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Experimental Lactic Acidosis: Effect of Nitroprusside[1] (40218)

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Since Huckabee described lactic acidosis as a distinct entity (1), many clinical conditions have been associated with lactic acidosis (2). The diagnosis of lactic acidosis is evident when there is a metabolic acidosis with an increase in unmeasured anions, elevated arterial lactate concentration, increased arterial lactate to pyruvate ratio and formation of excess lactate. Many disease states associated with lactic acidosis demonstrate increased lactate production and decreased lactate oxidation with resultant accumulation of excess lactate (2).

Therapeutic maneuvers employed for the treatment of lactic acidosis have not been successful unless the associated disease can be altered. Mortality has remained high despite the use of alkalinizing solutions, methylene blue and hemodialysis (2).

Recently, Taradash and Jacobson (3) used sodium nitroprusside, a vasodilator, to treat a normotensive patient with lactic acidosis. There was significant improvement in cardiac performance in association with resolution of the lactic acidosis. These observations raised the question of whether the beneficial effect of sodium nitroprusside was due to the improvement in circulatory dynamics or to some direct effect on lactate/pyruvate metabolism.

In light of these observations and questions, we developed an animal model of lactic acidosis associated with hypotension. We examined the effect of sodium nitroprusside on the lactic acidosis and cardiovascular hemodynamics.

Materials and methods. All studies were performed on mongrel dogs, 15–20 kg in weight, fed a standard kennel ration. The day of the study the animal was anesthetized with intravenous pentobarbital, 25 mg/kg, and supplemental doses were given throughout the experiment to maintain anesthesia. The animal was intubated with an endotracheal tube and ventilated with a mechanical respirator to maintain arterial pH near 7.45. Two catheters were placed in the right atrium via the external jugular veins for obtaining central venous blood samples, sodium nitroprusside infusion, and indocyanine green injection for cardiac output measurements. A carotid artery catheter was placed for measurement of mean arterial pressure and arterial blood sampling. An external electromagnetic flow probe was placed on the exposed femoral artery (FA) and led to an electromagnetic flow meter; this system was calibrated *in vivo* at the end of each experiment. A retention catheter was placed in the urinary bladder. A catheter connected to an overhanging reservoir bottle was placed in the other femoral artery. After completion of surgery, the animal was given heparin, 100 U/kg, intravenously. Thirty minutes were allowed for equilibration before proceeding with the experiment.

The experiment was divided into three twenty minute periods: control (C) period, lactic acidosis (LA) period, and sodium nitroprusside (NP) period. During the control period, two carotid artery and central venous blood samples were collected ten minutes apart. Simultaneous arterial pH, pO_2, and pCO_2 measurements were made. Duplicate cardiac output measurements were performed at the end of the period using the indocyanine green dye dilution technique. To create lactic acidosis, the dog was hemorrhaged over 45 min from the femoral artery into the overhanging reservoir bottle until a mean arterial pressure of 50 mmHg was achieved. Periodic adjustment of the height of the hemorrhage reservoir bottle above the dog permitted maintenance of mean arterial pressure at 50 mmHg. The animal was allowed to equilibrate for 30 min. Arterial

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blood samples were then drawn at 30-min intervals until a stable arterial pH of less than 7.25 was achieved, usually within 30–60 minutes. At this point, the sampling and measurements described in the control period were repeated.

When the lactic acidosis period was completed, intravenous sodium nitroprusside was infused using a constant infusion pump. The sodium nitroprusside period was designed to study the effect of a low and a high dose of sodium nitroprusside. Following 30 min infusion of the low dose of sodium nitroprusside, 0.27 μg/kg/min ($N = 9$), sampling and measurements were made as previously described. The infusion of sodium nitroprusside was increased to 2.23 μg/kg/min ($N = 6$) and, after 30 min, sampling and measurements were repeated.

Mean arterial pressure (MAP) was measured with a pressure transducer and recorded with the electromagnetic flow meter output on a direct writing recorder. Cardiac output (CO) was determined by the dye dilution method. Indocyanine green was injected into the right atrium and arterial blood was withdrawn from the carotid artery through a cuvette densitometer by a constant withdrawal syringe pump. Cardiac output was calculated according to the method of Kinsman, Moore and Hamilton (4). Femoral vascular resistance = MAP/Femoral Artery Flow, mmHg/ml/min and total peripheral vascular resistance = MAP/CO, mmHg/l/min. Arterial pH, pO_2 and pCO_2 were measured with an Instrumentation Laboratories Model 113 pH/Blood Gas Analyzer. Arterial plasma bicarbonate (HCO_3^-) concentration was derived from arterial pCO_2 and pH values and the Henderson-Hasselbalch equation using a pK of 6.11 and a solubility coefficient of CO_2 in plasma of 0.0307 mEq/l/mmHg.

Blood samples for lactate and pyruvate determination were collected into cold 5% metaphosphoric acid as described by Marbach and Weil (5); this permitted refrigeration of samples until centrifugation, separation and freezing of the supernatant. Lactate was determined by the enzymatic method of Marbach and Weil (5) while pyruvate was determined by the enzymatic method of Bucher et al (6). The two values for lactate and pyruvate in each period were averaged to give a single value for that period. Excess lactate, $xL = (L_o - L_n) - (P_o - P_n)(L_n/P_n)$ where L_o and P_o are observed arterial lactate and pyruvate concentrations and L_n and P_n are normal arterial lactate and pyruvate contrations, respectively (1). L_n and P_n were taken as the mean arterial levels of lactate and pyruvate during the control period; $L_n = 18.9 \pm 1.9$ mg%, $P_n = 2.30 \pm 0.21$ mg% and $L_n/P_n = 8.2 \pm 0.8$. The data in the text and figures are expressed as means ± SE. The Student's t test was used for statistical analysis of paired data within each group (7).

Results. The results are illustrated in Figs. 1–3. Controlled arterial hemorrhage (Fig. 1) resulted in a decrease in MAP from 113 ± 4 mmHg in the control period to 48 ± 2 mmHg in the lactic acidosis period. During both sodium nitroprusside periods, MAP decreased further. Cardiac output and femoral artery flow fell in parallel, 53% and 57% respectively, during controlled arterial hemorrhage. During both sodium nitroprusside periods, cardiac output remained unchanged while femoral artery flow showed a small but progressive increase. Femoral vascular resistance decreased from 3.23 ± 0.76 mmHg/

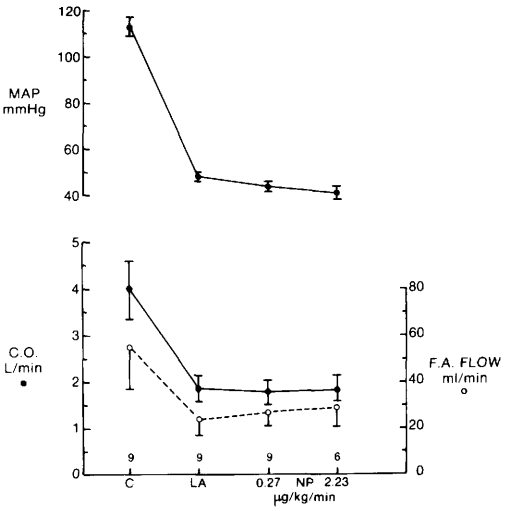

FIG. 1. Hemodynamic response to controlled arterial hemorrhage and sodium nitroprusside infusion. C = control, L = lactic acidosis, NP = sodium nitroprusside, MAP = mean arterial pressure, C.O. = cardiac output, F.A. = femoral artery. Data are means ± SE; numbers along abscissa indicate number of dogs studied in each period.

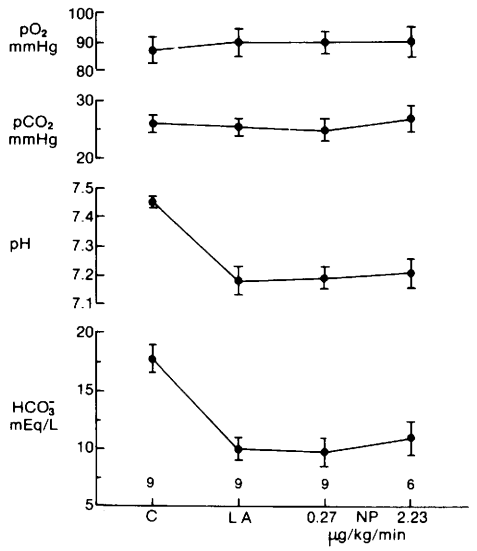

FIG. 2. Arterial blood gas and acid-base response to controlled arterial hemorrhage and sodium nitroprusside infusion.

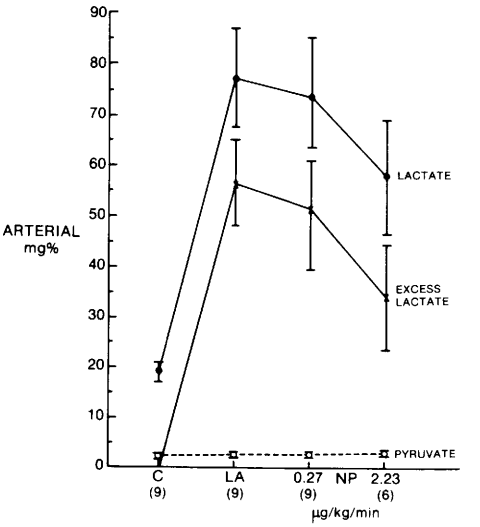

FIG. 3. Arterial lactate, pyruvate and excess lactate concentrations after controlled arterial hemorrhage and sodium nitroprusside infusion.

ml/min during the lactic acidosis period to 2.83 ± 0.87 and 1.92 ± 0.47 mmHg/ml/min during the low and high dose sodium nitroprusside periods, respectively. Total peripheral vascular resistance was unchanged from the lactic acidosis period level (29.4 ± 3.6 mmHg/l/min) by low dose sodium nitroprusside (30.4 ± 5.0 mmHg/l/min) but was decreased to 20.0 ± 2.2 mmHg/l/min by high dose sodium nitroprusside.

Figure 2 illustrates arterial blood gas and acid-base data. Arterial pO_2 and pCO_2 were relatively constant throughout the duration of the experiment at approximately 90 mmHg and 26 mmHg, respectively. Controlled arterial hemorrhage resulted in a sharp decrease in arterial pH from 7.45 ± 0.02 during the control period of 7.18 ± 0.05 during the lactic acidosis period; there was little effect of either dose of sodium nitroprusside on arterial pH. Arterial bicarbonate concentration decreased from 17.8 ± 1.2 mEq/l during the control period to 9.9 ± 1.1 mEq/l during the lactic bicarbonate concentration and the slight increase to 11.1 ± 1.3 mEq/l during high dose sodium nitroprusside was not significant.

Figure 3 illustrates the blood lactate and pyruvate data. Central venous lactate and pyruvate concentrations were not significantly different from arterial values. Neither controlled arterial hemorrhage nor low or high dose sodium nitroprusside had any effect on arterial pyruvate concentrations. Arterial lactate concentration increased fourfold during controlled arterial hemorrhage from 18.9 ± 1.9 mg% during the control period to 77.2 ± 0.7 mg% during the lactic acidosis period. Low dose sodium nitroprusside resulted in little change in arterial lactate concentration while high dose sodium nitroprusside resulted in a fall in arterial lactate concentration to 57.8 ± 11.6 mg%; this was not a statistically significant decrease and was still three times the control period value. Since arterial pyruvate concentration was unchanged, arterial excess lactate concentrations tended to parallel arterial lactate concentrations. Arterial excess lactate concentration fell from 56.5 ± 8.8 mg% during the lactic acidosis period to 34.0 ± 10.6 mg% during the high dose sodium nitroprusside period; this decrease was not statistically significant and high levels of arterial excess lactate remained. The lactate to pyruvate ratio was 8.2 ± 0.8 during the control period, 31.4 ± 3.3 during the lactic acidosis period, 25.2 ± 2.9 and 20.2 ± 2.6 during the low and high dose sodium nitroprusside periods, respectively.

Discussion. We have developed an animal

model of lactic acidosis as reflected by a metabolic acidosis with an increase in arterial lactate concentration, increase in lactate to pyruvate ratio and formation of excess lactate. This was achieved with controlled arterial hemorrhage which resulted in sustained reductions of approximately 50% in mean arterial pressure, cardiac output and femoral artery flow. These results are in agreement with the studies of Nahas *et al.* (8). They subjected dogs to controlled arterial hemorrhage and noted that arterial pH (7.26–7.31), arterial bicarbonate concentrations (6.3–10.6 mEq/l) and arterial excess lactate concentrations (56.6–62.7 mg%) were stable for a period of 120 min beginning 30–60 min after the end of the hemorrhage. Similar observations were made by Seligman *et al.* (9). Therefore, this experimental technique results in a stable steady-state of lactic acidosis during which the effect of various interventions can be evaluated. The nitroprusside was administered during the time of predicted stable steady-state lactic acidosis as demonstrated by the studies of Nahas *et al.* (8) and Seligman *et al.* (9). Further evidence that the experimentally produced lactic acidosis was in a stable steady state at the time of the nitroprusside infusions derives from the fact that the values for arterial pyruvate, lactate and excess lactate concentrations in the two samples taken 10 minutes apart in the lactic acidosis period differed by less than 5%. During low and high dose sodium nitroprusside infusion, there were progressive small decreases in mean arterial pressure and increases in femoral artery flow which were accompanied by progressive decreases in both femoral and total peripheral vascular resistance. These observations are consistent with the known vasoactive actions of sodium nitroprusside (10) and support the view that the doses of sodium nitroprusside used in this study were effective vasodilator doses. Despite this no statistically significant or clinically important changes in arterial acid-base status or lactate/pyruvate metabolism were observed.

Therapeutic interventions short of altering the underlying or associated disease have not proved beneficial in lactic acidosis. It is of some interest that Taradash and Jacobson (3) were able to reverse lactic acidosis using sodium nitroprusside vasodilator therapy. Their patient presented in congestive heart failure with lactic acidosis and normal blood pressure. Sodium bicarbonate therapy led to pulmonary edema. Sodium nitroprusside infusion at 0.6 μg/kg/min resulted in a decrease in pulmonary artery wedge pressure from 28 mmHg to 12 mmHg, a decrease in arterial pressure from 140/50 mmHg to 120/50 mmHg, a decrease in systemic vascular resistance from 1030 dyne·sec·cm^{-5} to 920 dyne·sec·cm^{-5} and an increase in cardiac output from 5.5 l/min to 6.0 l/min. The improvement in cardiac performance, as reflected by the increase in cardiac output and stroke-work index with a drop in left-ventricular filling pressure (i.e. pulmonary artery wedge pressure), is the usual response of a poorly functioning left ventricle to sodium nitroprusside (10). Significant lactic acidosis is known to occur in severe pulmonary edema due to acute congestive heart failure; furthermore, as the pulmonary edema regresses, the lactic acidosis is completely corrected without the administration of sodium bicarbonate (11). Therefore, it is likely that the beneficial effect of sodium nitroprusside on lactic acidosis noted by Taradash and Jacobson related to the improvement in myocardial performance with resultant improvement in tissue perfusion rather than any specific effect of sodium nitroprusside on lactate/pyruvate metabolism.

Summary. Lactic acidosis was produced in the dog by controlled arterial hemorrhage to a sustained mean arterial pressure of 50 mmHg. A stable metabolic acidosis with increased arterial lactate concentration, increased arterial lactate to pyruvate ratio and increased excess lactate occurred. Intravenous sodium nitroprusside at both low (0.27 μg/kg/min) and high (2.23 μg/kg/min) doses produced regional and systemic vasodilatation but failed to produce a statistically significant or clinically important change in arterial acid-base status or lactate/pyruvate metabolism. It is concluded that stable lactic acidosis can be produced in dogs by graded arterial hemorrhage with sustained hypotension. Vasodilator therapy with sodium nitroprusside is without significant effect on lactic acidosis associated with hypotension in the dog.

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