STARVATION INDUCED STRESS RESISTANCE IN MICE

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Abstract

Starvation protects normal cells against toxicity stress but cancer cells which being mutated do not have protective mechanisms therefore stress results in death of cancer cells. In the present study we exposed normal and starved mice (24-36 hrs) to etoposide which induces oxidative stress. Starved mice remained physically healthy and active and they also gained weight while normal fed mice were lazy and there was only minor increase in weight as compared to starved mice. The present study provide evidence that the starvation has protective effects against stresses.

Introduction

Cancer is one of the most threatening diseases of the world. Cancer is considered uncontrolled cell growth due to mutations in pre-oncogenes. There is a number of treatments to cure cancer i.e. chemotherapy, radiotherapy, gene therapy etc. All the treatments have their own pros and cons, however chemotherapy is the most widely used treatment for the past 50 years. Firstly it was known that chemotherapeutic drugs selectively kill the tumor cells, but now it is a well understood fact that these drugs also damage the normal cells. So there should be such strategy which would selectively kill the cancer cells but not the normal cells.

Fasting or short term starvation have been proved to induce differential protection of normal cells but not the cancer cells against a vast variety of chemotherapeutic drugs in yeast mice and cancer cell lines (Lee *et al*, 2010, Raffaghello *et al.,* 2008). There is also an evidence of safe and efficient application of starvation based differential chemotherapy in human beings which greatly reduced the side effects induced by chemotherapy (Safdie *et al.,* 2009) but still not applicable due to susceptible side effects of starvation. Most reasonably starvation based differential chemotherapy works on the principle fact that cancer cells being mutated do not show any response against external stresses (temperature, pressure, toxicity etc.) unlike normal cells which show resistance against environmental stresses. The recent study was designed to evaluate the protective effects of starvation against oxidative stress in mice. The mice were evaluated for 10 days (including starvation time) for weight and behavior (lazy or active).

Materials and Methods

Starvation and induction of stress: Selected 6 mice (Swiss Webster) and categorized them into three groups as group A, B and C. The mice were weighed and allowed group A and group B to starve for 24hrs and 36hrs respectively. Group 3 was used as a control with no starvation. After starvation weighed the mice again and injected each mouse with 80mg/kg of etoposide including control (Table 1). The mice were observed for weight gain/loss and behavior for 8 days after drug treatment.

Table. 1. Starvation time and weight of mice before and after starvation.

Mice No.	Group No.	Starvation time (hrs)	Weight before starvation (gm)	Weight after starvation (gm)	Drug dose (etoposide) (µl)
1	A	Control	34	34	136
2		Control	29	29	112
3	B	24	41	36	164
4		24	35	32.3	130
5	C	36	41	36.5	140
6		36	33	29.4	120

Results

We examined SW mice to study the effect of starvation on chemotherapy and its subsequent effects. Mice were starved for 24 and 36 hrs (Table 1). There was around 10% decrease in body weight after starvation (Fig. 2a). Starved mice lost their weight during starvation but regained within 2-3 days of re-feeding.

Table 2. Weight changes after starvation.

Group No.	Starvation time	Weight after starvation (gm)							
		Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8
A	0hrs	34	34	33	34	34	34	34	34
	0hrs	28	29	29.5	30	29	30	29.5	30
B	24hrs	36	42	42	41.6	41.6	43.5	43.3	44
	24hrs	31	35.5	35	33	34.4	33.6	36.3	37
C	36hrs	40	41.5	43	44	41	42.5	43	44
	36hrs	31	33.6	34.5	35	33.8	35	34.3	35

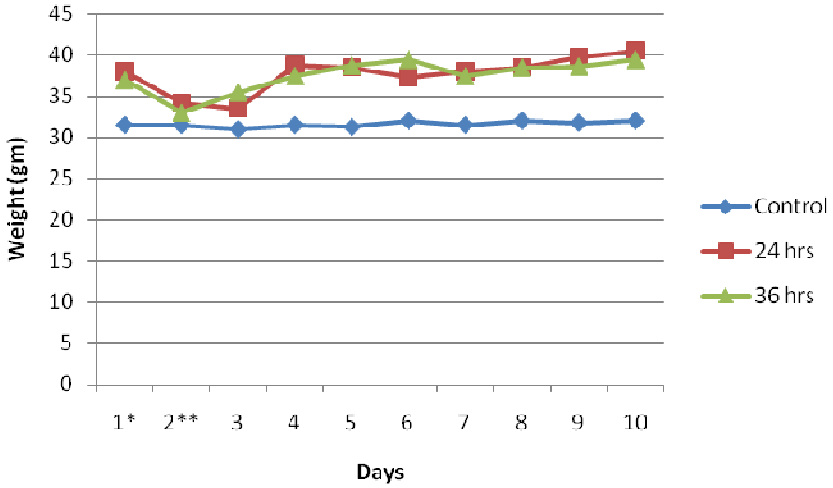

Fig. 1.Weight changes after starvation and etoposide treatment. Group A (control) showed no significant changes in weight while group B (24 hrs) and C (36 hrs) showed a remarkable weight gain.

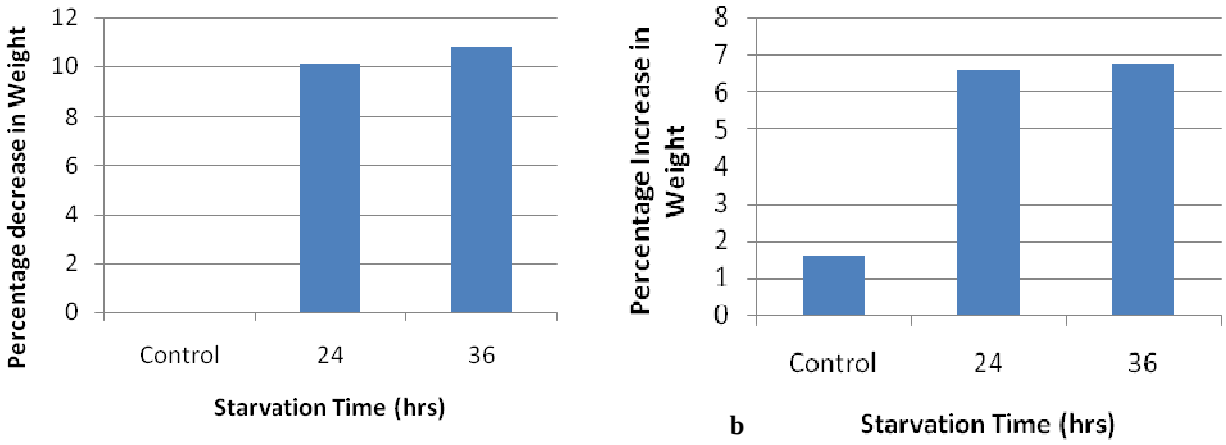

Fig. 2. Effect of starvation on weight of mice.**a**. Percentage decrease in weight of mice after 24 and 36 hrs of starvation, the weight remained unchanged in control. **b**. Percentage increase in weight from original weight after starvation and etoposide treatment in control and starved mice.

All the three groups were injected with 80mg/kg of the drug which quit high for human beings (30-40mg/kg) (Kroger *et al.,* 1998). All the mice survived including control. Pre-starved mice lost their weight upto5 gm while regained their original weights within 2 days after starvation on normal feeding. Mice were observed for 10 days and weighed them daily (Fig. 1, Table 2). Within this time period control group did not gain any remarkable weight (1.58%) whereas pre-starved mice showed 6.57 and 6.75% increase in their body weight after 8 days of drug induction and normal feed (Fig. 2b). After etoposide treatment controlled mice were lazy and inactive as compare to the pre-starved ones.

Discussion

Low calorie intake is good to treat many diseases including cancer. Studies indicated that short term starvation is effective in protecting normal cells when patient is treated by chemotherapeutic drugs (Raffagheloo *et al.,* 2008). Under the normal conditions the total energy of cells is balanced by cell division, cell maintenance or repair, while under stress condition all energy goes to cell maintenance. Cancer cells being mutated do not have such mechanism hence they do not response to stressed conditions (Raffagheloo *et al.,* 2008; Safdie *et al.,* 2009). The signaling pathway that are related to nutrient and stress resistance have key transcription factors that play an important role in stress resistance. These stress and nutrient signaling pathway have important role in growth, metabolism and stress against oxidants and toxins. These pathways are also conserved from yeast to human (Fontana *et al.,* 2009). Etoposide was used as a chemotherapy drug to induce stress, it is a widely used drug for cancer treatment which damages the cells by multiple mechanisms (Sawada *et al.,* 2001).

Some of the experiments were done on yeast and they were cultured in water instead of glucose (to induce stress) for some duration that resulted in increase in life span as well as resistance to oxidants and heat shock (Wei *et al.,* 2008).

In our experiment when we exposed the mice to etoposide after 24 and 36 hrs starvation, there was rapid increase in weight gain in starved mice as compared to control. Starved mice were also very active while there was minimal weight gain after 8 days of feeding in control mice and they were lazy. In another study when mice were on diet restriction, there was increased stress resistance. When mice were starved for 48-60 hrs and exposed to etoposide, they were protected against oxidative effects of chemotherapy (Raffaghello *et al.,* 2008). In a different study when CD-1 mice were starved for 72 hrs and then exposed to lethal dose of doxorubicin, they survived which normally cause death in normally fed mice due to oxidative stress induced cardiotoxicity (Lee *et al.,* 2010).

These results clearly indicate that starvation has protective effects on the body under different type of stress condition and on this basis differential chemotherapy could be very helpful for cancer patients to kill the cancer cells and protect the person from adverse side effects of chemotherapy.

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