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A PHYSICOCHEMICAL ASPECT OF RADIOPROTECTION AND SENSITIZED CANCER RADIOTHERAPY

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Both protection of normal tissue cells and sensitization of tumor cells are of basic importance in cancer radiotherapy. Cellular inactivation by radiation is promoted by water and oxygen. It is well known that the well oxygenated normal tissue cells are radiosensitive and the hypoxic cells in solid tumor are radioresistant.

We have studied the correlation between the radiation damage of DNA-constituents, especially of the thymine base moiety, and radiation inactivation of biological cells. This correlation provides physicochemical understanding on the protection of normal tissue and oxygen-mimetic sensitization of tumor cells in cancer radiotherapy.

Thymine Hydroxylation and Cellular Inactivation by Radiation Apparent reactivities of DNA-related compounds in the radiolysis of deoxygenated aqueous solution increase in the following orders: adenine (G = 0.43) < cytosine (G = 1.10) < thymine (G = 1.81) for DNA bases, and thymine (G = 1.81) < thymidine

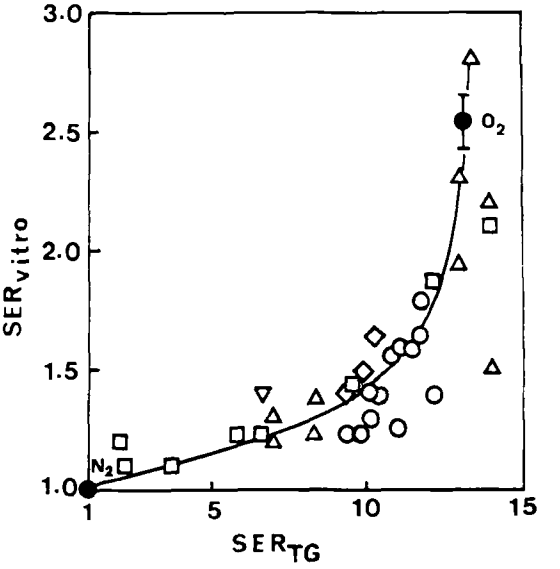


FIGURE 1 Correlation between the sensitizer enhancement ratios of V 79 cell killing (SER_{vitro} with 1.0 mM sensitizer) and TG formation ($SER_{TG} = G_S(TG)/G(TG)$ with 1.0 mM thymine and 0.2 mM sensitizer) for various nitroaromatic compounds: (Δ) nitrobenzenes, (▽) nitrothiazoles, (◇) nitrophyridines, (□) nitroimidazoles, (○) nitrotriazoles.

(G = 2.14) < thymidylic acid (G = 2.76) for thymine derivatives.[1] This result indicates that thymine and its derivatives are more radiosensitive sites in DNA. The radiolyses of deoxygenated and oxygenated aqueous solutions of thymine showed that oxygen and nitroazole radiosensitizers promote the formation of oxidation products such as thymine glycol (TG).

The lethal damage of V 79 Chinese hamster ovary cells on exposure to radiation is 2.5-times more enhanced under oxic (O_2) conditions than that under anoxic (N_2) ones. The sensitzer enhancement ratio (SER) in the *in vitro* cellular system is correlated with that of TG formation in the chemical system (see Figure 1). This result indicates that the cellular inactivation by radiation occurs more easily under the conditions giving higher TG yield.

Although the biological meaning of the correlation in Figure 1 has not yet been elucidated, the extent of radiation cell-killing *in vitro* is predictable from the G value of TG as a radiation chemical measure.

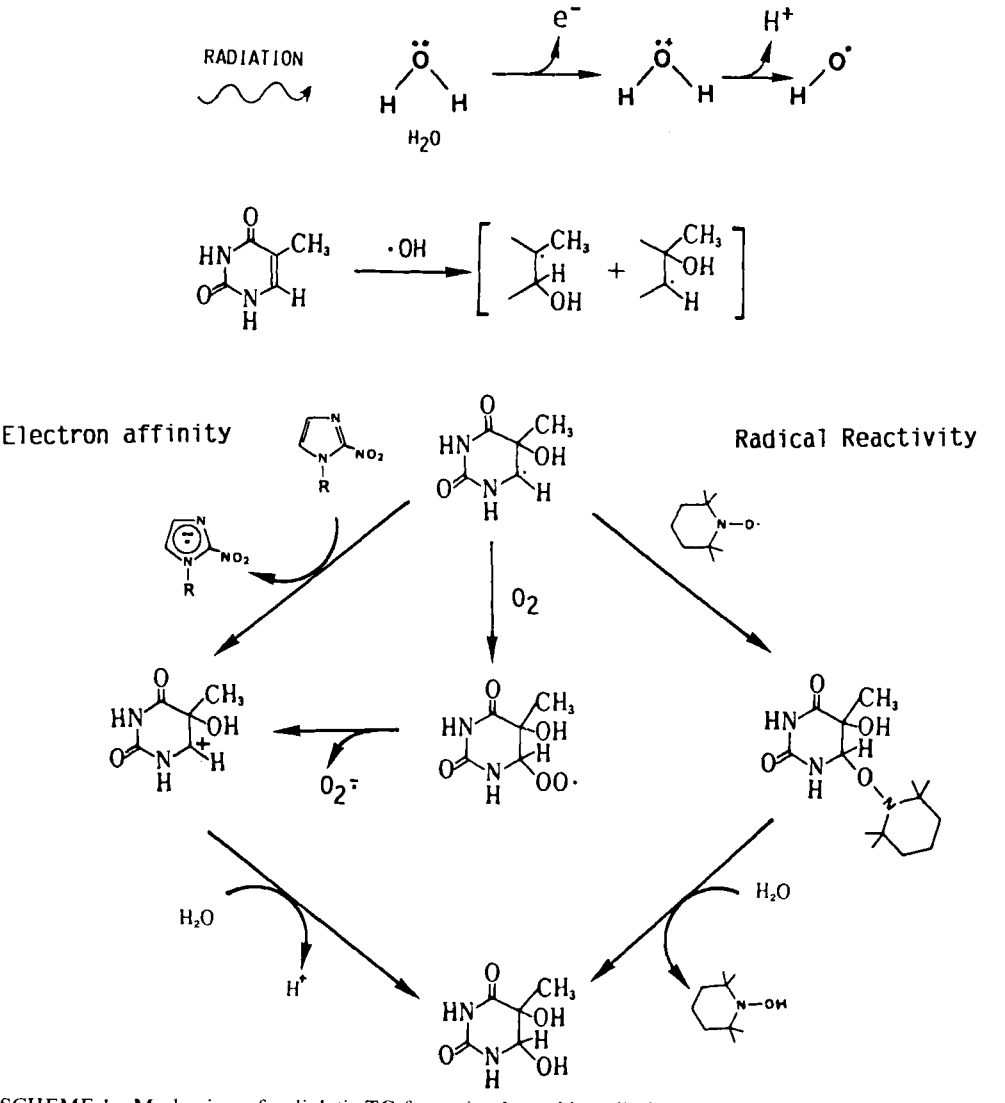
Radioprotection by Hydrogen-Atom Donating Compounds N_2O-saturation leads to 4.6-times larger G-value of TG in the radiolysis of aqueous thymine solution[2] and 2.9-times higher radiosensitivity of *Pseudomonas radiora* 0–1 in aqueous suspension,[3] because of higher yield of ·OH radicals in these systems. The TG formation in N_2O-saturated solution is retarded efficiently by the addition of various alcohols such as ethanol and ethylene glycol. The radiation lethal damages of bacterial cells (*Pseudomonas radiora* 0–1)[3] and mammalian V 79 cells[4]are also retarded by these alcohols. These results show that alcohols deactivate ·OH radicals and thereby protect intracellular DNA.

In the radiolysis of deoxygenated aqueous thymine solution, the TG formation is inhibited almost completely, whereas that of 5,6-dihydrothymine (DHT) is 2.0-times promoted, by the addition of glutathione (GSH) as one of intracellular sulfhydryl compounds.[1] Thus, GSH is an efficient hydrogen-atom donor for not only ·OH radicals but also for the 5,6-dihydrothymyl radical intermediate. Biological cells are protected by intracellular GSH. It decreases by the addition of N-ethylmaleimide (NEM) which reacts quantitatively with GSH. The radiation cell-killing is enhanced by NEM under anoxic conditions.[5]

Radiosensitization by Oxygen-Mimetic Compounds Two types of oxygen-mimetic stable radicals and electron affinic compounds have been studied for radiosensitization of hypoxic tumor cells.

In the radiolysis of N_2-saturated aqueous solution, stable radical compounds such as 2,2,6,6-tetramethyl-piperidine-N-oxyl(TEMPO·) promoted markedly (8.5–12.5 times) the hydroxylation of thymine to TG with almost complete depression of side reactions.[6] This sensitized TG formation proceeds by a hydrolysis mechanism (see Scheme 1). The N-oxyl derivatives sensitize the radiation cell-killing *in vitro* under anoxic conditions.[7]

Nitroaromatic compounds such as nitroazole derivatives having large one-electron reduction potentials also promoted the radiolytic TG formation under deoxygenated conditions.[2] This sensitzed TG formation proceeds by a one-electron oxidation mechanism (see Scheme 1). These electron-affinic compounds enhance the radiation inactivation of V 79 cells under anoxic conditions. The SER of cell killing in the *in vitro* system increases with increasing the SER of TG formation in the chemical system (see Figure 1).



SCHEME 1 Mechanism of radiolytic TG formation by stable radical and electron affinic compounds.

Radiosensitization of Solid Tumor Cells For clinical use, radiosensitizers must have higher tumor affinity and lower brain affinity *in vivo* with higher sensitizing activity *in vitro* cellular systems.

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