USE OF HYPERBARIC OXYGEN IN EXPERIMENTAL FROST-BITE

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Frost-bite was produced in rats by exposing them to —15°C and the extent of injury in the hind limbs and the tail was assessed at the end of 15 days. Hyperoxic treatment at normal atmospheric pressure and 1·5 atmosphere was given to experimental groups for 30 minutes daily for seven days in a hyperoxic chamber immediately after cold exposure. The controls were not given any treatment. Single oxygen treatment at normal atmospheric pressure was of no value, however, repeated hyperoxic treatment showed limited improvement but repeated treatment for seven days with hyperbaric oxygen at 1·5 atmosphere showed distinct recovery of frozen parts.

Extensive studies have been carried out for the prevention and treatment of cold injuries, but the problem still remains unsolved. Hyperbaric oxygen (OHP) treatment for cold injuries may be considered optimistic. While a number of experimental studies reported the effectiveness of OHP therapy in healing and tissue survival[1,2], this was, however, not corroborated by subsequent observation[3]. The present study was undertaken to evaluate the effect of OHP therapy as a treatment for cold injury.

EXPERIMENTAL

Cold injury was produced in five groups of male rats, each group consisting of 15 rats. The body weight of the rats ranged from 172 to 240 gm. with a mean of 208 $\pm$ 1·89 gm. The rats were exposed to — 15°C $\pm$ 1°C in a thermostatically controlled deep freeze. The groups were made by random selection. The experiments were conducted in two series.

Series A : Three rats, one each from group 1, 2 & 3 were exposed to cold simultaneously, restrained in individual perspex harness with the tail protected[4]. At the end of the cold exposure the rats were taken out, the rat from group one was kept at room temperature and served as control. The other two were treated in the hyperoxic chamber for 30 minutes at atmospheric pressure. The rat from group two, received treatment only for one day and that from group three, received treatment for 30 minutes daily for seven days. Fifteen such experiments were conducted.

Series B : Two rats, one each from group 4 and 5 were simultaneously exposed to —15°C $\pm$ 1°C in a metallic container for three hours. Air circulation was maintained through a rubber tube and a circulation pump kept outside the deep freeze. The rats were free to move and the tail was not protected in order to assess the degree of injury in the tail as well. During exposure the rat assumes a posture to reduce its surface area and also the heat loss from the body. At the end of the experiment the rat from group four was kept at room temperature and served as control and the other from group five was given hyperbaric oxygen treatment at 1·5 atmospheric pressure for 30 minutes daily, for seven days. Fifteen such experiments were conducted.

The thawing in all animals was done at room temperature (20–25°C). After the treatment the rats were kept in experimental room for a period of one hour and then the legs were bandaged using dry sterile cotton to prevent infection and mechanical damage. The rats were transferred to the animal house which is

maintained at 20–25°C. The bandages were replaced daily for a period of 15 days at the end of which the degree of injury was assessed using the following criteria[5].

> Very severe, loss of entire paw or more (++++); Severe, loss of all the digits (+++); Moderate, loss of two phalanges or equivalent (++); Primary, minimal injury without loss of phalanges (+) and No tissue damage (0).

The hyperoxic and hyperbaric environment in the exposure chamber (volume 16·5 litres) was obtained by introducing oxygen from a pressurised oxygen cylinder. No water vapour was added to the system.

RESULTS AND DISCUSSION

The data of the A series of experiments (hyperoxic treatment) is given in Table 1. Statistical analysis has been done by Mann-Whitney U Test, noting the degree of injury expressed in numbers for each rat in all the groups. Statistical comparison in controls of series A and B shows no significant difference. It is seen that there is no significant difference in recovery between groups 1 and 2 but group 3 (hyperoxic) treated for seven days shows significant ($P < 0{\cdot}02$) improvement compared to control group 1.

TABLE 1

EFFECT OF HYPEROXIC TREATMENT IN EXPERIMENTAL FROST-BITE

Sl. No.	Group 1 (Control) Without O_2 treatment		Group 2 O_2 treatment only for one day		Group 3 O_2 treatment for seven days	
	Lt Paw	Rt Paw	Lt Paw	Rt Paw	Lt Paw	Rt Paw
1	+	0	+	+	+	0
2	++	++	++	+	+	0
3	0	+	+	++	0	0
4	+	0	+	+	+	+
5	+++	+	+	0	+	+
6	++	+	+	+	0	+
7	++	+++	++	++	+	+
8	+	++	++	++	+	+
9	++	+	+	+	0	+
10	+	0	0	0	++	0
11	++	++	+	+	+	+
12	+	+++	++	+	+	++
13	+++	++	++	+	+	+
14	+	++	+	+	+	+
15	++	++	0	+	0	+

The statistical comparison shows no significant difference between group 1 and 2. A significant difference ($P < 02$) is seen between group 1 and 3.

In the B series of experiment in which hyperbaric treatment was given, there was highly significant difference ($P < 0{\cdot}001$) in recovery between the experimental and control groups (Table 2).

TABLE 2

HYPERBARIC OXYGEN TREATMENT IN EXPERIMENTAL FROST-BITE

Sl. No.	Group 4 (Control) Without O_2 treatment			Group 5 (Experimental group) O_2 treatment at 1·5 atm. for seven days		
	Lt Paw	Rt Paw	Tail affected (%)	Lt Paw	Rt Paw	Tail affected (%)
1	+	+	67·4	0	0	0
2	+	++	70·1	0	0	0
3	++	++	84·4	0	0	0
4	+	+	34·9	0	0	2·7
5	0	0	0	0	0	0
6	++	+++	65·6	+	+	0
7	+	++	85·2	+	+	17·4
8	+++	++++	83·9	+	++	23·5
9	+	+	29·9	0	0	29·5
10	++	++	76·5	0	+	0
11	+	+	54·5	0	0	29·2
12	++	++	56·9	+	+	0
13	++	+	59·3	+	0	0
14	+	+	30·0	0	0	0
15	++	++	47·7	+	+	30·2
			Mean 56·4%			Mean 8·8%

Statistical comparison shows significant ($P < 0{\cdot}001$) difference between the groups.

The main conclusions derived from the data are (*i*) Hyperoxic treatment for a single day at atmospheric pressure does not enhance the recovery of the frozen parts in the experimental rats but if the treatment is repeated for seven days, a marginal improvement is seen. (*ii*) Hyperbaric oxygen therapy at 1·5 atmosphere leads to significant recovery of the frozen parts when the treatment is continued for seven days.

Frost-bite, the severest type of cold injury results from actual freezing of the tissues. Freezing below —5 to —7°C crystallises cell protoplasm which results in tissue damage. During thawing, blocking of blood vessels obstructs the circulation resulting in conglutination[6]. Liberation of histamine like substances, causes increased permeability of the capillary wall, collection of exudates in tissue spaces, edema and blisters[7]. In the final stage degeneration of red blood cells leads to hyaline mass production and the tissue damage is due to hypoxia.

Necrosis is primarily the result of vascular stasis and anoxemia and is not influenced by cold[8]. It is reported that hyperbaric treatment improves tissue oxygen content[9]. With its bacteriostatic action hyperbaric oxygenation may accelerate the tissue regeneration [9, 10]. In our study it is believed that hyperbaric oxygen treatment may increase the amount of oxygen dissolved in the blood, thereby supplementing more of oxygen which relieves tissue hypoxia. A similar effect may be achieved to a lesser extent, by repeated hyperoxic treatment as seen in group 3. Further, the hyperbaric oxygen treatment may lead to epithellal regeneration, and capillary proliferation[11]. When oxygen is administered under pressure, it increases the oxygen saturation of the blood plasma which helps the affected cells to receive the requisite amount of oxygen during thawing.

In the present study, OHP treatment was given immediately after freezing of the paw. Okuboye et al.[1], used similar procedure for hyperbaric oxygen treatment in rabbits. Our results of OHP therapy in series B support their finding. On the other hand Hardenberg working on mice with the paw exposed to —30°C for 1–2 min., treated them with OHP, 3–4 hours after freezing[3]. The cold exposure was rather of a very severe degree and the damage to the tissues might have been complete and could not be reversed by OHP. The recovery by hyperbaric oxygen in our experiments shows beneficial effect of OHP in the prevention of cold injury of moderate to severe intensity when the therapy is instituted early. It eliminates the aggravating influence of hypoxaemia on tissue damage. Significant degree of recovery is possible when the therapy is instituted early and when the tissue damage is not complete.

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