INHIBITION OF METASTASIZATION OF LEWIS CARCINOMA IN MICE BY INTERFERON-INDUCING VASODILATOR DIPYRIDAMOLE

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According to data in the literature interferon inducers can inhibit tumor metastasization [3]. However, the clinical use of standard interferon inducers (for example, polyI:C) is difficult because of their toxicity and high cost. Interferon-inducing activity has more recently been discovered among certain vasodilators widely used in cardiologic practice [1]. One of these is dipyridamole (Curantil, Persantil), an effective coronary dilator. Interferon-inducing activity of dipyridamole is probably connected with its ability to inhibit cAMP phosphodiesterase, and with consequent accumulation of intracellular cAMP, followed by cAMP-dependent stimulation of immunocompetent cells and synthesis of type 1 pH-stable (α and β) interferons [1, 4, 8]. Maximal induction of interferon by dipyrisamole is usually observed 48 h after its peroral administration, and it depends essentially on the dose and mode of administration of the drug [1]. The present investigations showed that dipyridamole, together with radiotherapy, may be highly effective in the treatment of mice with experimental sarcomas [2].

The aim of this investigation was to compare the action of dipyridamole and the standard interferon inducer polyI:C (polyinosinic:polycytosinic acid) in a model of experimental metastasization in mice.

EXPERIMENTAL METHOD

Female C57BL/6 mice weighing 18-20 g and aged 10-12 weeks were used.

Lewis carcinoma was used as the model of metastasization. A suspension of living cells was prepared from a 14-day solid tumor. The number of living cells (60-80%) was determined by staining with trypan blue.

In the case of intravenous inoculation 2×10^5 tumor cells in 0.3 ml of Hanks' solution without heparin were injected into the caudal vein. On the 14th day after inoculation of the tumor the animals were killed and the number of metastases counted on the surface of the lungs, fixed beforehand in Boulin's solution. In the model with intramuscular inoculation 2×10^6 tumor cells in 0.2 ml of Hanks' solution were injected and the number of metastases counted on the 20th day.

The pharmacopoeial preparation of dipyridamole (Curantil, from VEB, East Germany) was administered perorally and intravenously in a dose of 500 µg, and polyI:C ("Calbiochem," Switzerland) parenterally in a dose of 50 µg in 0.2 ml physiological saline on the 1st, 7th, and 14th days after inoculation of the tumor.

EXPERIMENTAL RESULTS

Data on the antimetastatic activity of dipyridamole and polyI:C are given in Table 1. They show that after peroral administration of dipyridamole in the model with intravenous inoculation of the tumor cells, metastasization in the lungs was inhibited by almost 75%; dipyridamole, moreover, was found to be no less effective than the well-known interferon in-

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TABLE 1. Effect of Dipyridamole on Metastasization of Lewis Carcinoma in Mice

Experimental conditions	Number of metastases in lungs	
	after intravenous inoculation of 2×10^5 tumor cells	after intramuscular inoculation of 2×10^6 tumor cells
Control	24,4±1,2 (37)	21,5±1,1 (26)
Dipyridamole (500 μ per mouse, parenterally)	6,5±1,6 (9)	15,6±2,4 (8)
PolyI:C (50 μg per mouse, parenterally)	5,6±1,1 (11)	13,3±1,8 (8)
Dipyridamole (500 μg per mouse, intravenously)	42,3±3,7 (6)	44,8±3,9 (8)
PolyI:C (50 μg per mouse parenterally) + dipyridamole (500 μg per mouse, perorally)	5,9±1,3 (9)	16,8±1,4 (10)

Legend. Number of animals in each version of experiment shown in parentheses.

ducer, polyI:C. A special experiment also showed that preliminary injection of dipyridamole 24 h before intravenous inoculation of the tumor cells significantly ($p<0.01$) reduced metastasization to 11.6 ± 1.1 compared with 25.0 ± 1.8 in the control, i.e., dipyridamole also has a marked prophylactic action. Meanwhile intravenous injection of the same dose of dipyridamole caused a significant ($p<0.001$) increase in the number of metastases in the lungs.

In the case of combined administration of polyI:C (injected on the 1st, 7th, and 14th days) and dipyridamole (given on the 4th and 10th days after inoculation of the tumor) a synergic effect was not found and the reduction of metastasization corresponded to that of each preparation used separately.

In the model with intramuscular inoculations of the tumor dipyridamole, given perorally, also reduced the level of metastasization significantly ($p<0.01$), whereas the same dose injected intravenously led to an increase ($p<0.01$) in the number of metastases.

PolyI:C is known to stimulate antibody formation and cellular immune responses in animals in vivo and in vitro [5], a property linked with the ability of this compound to activate adenylate cyclase in immunocompetent cells and thereby to increase the intracellular cAMP pool [1]. The mechanism of the interferon-inducing action of dipyridamole also is based on ability to cause accumulation of intracellular cAMP by inhibiting phosphodiesterase [8]. The absence of a synergic effect with combined administration of polyI:C and dipyridamole also indicates that the two substances have a similar mechanism of interferon induction and, because of this, one cannot effectively potentiate the other. Considering that dipyridamole is an anticoagulant [7], and that for combined use of interferon inducers and anticoagulants in metastasization models synergic effects have been described [6], it can be concluded that it is unlikely that the mechanism of the antimetastatic activity of dipyridamole is based on its ability to prevent platelet aggregation, because of the absence of this effect in the present experiments (Table 1). The substantial increase in the level of metastasization following intravenous injection of dipyridamole is probably connected with inhibition of interferon synthesis due to considerable overdosage of the drug, for the interferon-inducing ability of dipyridamole is critically dose-dependent. The possibility cannot be ruled out, however, that dipyridamole, as a vasodilator, can affect the circulation and can hold up tumor cells in the capillaries, especially if the drug is injected intravenously.

It can thus be concluded from the results obtained by the use of a model of experimental metastasization in mice that the coronary dilator dipyridamole possesses high antimetastatic activity. Since dipyridamole is less toxic than the standard interferon-inducer polyI:C, clinical trials of it are likely to be promising.

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