Characteristics of Cell Damage in Breast Cancer with the Use of Modifiers of Radiotherapy

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Ultrastructural peculiarities of breast tumor cells damaged by radiotherapy modulated by thermomodifiers are studied. The destructive changes are characteristically more pronounced in the center of nodes rather than in their peripheral parts after hyperthermia of superhigh frequency. Alterations induced by high-frequency interstitial hyperthermia are distributed more evenly in the central and peripheral regions. Ferromagnetic hyperthermia activates phagolysosomes containig iron particles, while hyperthermia with current-conducting fluids causes intra- and intercellular edema.

Key Words: *breast carcinoma; ultrastructure; radiotherapy modifiers*

Radiation and thermoradiation pathomorphosis are compared mainly by histological manifestations in studies of the effectiveness of local hyperthemia as a modifier of tumor radiation therapy [1-3]. Electron microscopic (EM) analysis has as a rule not been performed or has been of a descriptive nature [3-5,7].

The aim of the present investigation was to determine the quantitative and qualitative EM characteristics of cell damage in breast carcinomas in local remote SHF-, interstitial ferromagnetic high-frequency (HF-), and HF-hyperthermia with current-conducting fluids.

MATERIALS AND METHODS

Surgical material from patients with infiltrating ductal carcinomas at clinical stages II-III was studied. Patients were treated surgically (11 persons), by radiation (13 persons), and by thermoradiation: 17 patients received local SHF-hyperthermia after every other radiation session (3rd group), 15 patients after the end of radiation treatment underwent interstitial ferromagnetic hyperthermia with the administration of reduced iron particles on gelatinol around the tumor bed (4th group), and 6 patients (5th group) after radiation were subjected to interstitial hyperthermia with the administration of current-conducting fluids instead of reduced iron. The procedures were performed no later than 2 days after the end of the course of treatment.

Samples from the central and peripheral regions of carcinoma nodes were prepared according to a described method [8] and viewed in an EMV-100 AK electron microscope. The surface area of outer membranes (S_O) and cristae (S_C) of mitochondria and endoplasmic reticulum (ER) (S_{ER}) [2] was measured using photonegatives, and the volume of mitochondria in a unit of cell volume (V_M) and of ribosomes bound with granular ER (S_R) was determined. The results were processed statistically using parametric and nonparametric tests [6].

RESULTS

The phenomena of radiation pathomorphosis (2nd group) manifested themselves in discomplexation of the tumor parenchyma, destruction of organelles, disorganization of the outer and inner membranes and lightening of the mitochondrial matrix, widen-

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TABLE 1. Morphometric Indexes of Cancer Cell Ultrastructure in the Groups under Observation (*M*±*m*)

Group	V_M	S_O	S_C	S_{ER}	S_R
1st (*n*=11)	8.1±0.9	12.1±0.9	5.2±0.7	13.4±1.2	7.7±0.4
2nd (*n*=13)	5.8±0.7	9.0±0.9*	2.6±0.3	5.3±1.7	4.0±0.7
3rd (*n*=17)	7.4±1.0^{*+}	11.0±1.0^{*+}	1.5±0.3	2.6±0.3	1.4±0.2
4th (n=15)	5.0±0.5^{+}	12.3±1.1^{*+}	0.9±0.1	5.6±1.0^{+}	1.5±0.2
5th (*n*=6)	5.3±0.8^{+}	9.0±0.8^{*+}	1.1±0.2	7.0±2.0^{+}	1.4±0.2

Note. *The differences are not reliable as compared to the 1st group of observations, $^{+}$as compared to the 2nd group.

ing of nuclear pores, and marginal condensation of chromatin (Fig. 1). A decrease of S_C, S_{ER}, and S_R was determined morphometrically (Table 1). The correlation (r=0.72, p<0.05) found between the values of S_{ER} and S_R points to the connection between the changes of membranes and ribosome degranulation.

Marked destructive changes were noted in the center of tumors after SHF-hyperthermia. Hydropic processes dominated in these changes: amorphous masses of cell detritus contained many vacuoles and lysed nuclei; swollen mitochondria with damaged cristae and empty lightened matrix were the only organelles found. Tumor elements with deep invaginations of the karyolemma and margination of lumpy chromatin were occasionally found on the periphery. Lysosomes and lipid inclusions were found in the protoplasm, and the swollen ER membranes were vacuolized and had areas of breaks and lysis. The majority of ribosomes had lost their orientation (Fig. 2). S_C, like S_{ER} and S_R, was significantly (p< 0.05) decreased as compared both to radiation and to surgical treatment (Table 1). This may indicate that thermoradiation therapy results in more pronounced inhibition of energy and protein-synthesizing processes in tumor cells than does gamma radiation.

Alterative processes were distributed uniformly in the peripheral and central parts of the tumor when a session of interstitial ferromagnetic hyperthermia was added to the radiation therapy, evidently due to a more even thermal action. An abundance of small and large phagolysosomes, some filled with carbonyl iron particles, some with destroyed membranes, was a distinctive feature. When these organelles were located in the perinuclear space, they caused partial lysis of the nuclear membrane. The greatly swollen ER had vacuolar inclusions and the mitochondria were characterized by double-contoured outer membranes, destroyed cristae and empty matrix, and nuclear edema (Fig. 3). V_M и S_C were lowered as compared to both surgical and SHF-thermoradiation treatment (Table 1). Ultrastructural transformations of tumor cells after interstitial HF-hyperthermia with current-conducting fluids was included in the therapeutic protocol were much the same as changes after ferromagnetic hyperthermia. Intra- and pericellular edema and intracytoplasmic electron-dense granules were characteristic (Fig. 4). S_C и S_R were decreased as compared to surgical and radiation treatment. As in interstitial ferromagnetic hyperthermia, the mean value of S_{ER} was significantly higher for heating with current-conducting fluids as compared to SHF-heating (Table 1).

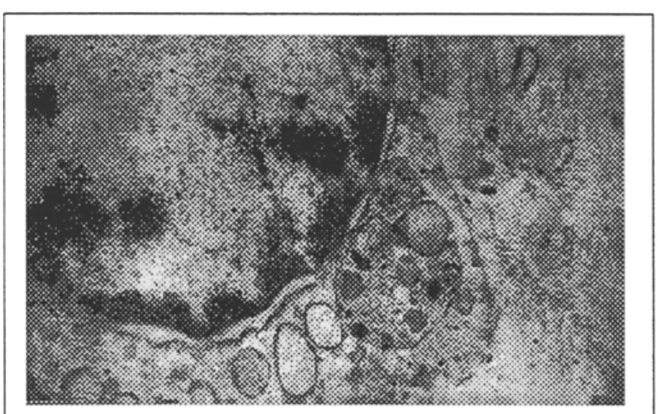

Fig. 1. Lightening of mitochondrial matrix, widening of nuclear pores and perinuclear space, and marginal condensation of chromatin. Electronogram, × 20,000.

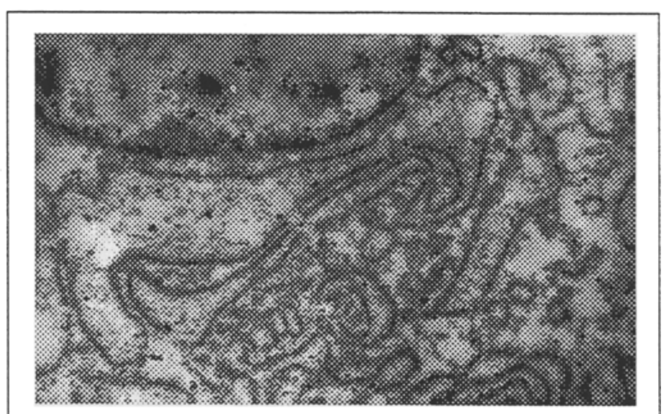

Fig. 2. Vacuolization, breaks, and lysis of endoplasmic reticulum with loss of ribosome orientation. Electronogram, × 20,000.

Fig. 3. Double—contoured outer membranes, destruction of cristae, and empty matrix of mitochondria; edema and invagination of nuclei. Electronogram, ×20,000.

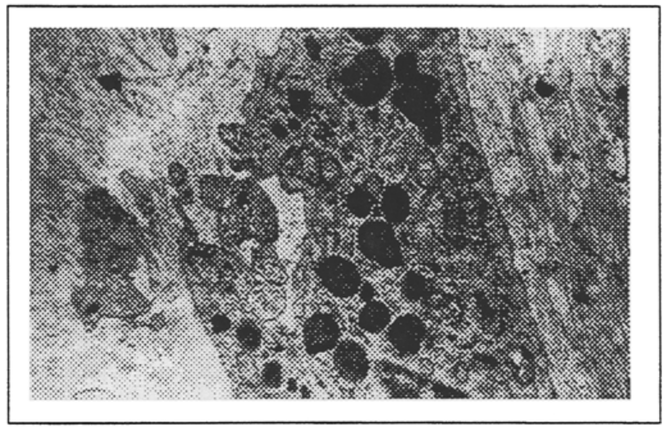

Fig. 4. Intra— and pericellular edema, and cytoplasmic electron—dense granules.
Electronogram, ×20,000.

Therefore, the damaging effect of thermomodifiers of radiation therapy on breast carcinomas is manifested on energy-forming and protein-synthesizing ultrastructures. Both SHF- and ferromagnetic HF-therapy cause a marked degranulation of the reticulum. In thermoradiated groups correlations of the same direction and force (r= 0.6, p<0.02) were found between S_{ER} and S_R. But there are differences in S_{ER} in the two thermoradiation methods. For example, the S_{ER} values after ferromagnetic hyperthermia are higher (p< 0.05) than in SHF radiation, perhaps due to the hyperplasia of ER membranes after interstitial hyperthermia and activation of hydropic processes with membrane vacuolization.

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