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Gadolinium-based nanoparticles as sensitizing agents to carbon ions in head and neck tumor cells

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Abstract

Hadrontherapy presents the major advantage of improving tumor sterilization while sparing surrounding healthy tissues because of the particular ballistic (Bragg peak) of carbon ions. However, its efficacy is still limited in the most resistant cancers, such as grade III-IV head and neck squamous cell carcinoma (HNSCC), in which the association of carbon ions with gadolinium-based nanoparticles (AGuIX®) could be used as a Trojan horse. We report for the first time the radioenhancing effect of AGuIX® when combined with carbon ion irradiation in human tumor cells. An increase in relative biological effectiveness (1.7) in three HNSCC cell lines (SQ20B, FaDu, and Cal33) was associated with a significant reduction in the radiation dose needed for killing cells. Radiosensitization goes through a higher number of unrepaired DNA double-strand breaks. These results underline the strong potential of AGuIX® in sensitizing aggressive tumors to hadrontherapy and, therefore, improving local control while lowering acute/late toxicity.

Key words: Gadolinium nanoparticles; Radiosensitization; Residual double strand breaks; Head and neck squamous cell carcinoma (HNSCC); Carbon ion irradiation

Nanoparticles (NPs) containing high-Z elements are promising candidates for enhancing radiotherapy efficiency. The interaction of radiation with metals produces secondary particles (photoelectrons, Auger, Compton electrons, etc.),[1,2] depending on the beam energy, which leads to the neighboring generation of reactive oxygen species and, subsequently, to a local dose enhancement.[3] After the pioneering work of Hainfeld[4] using gold NPs (GNPs), a large number of reports confirmed the advantage of metallic NPs in association with radiotherapy to overcome tumor radioresistance.[5,6] Gadolinium-based nanoparticles (GBNs) proved to be efficient radiosensitizers in different *in cellulo* and *in vivo* tumor models.[7] This effect should be theoretically observed at the K-edge of gadolinium (around 50 keV); however, surprisingly, we[8] and others[7,9,10] observed an enhancement of photon effects in different cellular models at energies between 220 kV and 6 MV (clinical energy). In particular, we demonstrated the radiosensitizing effect of a first generation of GBNs (DTPA as chelator) combined with 250 kV photon radiation in radioresistant cellular and animal models of head and neck squamous cell carcinoma (HNSCC).[8] One of the major criticisms of GBNs is their lack of specific targeting, despite tumor enrichment by the enhanced permeability and retention (EPR) effect, and the potential risk of toxicity in healthy tissues when combined with conventional radiotherapy.

Hadrontherapy with protons or carbon ions ($^{13}C^{+6}$) has been demonstrated to target tumors based on its high-energy delivery

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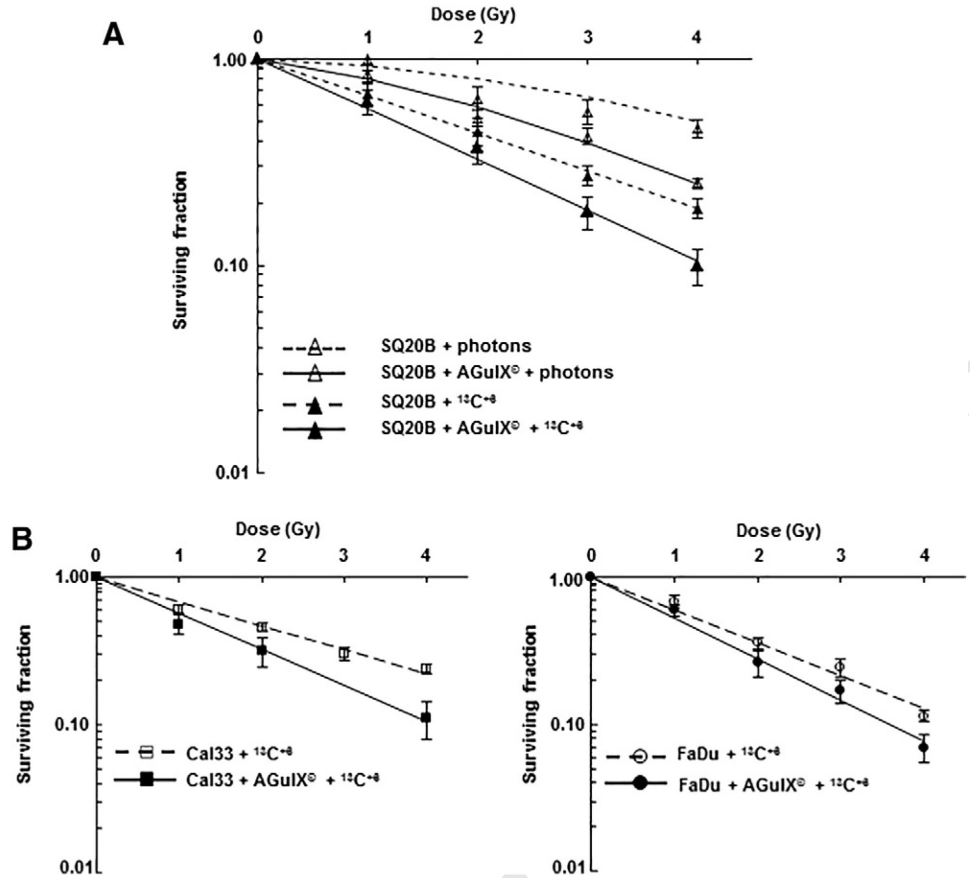

Figure 1. Radiosensitizing effect of AGuIX® in SQ20B (**A**), Cal33 and FaDu cells (**B**) irradiated with $^{13}C^{+6}$ or photons.

Table 1
Radiobiological parameters for HNSCC cell lines irradiated with photons or carbon ions in the presence or not of 0.8 mg/ml AGuIX®.

		$\alpha(Gy^{-1})$	$\beta(Gy^{-2})$	SF2	D_{10}(Gy)	RBE
Photons	SQ20B	0.07	0.03	0.74	7	
	SQ20B + AGuIX®	0.19	0.04	0.58	5.1	1.37
$^{13}C^{+6}$	SQ20B	0.42		0.43	5.5	1.27
	SQ20B + AGuIX®	0.56		0.33	4.1	1.7
	Cal33	0.38		0.47	6.1	1.14
	Cal33 + AGuIX®	0.56		0.32	4.1	1.7
	FaDu	0.51		0.36	4.5	1.33
	FaDu + AGuIX®	0.64		0.28	3.6	1.66

SF2, survival fraction at 2Gy; D_{10}, dose of radiation corresponding to 10% of survival; RBE, Relative Biological Effectiveness at 10% survival.

(Bragg peak) at the end of the course.[11] This specific characteristic affords a limited energy deposition in surrounding healthy tissues as well as a massive transfer of energy within the tumor. Furthermore, $^{13}C^{+6}$ exhibits a higher relative biological effectiveness (RBE) compared with photons because a higher local dose is delivered along the particle tracks, leading to complex, unrepairable DNA damage and cell death.[12–14]

Thus, combining hadrontherapy with GBNs may be of particular interest for amplifying the local energy deposition in radioresistant tumors, such as grade III-IV HNSCC (35% survival at 5 years), which relapse even after carbontherapy.[15]

Table 2
Isobolographic analyses of radiation and AGuIX® in three HNSCC cell lines.

	Photon irradiation	D (Gy)	Isobolographic Analyses	
			50% survival	10% survival
SQ20B		1	Syn	Syn
		2	+	+
		3	+	Syn
		4	Syn	Syn
	$^{13}C^{+6}$ irradiation			
SQ20B		1	+	Syn
		2	Syn	Syn
		3	Syn	Syn
		4	Syn	Syn
Cal33				
		1	+	+
		2	+	Syn
		3	NA	NA
		4	Syn	Syn
FaDu				
		1	+	+
		2	+	+
		3	+	+
		4	Syn	Syn

Syn: synergistic effect; Ant: antagonistic effect; +: additive effect.