Evaluation of Antitumor Efficiency of Experimental Interstitial Photodynamic Therapy on the Model of M1 Sarcoma

O. A. Skugareva, M. A. Kaplan, A. I. Malygina, and A. A. Mikhailovskaya

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny,* Vol. 148, No. 11, pp. 561-563, November, 2009
Original article submitted April 8, 2009

Antitumor efficiency of interstitial photodynamic therapy was evaluated in experiments on outbred albino rats with implanted M-1 sarcoma. Interstitial photodynamic therapy was carried out using one diffusor at different output power and duration of exposure. The percentage of complete regression of the tumors increased with increasing exposure parameters.

Key Words: *interstitial photodynamic therapy; M-1 sarcoma*

Photodynamic therapy is a method for local treatment of the tumor involving the minimum injury to normal tissues. The photodynamic effect is realized at the expense of reaction of the photosensitizer (PS), selectively accumulating in tumor cells, and the light at a wavelength corresponding to the PS absorption spectrum. Photochemical reactions cause the formation of singlet oxygen and other biological oxidants with a direct cytotoxic effect promoting destruction of the tumor vascular network with subsequent activation of the immune response [3,5,6,8]. The efficiency of photodynamic therapy is determined by the PS characteristics (selective accumulation and photochemical activity), penetrating capacity, dose and power of laser, vascularization and oxygenation of tumor tissue [1,4,7]. One of the main factors limiting the use of photodynamic therapy in the treatment of cancer is the depth of photo radiation penetration. For red laser (661-670 nm) it does not exceed 0.8-0.9 cm [5]. Therefore, the search for methods of interstitial photodynamic therapy (IPDT) with greater volume and depth of tumor exposure is an important task [9].

Medical Radiology Research Center, Russian Academy of Medical Sciences, Obninsk, Russia. ***Address for correspondence:*** skugaro99@mail.ru. O. A. Skugareva

We evaluated the antitumor efficiency of IPDT under experimental conditions on the model of M-1 sarcoma.

MATERIALS AND METHODS

The study was carried out on 100 outbred female albino mice with subcutaneously implanted (into the hip) M-1 sarcoma. The animals were exposed after palpatory detection of the tumor node on days 10-12. The mean volume of the tumor was 0.25 cm^3. Before the procedure, the animals were narcotized with 0.25% sodium thiopental (0.1 ml/100 g). Photolone (chlorine PS) was injected intraperitoneally (5 mg/kg; Latus 0.4 semiconductor laser with λ=662 nm served as the source of radiation. A lightguide with a cylindrical diffusor (1-cm active part) was used. A needle with a mandrel was injected into the thickness of the tumor; the mandrel was then removed and the lightguide was inserted through the needle canal. The diffusor was located along the greatest size of the tumor. The lightguide position was verified by the light spot.

The tumor diameter was measured before agent injection (initial data) and on days 3, 7, 10, 14, and 21 after therapy [2]. The dynamics of tumor growth was evaluated by the coefficient of the absolute tumor in-

crement (C) [2]. The treatment efficiency was evaluated by the continuing growth coefficient and complete regression (CR) percentage in comparison with the control group. The absence of apparent palpated tumor was taken as CR (C=-1.00). The control group consisted of animals receiving no treatment of any kind (no PS and no irradiation). The results were processed by variation statistics methods, the significance of differences was evaluated using Mann–Whitney test. Experimental data were compared with the data in the control for each experiment.

RESULTS

The animals were divided at random into 8 groups (Table 1). Intense growth of M-1 sarcoma was observed in the control group. In none controls spontaneous regression of the tumor was observed. Directly after

TABLE 1. Coefficient of Absolute Increment of M-1 Sarcoma after IPDT

Parameter	Day of observation				
	3	7	10	14	21
Control (*n*=17)					
C	1.73±0.32	7.53±1.39	17.70±3.35	29.85±5.58	69.83±13.90
Power 100 mW, exposure 20 min (*n*=11)					
C	0	0	0.53±0.53	0.84±0.41	9.13±3.29
			p=0.008	*p*=0.001	*p*=0.002
CR, %	91	91	73	45.5	45.5
Power 100 mW, exposure 25 min (*n*=11)					
C	1.95	4.31	7.44	3.68±2.76	9.77±1.64
				p=0.03	*p*=0.03
CR, %	91	91	91	82	82
Power 200 mW, exposure 15 min (*n*=13)					
C	-1.0	-1.0	-1.0	2.90±1.98	5.32±2.92
				p=0.03	*p*=0.004
CR, %	100	100	100	67	54
Power 200 mW, exposure 20 min (*n*=14)					
C	-1.0	0.11	1.07	4.93	7.92±3.87
					p=0.04
CR, %	100	93	93	93	79
Power 200 mW, exposure 25 min (*n*=11)					
C	-1.0	-1.0	-1.0	-1.0	0
CR, %	100	100	100	100	91
Power 300 mW, exposure 10 min (*n*=12)					
C	-1.0	0	0.16±0.16	0.55±0.16	8.15±5.05
			p=0.06	*p*=0.002	*p*=0.006
CR, %	100	92	83.3	58.3	58.3
Power 300 mW, exposure 15 min (*n*=11)					
C	-1.0	-1.0	0	6.02	3.95±3.95
					p=0.03
CR, %	100	100	91	91	82

Note. C: coefficient of absolute tumor increment. Significant differences from the control according to Mann–Whitney test.

the IPDT session, all experimental animals developed cyanosis of the skin, which augmented with time. After 3-4 h the tumor was poorly discernible because of edema. The crust formed by days 3-5, its size varying from 0.7-1.0 to 1.5-1.8 cm. Edema of the adjacent tissues persisted until days 7-10.

In groups exposed to radiation of 100 mW, the tumor resolved under the crust and by day 21 was gradually replaced with the connective tissue; a fine cicatrix formed at the site of exposure. In groups exposed to 200 and 300 mW, the adjacent normal tissues were damaged, and by the end of the observation period ulcerative defects or a coarse cicatrix formed at the site of exposure, so that the animals spared the limb. The most pronounced changes were observed after 25-min exposure to radiation of 200 mW and to 10-min or 15-min exposure to 300 mW.

Analysis of experimental data demonstrated the increase of tumor CR percentage after exposure to higher output power. The increase of the power from 100 to 200 mW (20-min exposure) resulted in an increase of CR from 45.5 to 79%, the coefficient of absolute tumor increment in animals with continuing growth being 9.12 and 7.92, respectively. At a power of 200 and 300 mW (15-min exposure) the CR was 54 and 82%, respectively, the tumor increment coefficient being 5.32 and 3.95, respectively.

The most significant parameter was the duration of exposure. At the output power of 100 mW prolongation of exposure from 20 to 25 min led to CR increase from 46 to 82%. In the groups exposed to 200 mW the percentage of animals with tumor CR also increased with prolongation of exposure: 54% after 15 min, 79% after 20 min, and 91% after 25 min. At a power of 300 mW and exposure of 10 and 15 min the CR by the end of observation was 58.3 and 82%, respectively.

Hence, the use of IPDT for the treatment of M-1 sarcoma is sufficiently effective, but it is noteworthy that the optimal mode is low output power and long exposure of the tumor, because the increase in exposure parameters leads to enlargement of the damaged zone and hence, normal tissues are involved, which inhibits complete healing.

REFERENCES

1. D. V. Vasilyev, A. N. Stukov, and M. L. Gel'fond, *Ros. Bioter. Zh.*, No. 4, 61-66 (2003).
2. A. A. Mikhailovskaya, M. A. Kaplan, R. A. Brodskii, and L. N. Bandurko, *Byull. Eksp. Biol. Med.*, **147**, No. 1, 95-98 (2009).
3. E. F. Stranadko, *Ros. Onkol. Zh.*, No. 4, 52-56 (2000).
4. R. P. Allison, G. H. Downie, R. Cuenca, *et al.*, *Photodiag. Photodyn. Ther.*, **1**, No. 1, 27-42 (2004).
5. T. J. Dougherty, C. J. Gomer, B. W. Henderson, *et al.*, *J. Natl. Cancer Inst.*, **90**, No. 12, 889-905 (1998).
6. V. H. Fingar, T. J. Wieman, S. A. Wiehle, and P. B. Cerrito, *Cancer Res.*, **52**, No. 18, 4914-4921 (1992).
7. S. L. Gibson, K. R. VanDerMeid, R. S. Murant, *et al.*, *Cancer Res.*, **50**, No. 22, 7236-7241 (1990).
8. M. Korbelick and I. Cecica, *J. Photochem. Photobiol. B.*, **93**, No. 1, 53-59 (2008).
9. Z. Xiao, S. Halls, D. Dickey, *et al.*, *Clin. Cancer Res.*, **13**, No. 24, 7496-7505 (2007).