Evaluation of Antitumor Efficiency of Electrochemical Lysis on the Model of M-1 Sarcoma

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Antitumor efficiency of electrochemical lysis was evaluated on the model of M-1 sarcoma. At stage 1 of the study, the results of therapy with electrodes in different position were compared, at stage 2 various combination of electrochemical lysis parameters (current strength and duration of exposure) were evaluated. The increase in parameters was associated with the increase in the percentage of cases with complete regression of tumors, which was confirmed by morphological data.

Key Words: *electrochemical lysis; M-1 sarcoma*

Methods of local little invasive exposure of focal malignant tumors now attract special attention [7, 8]. A demonstrative example of this trend in oncology is electrochemical lysis (ECL) [2,3,9]. The ECL consists in exposure of the tumor node to direct current, which leads to the development of aseptic necrosis (stage I), and delayed chemical exposure of the tumor to electrolysis products (alkali, acid, and platinum compounds, stage II) [1,6]. Interstitial fluid and blood plasma are characterized by high electric conductivity. Hydrolysis (degradation of water in the fluids) leads to the formation of acidity on the anode (positively charged electrode) and alkalinity on the cathode (negatively charged electrode). Acid environment with HCl and gaseous Cl_2 and O_2 is forming in the anode zone, and alkaline environment with NaOH and H_2 is forming in the cathode zone. The portion of chlorine during the exposure is negligible, because its large molecules carrying no charge excess cannot compete with the destructive effect of H^+ ions. These protons easily migrate in the anode field more rapidly than other ions [4,10]. Exposure of the parenchymatous organs and tissues to acid causes coagulation necrosis of compact structure. Alkali causes colliquation necrosis with a liquid consistency [5].

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

MATERIALS AND METHODS

The study was carried out on 154 outbred female rats with M-1 sarcoma subcutaneously transplanted into the hip. After detection of the tumor node by palpation on days 10-11, the animals were used in the experiment. The fur in the tumor growth zone was removed by depilation cream. All animals were narcotized for the procedure with sodium thiopental (0.25%) in a dose of 0.1 ml/100 g.

Electrochemical lysis was carried out on an ECU device (Soring). The electrodes were inserted into the tumor vertically or horizontally under the tumor base at a distance of 10 mm from each other.

Tumor diameters were measured before therapy (initial data) and on days 3, 7, 10, 14, and 21 after treatment. Tumor volume was calculated by the formula:

$$V={}^{1}/_{6}\pi\times d_1\times d_2\times d_3,$$

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where: $d_{1,2,3}$ are three perpendicular diameters of the tumor, $1/6\pi=0.52$ is a constant, and V is tumor volume (cm^3).

The dynamics of tumor growth was evaluated by the coefficient of tumor absolute increment (K):

$$K=\frac{V_1-V_0}{V_0},$$

where V_0 is the initial tumor volume and V_1 is tumor volume for a certain period of observation.

Treatment efficiency was evaluated by the continuing growth coefficient and the tumor complete regression percentage (CR%) in comparison with the control group. The absence of apparent and palpated tumor (K=-1.00) was considered as complete regression.

The results were processed by method of variation statistics. The significance of differences was evaluated by Mann—Whitney test.

RESULTS

Directly after ECL, the animals were inert, did not use the affected limb during walking. The rat status normalized on day 3, the animals actively ate and moved in the cage. Analysis of treatment efficiency showed the following results (Table 1). Horizontal insertion of electrode proved to be the most effective for complete regression and for the remaining tumor increment coefficient. Tumor growth was observed in just 8% of experimental rats of this group.

Various combinations of current strength (10-30 mA) and duration of exposure (10 and 15 min; Table 2) were tried in order to choose the optimal conditions. The percentage of complete regression of the tumor node increased with increasing exposure parameters. However, the coefficients of continuing tumor growth on day 21 of observation were virtually the same in different groups, which can be explained by active multiplication of tumor cells in case of their incomplete destruction. The initial parameters of treatment were inessential for this.

Fig. 1. A fragment of the rat hip: day 21 after ECL (20 mA, 10 min), ×100. *1*) necrotic focus surrounded by leukocytic roll; *2*) remaining part of tumor tissue.

Macroscopic examination showed necrotic skin. Autopsy showed gas bubbles under the capsule and dark-red tumor tissue.

Morphological study confirmed that ECL exposure of experimental tumors caused their necrosis, its severity depending on the exposure mode (Fig. 1). The destructive effect was realized by destruction of not only the tumor parenchyma, but also its vascular network (most developed in the subcutaneous fat). This exposure of the vessels predominated in the zone of the cathode. The most pronounced destruction was seen after exposure to 30

TABLE 1. Coefficient of M-1 Sarcoma Absolute Increment after ECL (50 mA, 15 min, 45 C)

Parameter	Day of observation				
	3	7	10	14	21
Control (*n*=37)	1.68±0.26	4.11±0.49	8.39±1.15	12.23±1.52	22.62±2.53
Horizontally inserted electrodes (*n*=16)	0.85±0.19	0.85±0.23	1.80±0.59	3.09±0.72	2.82
	(*p*=0.12)	(*p*<0.001)	(*p*=0.002)	(*p*=0.002)	
CR%	—	—	—	—	92
Vertically inserted electrodes (*n*=19)	0.46±0.24	0.60±0.46	2.24±0.91	3.78±2.42	4.62±3.36
	(*p*=0.01)	(*p*<0.001)	(*p*=0.001)	(*p*=0.01)	(*p*=0.004)
CR%	—	—	—	—	58

Note. Here and in Table 2: *p*: significant difference *vs.* control.

TABLE 2. Coefficient of M-1 Sarcoma Absolute Increment after ECL (*M*±*m*)

ECL protocol	Day of observation				
	3	7	10	14	21
Control (*n*=50)	3.31±0.30	12.26±1.24	28.10±3.30	52.05±5.88	107.22±14.55
10 mA, 10 min, 6 C (*n*=12)	0.49±0.22	2.05±0.36	4.99±1.33	10.13±2.69	16.81±4.52
	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)
CR%	—	—	—	—	0
10 mA, 15 min, 9 C (*n*=9)	0.51±0.15	2.53±1.01	6.39±1.55	13.42±3.00	21.15±5.02
	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)	(*p*=0.01)
CR%	—	—	—	—	17
20 mA, 10 min, 12 C (*n*=21)	0.61±0.18	2.23±0.40	3.96±0.71	8.60±1.40	17.59±3.54
	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)
CR%	—	—	—	—	35
20 mA, 15 min, 18 C (*n*=8)	0.62±0.24	1.91±0.62	2.93±0.97	9.43±2.48	18.10±9.18
	(*p*=0.02)	(*p*=0.01)	(*p*<0.001)	(p=0.01)	(*p*=0.02)
CR%	—	—	—	—	40
30 mA, 10 min, 18 C (*n*=14)	0.57±0.19	2.70±0.75	4.17±1.27	8.65±2.62	21.23±6.25
	(*p*=0.01)	(*p*<0.001)	(*p*<0.001)	(*p*<0.001)	(*p*=0.01)
CR%	—	—	—	—	58
30 mA, 15 min, 18 C (*n*=9)	0.14	0.88±0.88	2.90±2.19	6.47±4.27	13.40±1.48
	(*p*=0.02)	(*p*=0.03)	(*p*=0.03)	(*p*=0.02)	
CR%	—	—	—	—	75

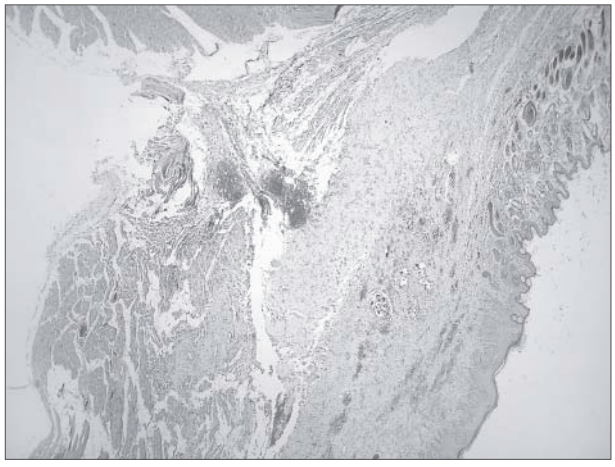

Fig. 2. Rat hip fragment: day 21 after ECL (30 mA, 15 min), ×100. *1*) thickened skin with edematous subcutaneous fat with microhemorrhages; *2*) fragments of muscle tissue with foci of myolysis under the skin.

mA current for 15 min. However, in this case tumor tissue death was paralleled by the development of necrotic changes in the adjacent tissues (Fig. 2).

Immunocompetent cells (segmented leukocytes, macrophages, lymphoid cells) were actively involved in the resorption of the necrotic tumor after ECL exposure. Intense lasting lesions of the vascular network seem to be responsible for inhibition of relapsing tumor growth in comparison with the control.

Hence, treatment of M-1 sarcoma by ECL with horizontally positioned electrodes is sufficiently effective. The only side effect is enlargement of the damaged zone with increase of the parameters of exposure and hence, a higher risk of death for normal cells adjacent to the tumor.

REFERENCES

1. A. V. Barsukov, O. D. Varchuk, and R. A. Alibegov, *Gastroenterol.* (St. Petersburg), No. 3, 31 (2002).
2. B. Chen, Z. Xie, and F. Zhu, *Eur. J. Surg.*, Suppl., No. 574, 75-77 (1994).
3. C. K. Chou, J. A. McDougall, C. Ahn, and N. Vora, *Bioelectromagnetics*, **18**, No. 1, 14-24 (1997).
4. R. Li, Y. Xin, Y. Gu, *et al.*, *Ibid.*, 2-7.
5. D. Maintz, R. Fischbach, N. Schafer, *et al.*, *Invest. Radiol.*, **35**, No. 5, 289-294 (2000).
6. G. S. M. Robertson, S. A. Wemyss-Holden, A. R. Dennison, *et al.*, *Surgery*, **85**, 1212-1216 (1998).
7. B. D. Teague, S. A. Wemyss-Holden, B. G. Fosh, *et al.*, *Ibid.*, **72**, No. 2, 137-141 (2002).
8. A. Turler, H. Schafer, N. Schafer, *et al.*, *Langenbecks Arch. Chir. Suppl. Kongressbd.*, **115**, Suppl. I, 611-614 (1998).
9. H. Von Euler, E. Nilsson, J. M. Olsson, and A. S. Lagerstedt, *Bioelectrochemistry*, **54**, No. 2, 117-124 (2001).
10. H. Von Euler, K. Strahle, A. Thorne, and G. Yongqing, *Ibid.*, **62**, No. 1, 57-65 (2004).