COMBINATION THERAPY OF TRANSPLANTABLE TUMOUR IN MICE WITH CISPLATIN AND GLUCOSE

SUNITI SARNA and R. K. BHOLA
Department of Zoology, Gauhati University, Guwahati 781 014, India.

ABSTRACT

Cisplatin was found to be more effective when used in combination with glucose. This combination therapy significantly enhanced the survival time of the tumour-bearing mice and the number of tumour-free survivals. Subtherapeutical dose of cisplatin along with glucose was found to be more effective than cisplatin alone at a therapeutical dose.

INTRODUCTION

ANTI-TUMOUR compound cisplatin was found to be effective in the treatment of a variety of experimental tumours[1-4]. It has been shown that cisplatin regresses transplantable tumours through the enhancement of the host's immune system[5-11]. Although this compound increases the survival time of the animals when administered at a therapeutical dose, a carcinogenic risk always remains in the treated animals[12, 13]. Some anti-tumour drugs are more effective when used along with glucose[14-16]. Several anti-cancer drugs were tried in acidic conditions induced by tumour cell glycolysis and among these carbazilquinone was most cytocidal when coupled with Ehrlich ascites tumour cell glycolysis[16]. Chemotherapeutical studies were done by injecting tumour cells preincubated *in vitro* with different drugs and glucose[16]. The present study involves the direct treatment of tumour *in vivo* with cisplatin along with glucose to evolve a non-toxic chemotherapy for the selective destruction of tumour cells.

MATERIAL AND METHOD

Cisplatin was obtained as a gift from Bristol-Myers Co., USA. C_3H/He strain of mice (both males and females, 8–10-weeks-old, weighing 20–22 g) bearing transplantable Dalton's lymphoma was used. The day of tumour transplantation was taken as zero. On day 6 tumour-bearing animals were treated with a single injection (i.p.) of cisplatin (9 mg/kg or 3 mg/kg) alone or in combination with single or repeated injections of glucose. Controls were run for each concentration of glucose. The increase in the survival time of tumour-bearing mice and the tumour-free survivals were studied in experimental and control groups (table 1).

RESULTS AND DISCUSSION

Tumour cells are known to have a higher rate of glycolysis[17]. It has long been accepted that this vigorous glycolytic activity of tumour cells decreases the extra and intracellular pH due to the production of excess amount of lactic acid after incubation with glucose[15, 18-20]. The release of lactic acid by tumour cells treated with glucose *in vitro* has already been reported[14, 16, 21]. When 75 mg/kg glucose was injected i.p. in tumoured mice, this quantity is utilized by the tumour cells which lowers the pH in

Table 1 *Effect of different combinations of cisplatin and glucose on survival time of tumoured mice*

Group	Tumour-free survivals (%)	Maximum survival time of tumour-bearing mice (days)
9 mg/kg cisplatin (i.p.) + 75 mg/kg glucose (i.p.)	nil	14
9 mg/kg cisplatin (i.p.) + 100 mg/kg glucose (i.m.)	60	70
9 mg/kg cisplatin (i.p.) + 150 mg/kg glucose (i.m.)	80	58
9 mg/kg cisplatin (i.p.) + 50 mg/kg glucose (i.p.)	100	—
9 mg/kg cisplatin (i.p.)	57	35
3 mg/kg cisplatin (i.p.)	nil	55
3 mg/kg cisplatin (i.p.) + 5 repeated injections of 75 mg/kg glucose (weekly, i.p.)	80	38

their vicinity. This lowering of pH has some effect on tumour growth resulting in an increase in the survival time of the tumour-bearing mice (figure 1). When therapeutical dose of cisplatin (9 mg/kg, i.p.) was injected in tumoured mice, almost 57% of the mice showed complete regression after 3–4 days of treatment, appearing as tumour-free survivals. The rest of them died after a few days due to re-appearance of the tumour. When therapeutical dose of cisplatin was injected with 75 mg/kg of glucose i.p., all the animals died after 6–8 days of treatment (table 1, figure 1). However, animals receiving subtherapeutical dose of cisplatin (3 mg/kg) along with a single injection of glucose (75 mg/kg) i.p. survived with tumour growth. Despite giving subtherapeutical dose of cisplatin, in all animals, the tumour increased gradually in size without any sign of regression, but repeated injections of glucose (75 mg/kg, i.p.) for 5 weeks slow down the growth rate causing an increase in the survival time of the tumoured mice. Almost in 80% of the mice there was complete regression of tumour after repeated injections of glucose and they appeared as tumour-free survivals. Controls (repeated injections of glucose without cisplatin) of this group also showed regression in a few animals (figure 2), suggesting that a low pH or acidic conditions around the tumour cells may by itself be hazardous for their multiplication[14]. Animals receiving therapeutical dose of cisplatin (i.p.) with different concentrations of glucose (100 mg/kg, 150 mg/kg, i.m.) survived with slower rate of tumour growth. Similarly if a low dose of glucose (50 mg/kg) was injected i.p. along with therapeutical dose of cisplatin, all animals survived with tumour growth. In this case also, tumour developed but grew at a slower rate because of the active glycolysis combined with the anti-tumour effect of therapeutical dose of cisplatin. Complete regression of tumour in 60–100% of the mice was observed after 24–25 days of treatment with therapeutical dose of cisplatin in combination with different concentrations of single glucose injection (table 1, figure 1), suggesting that the anti-tumour effect of cisplatin gets slightly repressed first after coupling with vigorous glycolytic activity of tumour cells but its action gets potentiated as the required pH maintained in the vicinity due to the production of lactic acid by tumour cells. Therapeu-

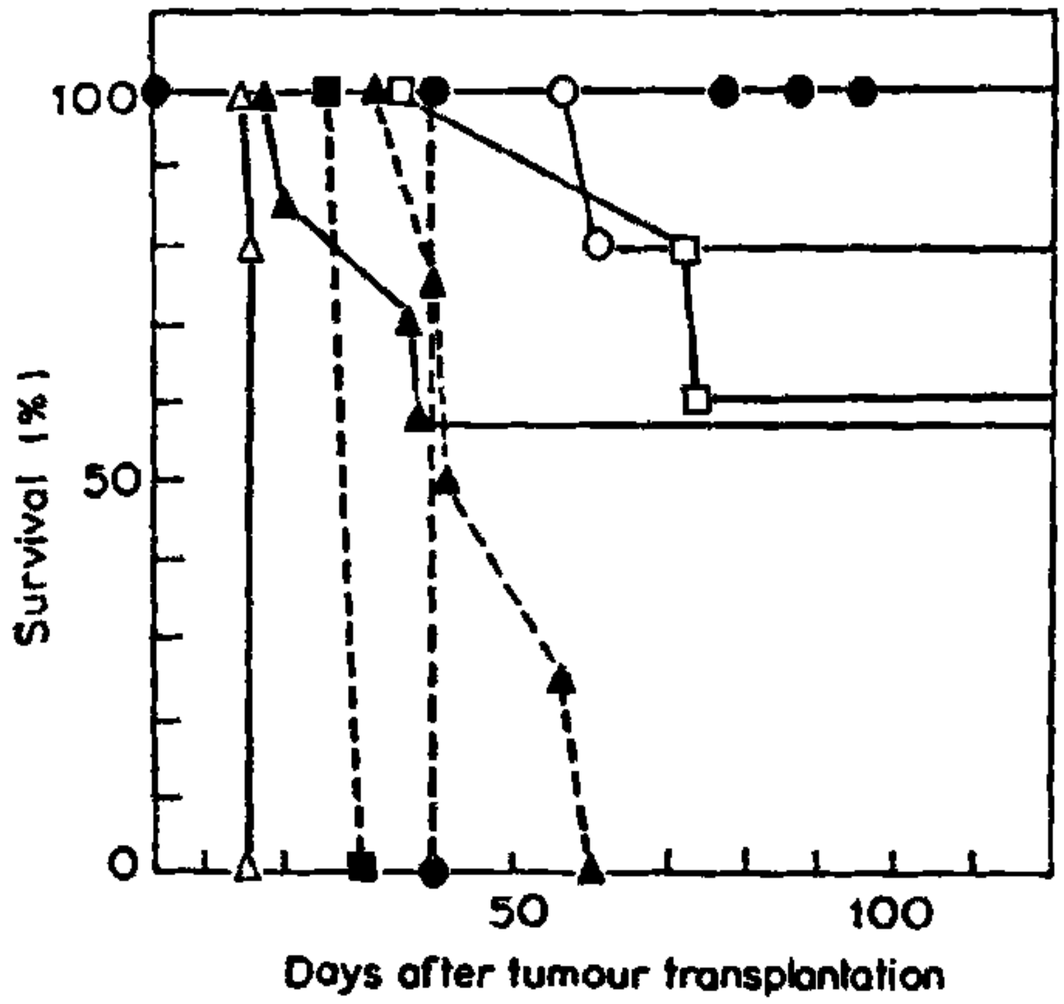

Figure 1. Therapeutical dose of cisplatin (9 mg/kg) with different doses of glucose (i.m. or i.p.) [●---●] 50 mg/kg glucose (i.p.) and 150 mg/kg glucose (i.m.), ●——● 50 mg/kg glucose (i.p.) + 9 mg/kg cisplatin (i.p.), ○——○ 150 mg/kg glucose (i.m.) + 9 mg/kg cisplatin (i.p.), □——□ 100 mg/kg glucose (i.m.) + 9 mg/kg cisplatin (i.p.), ■——■ 100 mg/kg glucose (i.m.), △——△ 75 mg/kg glucose (i.p.) + 9 mg/kg cisplatin (i.p.), ▲——▲ 9 mg/kg cisplatin (i.p.), ▲---▲ 75 mg/kg glucose (i.p.)].

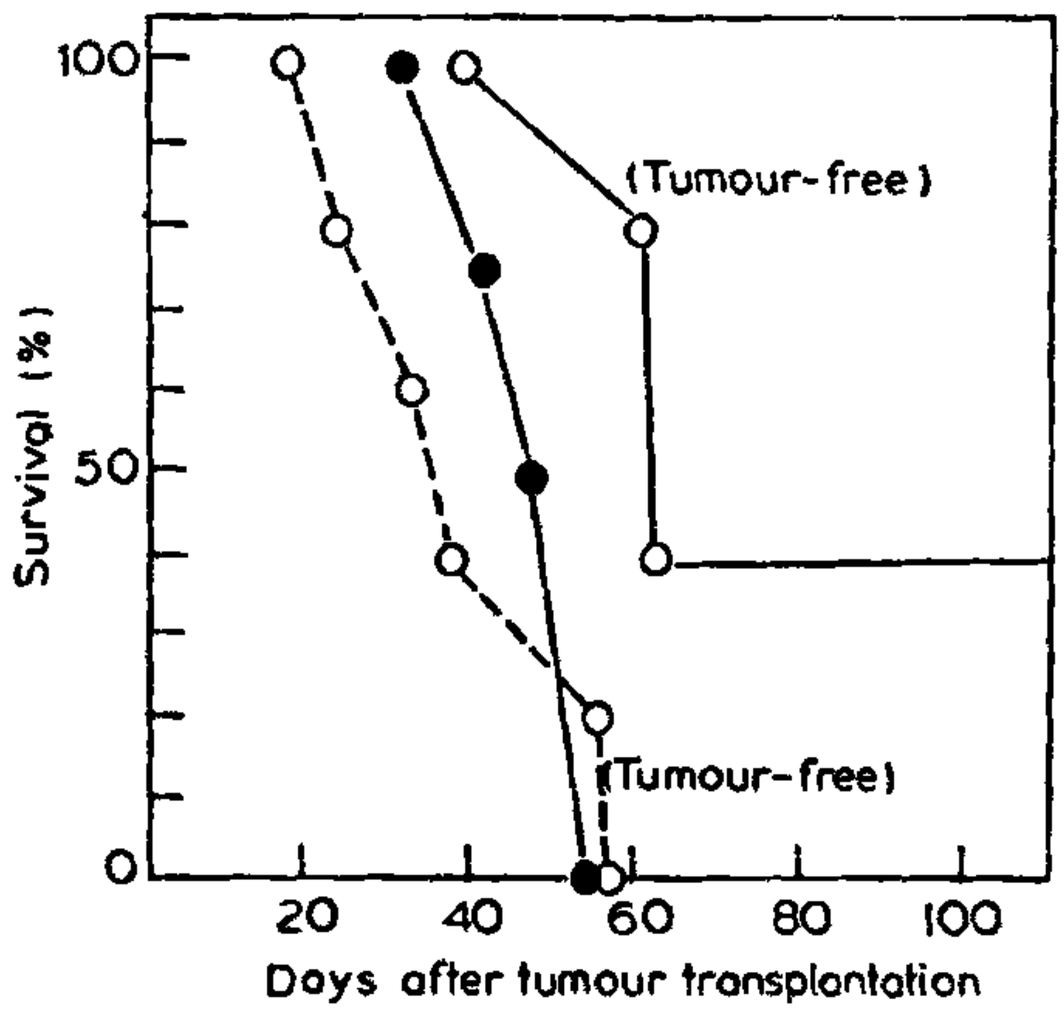

Figure 2. Subtherapeutical dose of cisplatin (3 mg/kg) combined with repeated injections of glucose. [○——○ 3 mg/kg cisplatin (i.p.) and 75 mg/kg glucose (i.p.) weekly, ○---○ 75 mg/kg glucose (i.p.) weekly, ●——● 3 mg/kg cisplatin (i.p.)].

tical dose of cisplatin with low (i.p.) or high (i.m.) dose of glucose exhibited an anti-tumour effect in the beginning to some extent but do not show regression even in a single mice, however, this combined treatment resulted in 60–100% tumour-free survivals at a later stage with an increase in the mean survival time of tumour-bearing mice up to 70 days. These results agree with earlier results where some anti-cancer drugs are reported more effective when coupled with tumour cell glycolysis[16]. It has been reported that cisplatin when given in a dose of 8.5 or 10 mg/kg produces a rapid increase in urinary protein and glucose associated with an increase in serum glucose and urea. The course of kidney injury produced by cisplatin (8.5 mg/kg) can be effectively followed over the period of 0–6 days[22]. In the present system, the mice usually die within 6–8 days of cisplatin and glucose (75 mg/kg i.p.) treatment (figure 1). This high dose of glucose may decrease the ascites pH very much, which instead of increasing the anti-tumour effect of cisplatin, adds to its side effects, unbearable by the animals. Despite this anti-tumour effect of therapeutical dose of cisplatin, a carcinogenic risk always remains at a later stage[12, 13]. To avoid this risk in the present system subtherapeutical dose of cisplatin with repeated injections of glucose was utilized to evolve a non-toxic therapy of this tumour. Such a low dose of cisplatin in combination with glucose is proved effective in causing anti-tumour effect in tumoured mice resulting in a significant increase in their survival time due to regression of tumour.

ACKNOWLEDGEMENT

One of the authors (SS) thanks UGC, New Delhi, for financial assistance.

4 June 1988; Revised 22 August 1988

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