Effects of Hyperbaric Oxygenation on Metabolism
VII. Succinate Protection Against Oxygen Toxicity in Large Animals
(35328)

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Significant protection was demonstrated against the development of convulsive activity in small animals using 0.4 *M* sodium succinate (1–4) injected intraperitoneally in a dosage of 12 m*M*/kg of body weight. Succinate protection against hyperbaric oxygen (HPO) at 5, 7, 9, and 11 ATA of 100% oxygen (5) was shown not to be due to the hyperosmolarity of the solution infused and led to our proposal that maintenance of normal ATP concentrations in brain and other tissues is of prime importance in protecting animals subjected to HPO. No significant delay of onset of convulsive activity was observed with hyperosmolar solutions of NaCl, sodium malate, an NAD-linked TCA cycle intermediate, or glucose. These results are consonant with the observation of Chance *et al.* (6) that HPO adversely affects NAD-linked substrates of oxidative phosphorylation, and have stimulated further studies using the FAD-linked substrates, succinate and alpha-glycerophosphate. We report here the results of experiments using sodium succinate as a protective agent against the toxic effects of HPO in large animals. These studies were carried out to determine the feasibility of using succinate in humans as a protective agent when HPO is used in the treatment of certain clinical conditions such as gas gangrene.

Dogs (12–18 kg) were infused intravenously with 0.4 *M* sodium succinate, pH 6.4, in a dosage of 8 m*M*/kg of body weight/hr, for 50 min prior to and during exposure to 100% oxygen at a pressure of 40 psig. Control experiments were conducted by subjecting the same animals, either untreated or infused with equivalent doses of saline or sodium malate, to identical conditions of HPO 48–72 hr prior to the succinate experiments.

Thus, each dog served as its own control, and time to convulsions could be compared directly (Table I). Extensive preliminary experiments have shown that the sequence of the two HPO exposures does not affect the susceptibility to seizures; the above sequence was designed to exclude the occasional dog which fails to convulse prior to death. No change in time to convulsion was observed when saline or sodium malate were infused into the control animals.

Table I indicates the differences observed in time to convulsions when a group of 13 animals was given no infusion and 2 days later were infused with sodium succinate. Prolongation of the time to convulsion was noted in all dogs infused with succinate, with protection ratios of 1.2 to 5.63. Those animals demonstrating protection ratios less than 2.0 appeared agitated and fearful of the hyperbaric chamber and experimental manipulations; without exception the calmer dogs had a much higher protection ratio. Consequently, studies using preliminary tranquilization with prochlorperazine (Compazine) were undertaken.

Table II shows the results obtained with a series of dogs that were given intramuscular Compazine (0.26 mg/kg of body wt) 30 min prior to treatment with HPO. Compazine was given to both control and succinate-treated animals, and thus played no direct role in the protection ratio obtained. In addition, the control convulsion times in Table I and II clearly demonstrate that this dosage of Compazine yields no protection against HPO. It is significant that the tranquilizer does calm the animals. At *t* test comparison of the pro-

tection ratios of the tranquilized and nontranquilized groups indicated that they were not significantly different. However, when using Compazine in conjunction with succinate all 15 protection ratios were equal or greater than 2.0. In contrast, four of the group of 13 nontranquilized animals had protection ratios less than 2.0 (1.20, 1.33, 1.54, and 1.68).

The results reported here are favorable to the use of sodium succinate as a protective agent in humans undergoing HPO treatment. Pharmacological studies now in progress indicate that these relatively large drug doses employed in the present experiments are satisfactorily tolerated by large animals.

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TABLE I. Time to Convulsions—Normal Dogs—No Tranquilizer (40 psig, 100% O_2).

Dog no.	Convulsion time (min) Control	Succinate[a]	Protection ratio
6485	25.0	>90	3.60
6486	13.0	20	(1.54)
6508	19.0	>90	4.74
6512	19.0	>90	4.74
6517	11.5	49	4.26
6518	9.25	45	4.86
6533	30.0	40	(1.33)
6550	16.0	>90	5.63
6559	19.0	61	3.21
6573	12.5	21	(1.68)
6574	30.0	90	3.00
6579	17.5	21	(1.20)
6580	16.0	66	4.13
n	13		
$\bar{X}$	18.3	59.5	3.38
σ	6.3	27.6	1.46
t		5.03	
p		<0.0005	(9/13)>2.0
Range	9.25–30	20–90	1.2–5.63

[a] Succinate: 8 mM/kg of body wt, 0.4 M sodium succinate (pH 6.4) iv infusion 50 min pre-HPO and during exposure.

TABLE II. Time to Convulsions—Tranquilized Dogs (Compazine im, 0.26 mg/kg, 30 min pre-HPO; 40 psig, 100% O_2).

Dog no.	Convulsion time (min) Control	Succinate[a]	Protection ratio
6594	17.0	>90	5.29
6596	17.0	75	4.41
6602	14.5	>90	6.21
6603	16.0	48	3.00
6604	24.5	57	2.33
6625	11.5	23	2.00
6632	11.0	24	2.18
6636	18.0	41	2.28
6638	20.0	43	2.15
6666	31.5	>90	2.86
6668	14.5	41	2.83
6669	12.0	71	5.92
6680	11.25	>90	8.00
6683	19.0	56	2.95
6686	9.5	>90	9.47
n	15		
$\bar{X}$	16.5	61.9	4.13
σ	5.6	24.0	2.27
t		6.902	
p		<0.0005	(15/15)>2.0
Range	9.5–31.5	23–90	2.00–9.47

[a] Succinate: 8 mM/kg of body wt, 0.4 M sodium succinate (pH 6.4) iv infusion 50 min pre-HPO and during exposure.

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