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METABOLIC AND FUNCTIONAL CHANGES IN THE HEART DURING PROLONGED HYPOTHERMIA

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ABSTRACT

The effect of hypothermia of 25° C for 24 hours on myocardial metabolism and efficiency was determined on dogs fasted for approximately 15 hours and anesthetized with sodium pentobarbital. Coronary blood flow, cardiac output, myocardial oxygen and substrate utilization, and mechanical efficiency of the heart were determined at normal and reduced body temperatures. Prolonged reduction of myocardial temperature, with concomitant reduction in coronary blood flow, led to diminished oxygen and substrate utilization. Myocardial glycolysis began following 12 hours of cooling when pyruvate utilization stopped in negative balance. After 24 hours the heart stopped utilizing carbohydrates, with negative arteriovenous differences for these substrates (in the presence of normal arterial carbohydrate levels), but continued to utilize nonesterified fatty acid. The coefficient of oxygen utilization for the heart increased following 24 hours of cooling, suggesting a relative state of myocardial hypoxia. The appearance of hypoxia and glycolysis during the late hours of cooling suggest that the heart's limit of tolerance to cooling was near.

PUBLICATION REVIEW

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SECTION 1. INTRODUCTION

Previous studies indicate that despite reduction in cardiac output during hypothermia, oxygen transport remains adequate since arteriovenous oxygen difference does not increase beyond a normal range. However, since the venous level of oxygen might not have been determined by the extraction of oxygen by the tissue but rather by the affinity of oxygen to hemoglobin, a constant A-V oxygen difference is not necessarily indicative of adequate tissue oxygenation during hypothermia. Furthermore, even an increase in A-V oxygen difference within the normal normothermic range in the hypothermic heart might reflect serious impairment in metabolism and function of the heart.

Fisher, Russ and Fedor (1957), Lewis (1961) and Connaughton, Holt and Lewis (1961) have observed that during the late hours of prolonged hypothermia, cardiac output is markedly depressed and stagnant anoxemia develops. Fisher et al (1957) reported that most dogs dying during the late hours of hypothermia did so following stagnant anoxemia. Russ and McCollester (1959) have observed that the decline in cardiac output and venous oxygen saturation appears to be associated with an increase in plasma transaminase activity. In view of these changes and since transaminase is notably high in the myocardium, our attention in the present study has been directed to the heart and its role in the causation of death in dogs exposed to prolonged hypothermia.

SECTION 2. METHODS

Six mongrel dogs varying in weight from 18 to 22 kg were fasted for approximately 15 hours and anesthetized with sodium pentobarbital.

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* This research was conducted in accordance with the "Principles of Laboratory Animal Care" of the National Society for Medical Research.

Anesthesia was administered as often as needed during the experiment. The coronary sinus and right ventricle were catheterized with the aid of fluoroscopy. Both femoral arteries were cannulated. Each animal was studied at normal and reduced body temperature. Left ventricle coronary flow was measured over a ten-minute period using the nitrous oxide saturation technique (Eckenhoff et al, 1948). Arterial and coronary sinus blood samples were drawn simultaneously for measurement of myocardial oxygen (Peters and Van Slyke, 1932), glucose (Somogyi, 1945), pyruvate (Friedemann and Haugen, 1943), lactate (Barker and Summerson, 1941) and nonesterified fatty acid (NEFA) (Dole, 1956) utilization. Blood was analyzed for nitrous oxide by the method of Galla, Ottenstein and Sancetta (1962). Coronary sinus and arterial plasma glutamic oxalacetic transaminase (PGO-T) activity was determined by the method of Karmen, Wroblewski and LaDue (1955). Left ventricle work, efficiency and substrate utilization were calculated (Goodale and Hackel, 1953).

Immediately after collecting samples for normothermic or control studies, the animals were immersed in a 4° C water bath and removed after the rectal temperature reached 28° C. The temperature was allowed to drift to 25° C where it was maintained for 24 hours with a Therm-O-Rite blanket. After cooling, all animals were administered 95% oxygen and 5% carbon dioxide by positive pressure, except during measurement of coronary blood flow when respiration was maintained by an electrophrenic respirator. Samples were collected at normal temperature and immediately following 12 and 24 hours of cooling. Two dogs died immediately after removal from the bath, while four dogs survived 24 hours of hypothermia. The following results were obtained from the four dogs surviving 24 hours of cooling.

SECTION 3. RESULTS

Hemodynamics

The reduction in left ventricle work following 12 hours of hypothermia was insignificant. After 24 hours a significant reduction in left ventricle work occurred, which corresponded to a significant fall in cardiac output and arterial blood pressure. Coronary blood flow and cardiac output were relatively constant following 24 hours of cooling. While myocardial oxygen consumption and the coefficient of oxygen extraction decreased significantly after 12 hours, the oxygen extraction coefficient rose following 24 hours as oxygen consumption continued to decline. There was a proportionately greater decrease in myocardial oxygen consumption than left ventricle work after 12 hours, which resulted in an increase in myocardial efficiency. The increase in oxygen consumption and left ventricle work was proportionately the same following 24 hours, indicating stabilization of myocardial efficiency (Table I).

TABLE I

MEAN EFFECTS OF PROLONGED COOLING
ON HEMODYNAMICS AND OXYGEN METABOLISM

(MEAN ± SE_M)

Time (Hours)	Control	12	24
Temperature (^{o} C)	35.5	24.9	24.6
Cardiac Output (liters per min)	3.165 ±0.29	3.151 ±0.44	2.152 ±0.18
Coronary Blood Flow (cc per 100 gm per min)	189.47 ±17.46	162.10 ±17.2	95.42 ±31.13
Blood Pressure (mm Hg)	144.167 ±12.92	110.333 ±7.75	84.467 ±9.79
Heart Rate (beats per min)	128 ±23	66 ±5	70 ±12
Myocardial Efficiency (%)	17.41 ±2.52	24.75 ±6.54	35.52 ±14.82
Left Ventricle Work (kg-m per min)	6.726 ±0.932	5.375 ±0.906	2.375 ±0.376
O_2 Arterial Concentration (vol %)	16.87 ±0.71	17.45 ±1.58	10.74 ±1.05
O_2 Consumption (cc per 100 gm per min)	21.63 ±2.53	11.97 ±1.22	5.59 ±2.37
O_2 Myocardial Coefficient of Extraction (AV/A) (%)	69.23 ±3.68	43.93 ±5.57	60.16 ±17.32

Metabolics

Carbohydrates. Arterial lactate decreased significantly from a control level of 10.92 mg % to 3.97 mg % after 24 hours. Myocardial extraction and utilization of lactate decreased after 12 hours and stopped most strikingly after 24 hours of cooling, with a negative arteriovenous difference. Arterial pyruvate declined from a control level of 1.50 mg % to 0.82 and 0.76 mg % following 12 and 24 hours respectively. Myocardial pyruvate extraction stopped after 12 hours, with a negative arteriovenous difference. Arterial levels of glucose were relatively constant. Myocardial glucose extraction decreased following 12 hours and stopped after 24 hours, with a negative arteriovenous difference.

Nonesterified fatty acids. Arterial NEFA decreased from a control level of 15.47 mg % to 9.62 mg % after 12 hours of hypothermia but rose to 13.87 mg % following 24 hours. Myocardial consumption of NEFA declined after 12 hours but remained relatively constant after 24 hours. The coefficient of extraction for this substrate decreased from 18 to 12% after 12 hours but rose to 52% following 24 hours.

Plasma glutamic oxalacetic transaminase. Since it is generally agreed that the range of normal for transaminase varies according to the method of analysis and the laboratory performing the test, present arbitrarily fixed normal and abnormal values, as suggested by Sampson (1958), are not separated by a "gray" zone of marginal abnormalities. In this study the mean control value was selected as the normal level of PGO-T activity (per ml of plasma): normal 17 units, marginal 17-42 units and abnormal over 42 units. Prolonged myocardial cooling resulted in an insignificant marginal and abnormal increase in PGO-T after 12 and 24 hours respectively. Mean normothermic coronary venous PGO-T exceeded that of the arterial plasma, but following cooling arterial PGO-T activity was higher and remained higher after 24 hours (Table II).

SECTION 4. DISCUSSION

The normothermic or control observation in this study agrees with those reported from other laboratories that myocardial substrate utilization is a function of arterial substrate concentration. However, prolonged reduction of myocardial temperature, with concomitant reduction in coronary blood flow, leads to diminished oxygen and substrate utilization and ultimately to negative carbohydrate balance. These changes occurred at higher than normothermic threshold levels of utilization for each carbohydrate substrate.

TABLE II

MEAN METABOLIC CHANGES DURING PROLONGED COOLING
(MEAN ± SE_M)

TIME (Hours)	Control	12	24
TEMPERATURE (° C)	35.5	24.9	24.6
LACTATE			
Arterial Concentration (mg%)	10.92 ±0.79	3.97 ±1.81	6.95 ±2.11
Utilization (mg per 100 gm per min)	9.085 ±3.22	3.25 ±2.91	0
Myocardial Coefficient of Extraction (AV/A) (%)	43.602 ±15.15	21.93 ±14.08	0
PYRUVATE			
Arterial Concentration (mg%)	1.5 ±0.26	0.82 ±0.21	0.76 ±0.19
Utilization (mg per 100 gm per min)	0.72 ±0.18	0	0
Myocardial Coefficient of Extraction (AV/A) (%)	30.6 ±4.92	0	0
GLUCOSE			
Arterial Concentration (mg%)	75.03 ±6.7	74.03 ±21.2	79.43 ±14.7
Utilization (mg per 100 gm per min)	24.13 ±9.89	18.79 ±9.41	0
Myocardial Coefficient of Extraction (AV/A) (%)	15.51 ±5.86	16.19 ±9.36	0
NONESTERIFIED FATTY ACIDS			
Arterial Concentration (mg%)	15.47 ±2.76	9.62 ±4.4	13.87 ±2.98
Utilization (mg per 100 gm per min)	5.996 ±8.02	4.43 ±4.2	4.67 ±4.5
Myocardial Coefficient of Extraction (AV/A) (%)	18.4 ±26.2	12.16 ±21.65	52.18 ±16.03
PLASMA GLUTAMIC OXALACETIC TRANSAMINASE			
Arterial Units	11.3	38.7	57.0
Coronary Sinus Units	16.7	32.3	48.0

An important factor in determining the metabolic response of the heart to reduced coronary blood flow during hypothermia is the ratio of oxygen demand of the myocardium to its supply. That the oxygen demand of the heart following 12 hours of cooling was proportionately less than its supply, in the present study, is reflected by a decreased coefficient of oxygen extraction. However, the coefficient of oxygen extraction increased after 24 hours, indicating that myocardial oxygen demand had become proportionately greater than supply and suggesting relative myocardial hypoxia. The fall in arterial oxygen does not appear to be related to the increase in the coefficient of oxygen extraction, since these two factors have an inverse relation with a correlation coefficient of 0.07. This observation is supported by Hackel, Goodale and Kleinerman (1954), who reported that the oxygen extraction coefficient of the normothermic heart had no relation to arterial oxygen levels as long as the arterial blood maintained its content above a threshold level of 8 volumes per cent. Below that level the two factors had an inverse relationship with a coefficient of correlation of 0.89. Consequently, the fall in arterial oxygen does not appear to be the cause of myocardial hypoxia in these experiments.

Myocardial glycolysis appears to have begun after 12 hours of cooling when pyruvate utilization stopped in negative balance. As the hours of cooling were prolonged, an increase in glycolysis was indicated when myocardial carbohydrate metabolism was observed to be in negative balance. The concomitance of hypoxia and glycolysis in the hypothermic heart in this study resembles that observed in the normothermic heart when subjected to partial and complete oxygen deprivation (Hackel et al, 1954.) Since these two factors are related with a coefficient of correlation of 0.99, glycolysis in the present study probably resulted from relative myocardial hypoxia.

Inasmuch as myocardial utilization of carbohydrate stopped in the presence of relatively normal arterial carbohydrate levels and since the heart continued to utilize NEFA, preferential utilization of this substrate is suggested during the late hours of cooling. The reason for selective demand of NEFA by the heart is not clear. Bernhard et al (1961) observed selective utilization of NEFA by the heart in dogs subjected to hypothermic cardiac arrest at 10° C from 20 to 30 minutes. They suggested that utilization of NEFA by the heart, in their study, might be related to the oxygen debit and metabolic lactic acidosis which appeared in all animals at the conclusion of rewarming.

Russ and McCollester (1959) have observed PGO-T activity to increase in mixed venous blood of dogs after prolonged sodium pentobarbital anesthesia. White (1956) has demonstrated an increase in serum GO-T activity in cancer patients suffering from inanition which paralleled the loss of creatinine, potassium and nitrogen indicating muscle damage. The increased activity returned to normal when the patients were fed large amounts of protein, suggesting that the muscle was transiently damaged. In the present

study, myocardial metabolism was altered after cooling, indicating a change in the state of nutrition of the heart; perhaps the rise in coronary venous PGO-T activity is a reflection of the state of nutrition of the hypothermic heart. Whether the increased venous PGO-T observed in this study resulted from relative myocardial hypoxia, prolonged sodium pentobarbital anesthesia, the nutritional state of the heart of (as has been suggested) increased permeability of the cellular membrane, or a combination of these factors is not known. However, since it has been demonstrated that the rise in SGO-T is proportional to the amount of myocardial damage (Lemley-Stone et al, 1955: Wroblewski, 1957; Wroblewski and LaDue, 1956), it seems reasonable to assume that the amount of damage under the conditions of these experiments is reflected by the magnitude of increase of coronary venous PGO-T. Based on this assumption and the arbitrarily fixed values assigned to levels of PGO-T, damage to the myocardium was minimal.

It is rather significant that a slightly damaged hypothermic heart managed to maintain an improved mechanical efficiency in a state of relative hypoxia and glycolysis. Whether this is due to preferential utilization of NEFA is not known. However, Katz (1960) has made an observation of similar significance. He observed that when the heart rate, cardiac output and blood pressure were maintained constant, severe hypoxia caused oxygen consumption of the normothermic heart to decline, so that utilization of the energy of the heart for propelling blood actually improved. Katz (op. cit.) also reported similar improvement in external mechanical efficiency of the heart when put under great working stress. It was not established whether this improvement in mechanical efficiency was due to the heart making better use of its chemical fuel, to the heart size being reduced so that tension is more effectively converted to cavity pressure, or to the heart making more use of the substrate passing to its muscle. Further study is needed to establish the role of substrate utilization in the improvement of mechanical efficiency of the hypothermic heart in a state of relative hypoxia and glycolysis. Despite the maintenance of improved mechanical efficiency of the heart after prolonged cooling in the present study, the appearance of a relative state of hypoxia and glycolysis indicates that the heart's limit of tolerance to cooling was near.

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