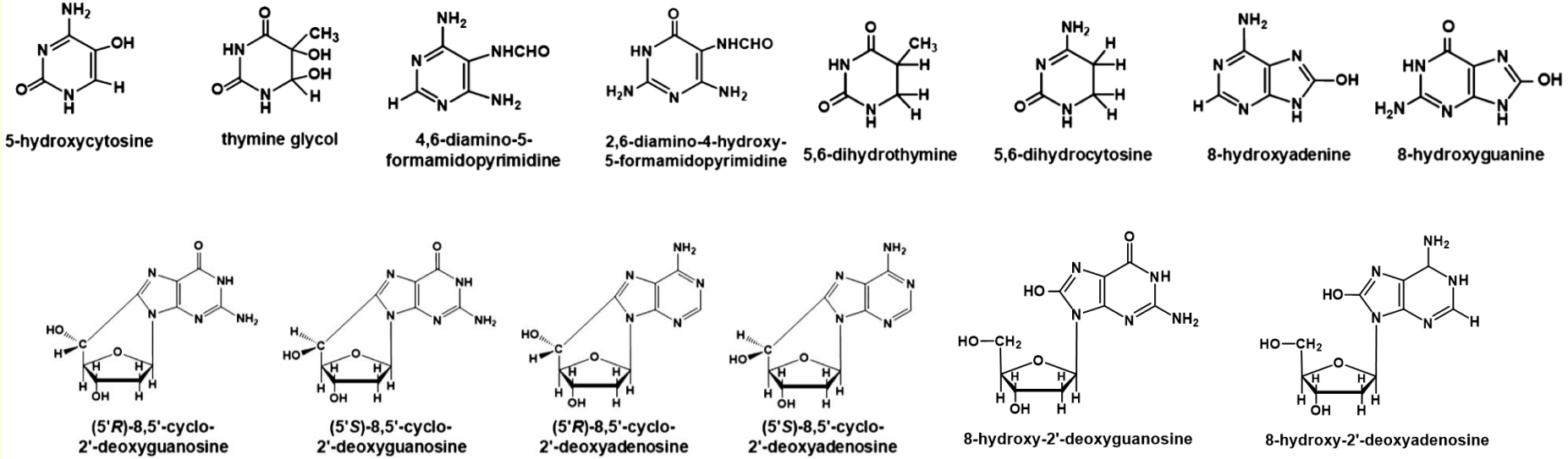
DNA-radicals formed via the reductive (i.e., electron-gain) pathway



Adhikary, A., Kumar, A., Becker, D., and Sevilla, M. D. (2012) Theory and ESR spectral studies of DNA-radicals. In **Encyclopedia of Radicals in Chemistry, Biology and Materials**. (C. Chatgilialoglu, A. Struder (Eds.)), John Wiley & Sons Ltd, Chichester, UK, pp 1371-1396.

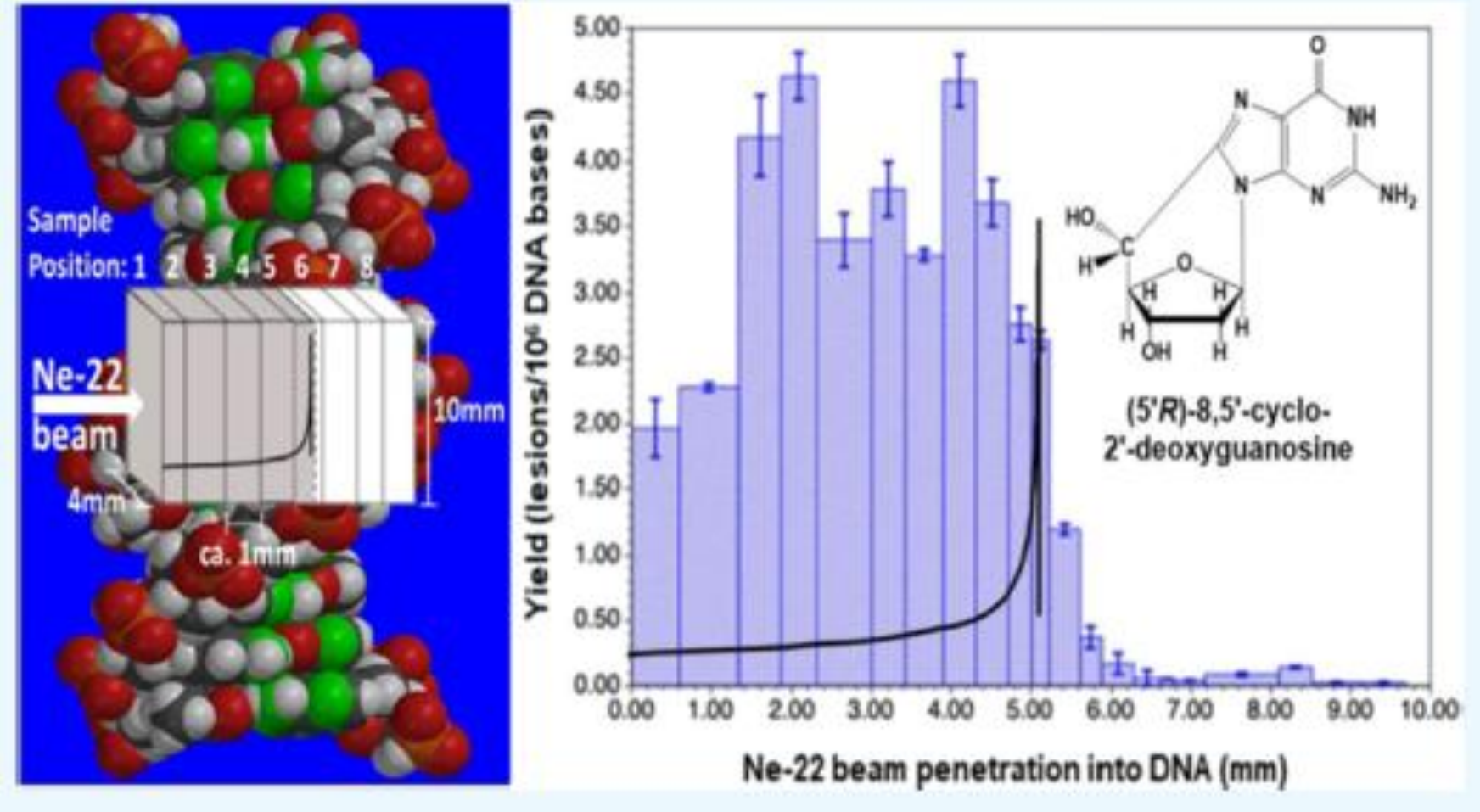
Room Temperature Base Damage Products

Final Damage Products Considered



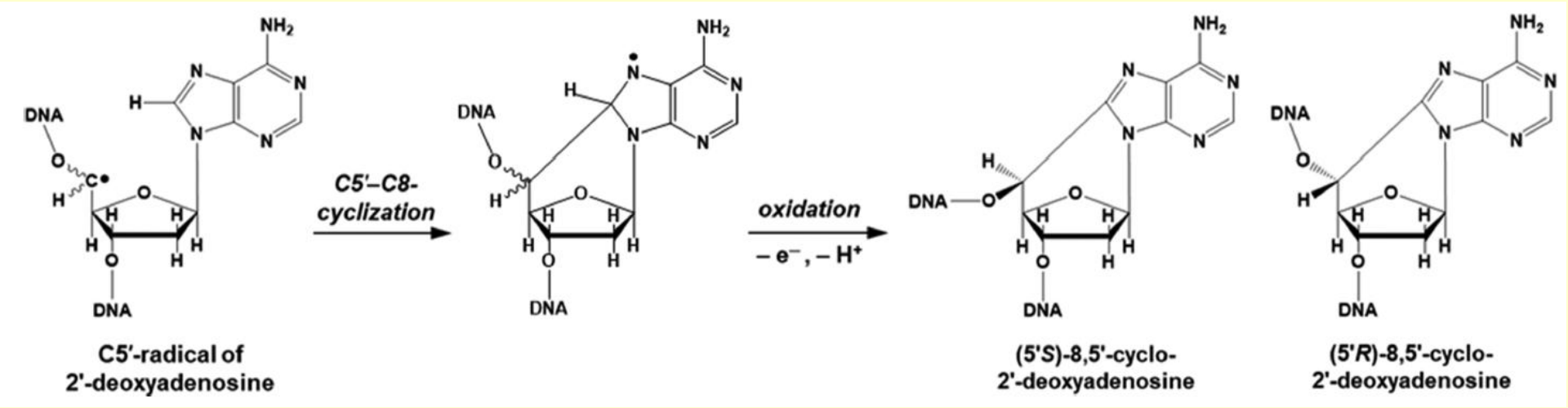
ACS Omega, 2021, 6, 16600-16611

Identification of highly mutagenic 8,5′-cyclo-2′-deoxyguanosine in ion-beam irradiated DNA



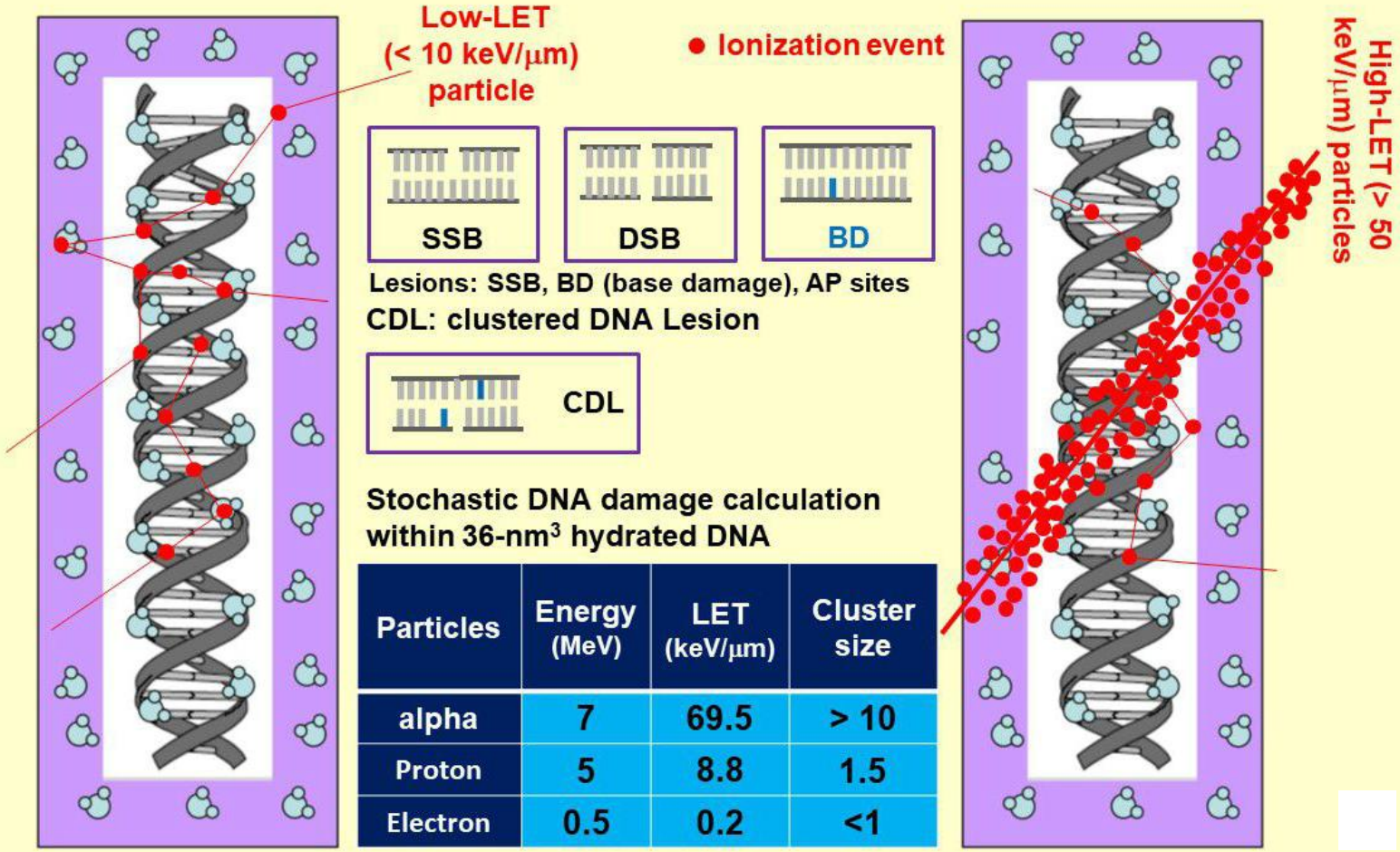
ACS Omega 2021, 6, 16600−16611

Mechanism of 8,5′-cyclo-2′-deoxyguanosine formation



ACS Omega 2021, 6, 16600–16611

Geant4-DNA calculations



Stochastic DNA damage calculation within 36-nm^3 hydrated DNA

Particles	Energy (MeV)	LET (keV/μm)	Cluster size
alpha	7	69.5	> 10
Proton	5	8.8	1.5
Electron	0.5	0.2	<1

DNA **2024**, 4, 34–51. https://doi.org/10.3390/dna4010002

ChemBioChem 2022, e202200391 doi.org/10.1002/cbic.202200391

Processes that did NOT significantly affect damage to 2686 bp plasmid DNA during repeated freeze-thaw cycles:

- mechanical stress alone

- Vortexing

- pH 6

- water or NaCl solution under tested conditions

Processes that DID affect damage:

- Fe^{2+} contamination

- ROS generation during freeze-thaw

- damage strongly suppressed by radical scavengers DMSO and Ectoine

- long-term frozen storage < −25 °C caused only slow degradation over months to years

Future Directions and CIRMS Needs

Predictive Radiobiology

- **Standardization:** Establishing a quantitative framework that links physical "track-structure" physics to chemically defined DNA-lesions.
- **Predictive Power:** Moving from simple dose measurement to **predictive radiobiology**, thereby forecasting biological risk based on the specific type and quality of radiation (Joey Robertson, also see the poster of Mohammad Rezaee and John Wang) .
- **Clinical Relevance:** Advancing radiation measurement science to improve outcomes in Radiation Oncology, Radiation and Nuclear Countermeasure Program, Space radiation protection etc.

Future: Toward Predictive Biodosimetry

- Standardize lesion yields
- Develop (LET and fluence)-specific biodosimetry
- Integrate into clinical planning

FINAL TAKE-HOME

•The mechanism based biodosimetry model predictions are **to be validated by experiments.**

•DNA-lesions are the **true measurable quantity**

•LET and track structure define **damage quality**

•This framework enables **predictive radiobiology**

By linking track-structure physics to chemically defined DNA-lesions, we transform biodosimetry from observation into quantitative prediction.

Grateful to:

- **Free Radical Research and Radiation Biology Program, The University of Iowa, and to Dr. Andrean Simons-Burnett.**
- **Holden Comprehensive Cancer Center.**
- **Veterans Affairs, The University of Iowa.**

Present Collaboration:

- **(Late) Miral Dizdaroglu (NIST)**
- **Mehran Mostafavi (U Paris-Saclay)**
- **Stanislaw F. Wnuk (FIU)**
- **Mohammad Rezaee (JHU)**
- **Sudipta Seal (UCF, Orlando)**
- **Peter Glazer (Yale)**
- **Arnab Chakravarti (OSU)**
- **Melanie Coathup (UCF)**
- **Stephen Brown (HFH)**
- **Doughlas Boreham (NSOM)**
- **Achintya Dutta (IIT, Mumbai, India)**
- **Marc Greenberg (JHU)**

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- **Indebted to (Late) Prof. C. von Sonntag**
- **(Late) Prof. Bill Bernhard**

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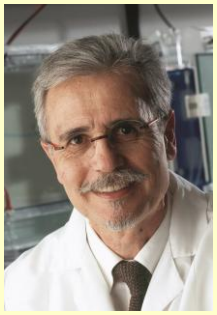
DNA



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MDPI

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Scope:

- Nucleotides; nuclear DNA; mitochondrial DNA; chloroplast DNA; mobile DNA
- DNA replication; DNA damage and repair; nucleic acid enzymes
- Radiation damage to DNA: Mechanisms and Consequences
- Gene regulation; gene expression; DNA methylation; epigenetics
- Sequencing technologies; DNA testing; genomics; bioinformatics
- DNA origami; DNA-based hybrid materials; DNA-based sensors
- Chromosome organization; genome structure and function; chromatin remodelling and dynamics; nuclear organization
- Genome evolution from sequence to structural level

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Support

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- **DNA (MDPI)**
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Thank you for your attention!

Revised model structure

Layer 1: Physical and chemical induction
- dose, LET, fluence, track structure
- direct/quasi-direct/indirect effects
- lesion spectrum per exposure

Layer 2: Measurement and persistence
- immediate clustered lesions as exposure signal (Sutherland)
- handling/storage artifacts and stability constraints (Hahn)
- residual unrepaired or unrepairable clusters as possible biological memory

Where is ‘old radiation’ in this model?

1. Not stored as prior Gy
2. Stored, if at all, in residual biological state: unrepaired clustered…
3. Sutherland explicitly suggests some clusters may be unrepairable and may…
4. Hahn reminds us that some apparent persistence can also come…

How re-irradiation should be represented

1. A fresh 50 Gy course should not be modeled as…
2. Instead, the model needs state variables carried forward through time
3. Examples: residual clustered damage burden, repairable vs non-repairable lesion pools,…
4. Time between courses changes the retained state through repair, repopulation,…