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Effects of Local Acidosis on Vascular Resistance in Dog Skeletal Muscle (37791)

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Acidosis from moderate to severe levels occurs regularly during normal as well as pathological situations (1). For example, partial occlusion of a supplying artery, or an increase in metabolic rate are frequently accompanied by a moderate decrease in plasma pH (1). Also, pH falls to low levels during conditions such as shock. It is generally agreed that the hydrogen ion is vasodilator and this has been documented in several vascular beds. Local acid administration causes a fall in resistance in the dog forelimb (2), hind limb (3, 4), coronary (5), cerebral (6), cutaneous (7), and skeletal muscle (7, 8) vascular beds. However, the effect of steady-state acid infusion was determined in only one of the latter studies (8). The current study was undertaken because of the relatively small amount of information available about this vasculature, which composes approximately half of the body mass and is capable of demanding a large portion of the cardiac output. Therefore, the primary objective of this study was to determine the effects on skeletal muscle vascular resistance of steady-state decreases in blood pH over a wide range of values.

Methods. Experiments were completed in 17 mongrel dogs of both sexes, anesthetized with sodium pentobarbital (30 mg/kg) and anticoagulated with heparin sodium (3 mg/kg). The innervated right gracilis muscle was isolated from the body and skin by appropriately placed ligatures and by cauterization. Drying was prevented by bathing the muscle with warm mineral oil and covering it with Saran Wrap. Muscle temperature was maintained at 37° with a heat lamp. A sigmamotor pump was interposed between the right femoral and gracilis arteries for constant flow perfusion of the muscle. Venous blood was returned to the animal through the uninterrupted gracilis vein. All branches of the gracilis muscle and vein were ligated except for one side branch of the vein which was cannulated for withdrawing blood samples. Completeness of vascular isolation was confirmed by stopping the perfusion pump and observing the perfusion pressure tracing. Blood pH was determined with a Radiometer pH meter. Femoral artery and gracilis artery (perfusion) pressures were recorded by means of Statham pressure transducers and a Sanborn direct writing recorder. Since blood flow was held constant, changes in perfusion pressure were directly proportional to changes in muscle vascular resistance. All dogs were ventilated with a Harvard positive pressure respirator.

Isotonic solutions of lactic or acetic acid were infused upstream to the blood pump with a Harvard infusion pump at sequentially faster rates (0.1–0.4 ml/min). Infusion at each rate was maintained until perfusion pressure became steady, usually 5–10 min. An isotonic solution of sodium chloride was infused at the same rate in each case to evaluate the dilutional effect of the test solution. Systemic pH of the dog was not effected at the infusion rates employed. The order of infusion of the three solutions was randomized.

Since changing pH of the blood is known to alter red cell size and hence viscosity of the blood (8), the effects on hematocrit of decreasing pH over the range used in this study were determined in several experiments. Blood samples were obtained proximal

and distal to the point of acid infusion, and from the gracilis vein before, during, and after acid infusion. In several dogs the experimental muscle and intact contralateral control muscle were weighed and compared at the end of the experiment as a rough check for possible edema formation; no differences were found.

Statistical comparisons of the data were made using Student's t test for a comparison of the mean and Student's t test modified for within group comparisons. P values below 0.05 were considered significant.

Results. Figure 1 illustrates graphically data obtained during lactic or acetic acid infusion on perfusion pressure, aortic pressure, and plasma pH. Decreasing pH stepwise from 7.43 to 7.38, 7.32, and 7.17 pH units ($p < 0.05$) was associated with falls in perfusion pressure averaging 2%, 14%, and 18%, respectively ($p < 0.05$). Blood pH and perfusion pressure returned to a level not different than control shortly after stopping lactic acid infusion ($p > 0.05$). The dashed lines indicate the response to acetic acid infusion. As in the case of lactic acid, decreasing pH with acetic acid in a step-wise fashion resulted in step-wise decreases of pH, however the decreases in perfusion pressure were of significantly greater magnitude than seen during lactic acid infusion ($p < 0.05$).

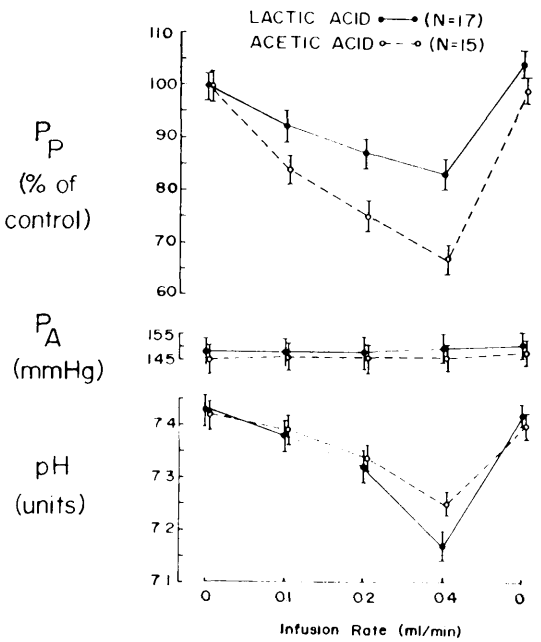

FIG. 1. Average effects ± standard errors of intra-arterial infusion of lactic and acetic acid on gracilis muscle perfusion pressure (P_P), dog aortic blood pressure (P_A), and muscle venous blood pH. Average muscle flow; lactic acid = 16.5 ml/min per 100 g; acetic acid = 18.9 ml/min per 100 g. Initial perfusion pressure: lactic acid = 152 ± 6 mmHg; acetic acid = 147 ± 5 mmHg.

Figure 2 shows representative tracings from one experiment in which lactic acid, acetic acid, and saline were infused to lower pH. In this example, infusion rates were increased to 2 ml/min. Note that with lactic and acetic acid there were step-wise decreases in perfusion pressure, reflecting decreases in resistance, until the infusion rate exceeded 0.4 ml/min. Further increasing the infusion rate, and decreasing pH to near or below 7.0 units, resulted in no change or a striking increase in perfusion pressure, indicating an increase in resistance. This increase in perfusion pressure at extremely low pH's usually occurred during high infusion rates with both lactic and acetic acid. Stopping the acid infusion was usually followed by a rapid decrease in perfusion pressure below the lowest level attained previously. Equivolume infusion of saline, shown on the lower portion of the figure, did not alter pH and did not affect perfusion pressure except at the highest infusion rate on the average, and over the infusion rates used to induce moderate decreases in pH, saline infusion produced no significant change in perfusion pressure. The representative tracings have been cut to uniform size for ease of presentation. Each complete run (from zero acid infusion to maximum infusion rate to the recovery period) required approximately 25 min.

Hematocrit data during pH changes are not shown here, however, in no case was a significant change in hematocrit noted during acid infusion over the range shown in Fig. 1 ($p > 0.05$). It is also of interest that hematocrit did not increase significantly ($p > 0.05$) during the abrupt pressure increase when pH was decreased to near or below 7 pH units.

Discussion. Step-wise decreases in pH over the physiological range produced by infusion of lactic or acetic acid were associated on

the average with step-wise decreases in skeletal muscle vascular resistance during the steady-state. Acetic acid caused a substantially greater decrement in resistance than did lactic acid. This difference is no doubt due to the vasodilator property of the acetate ion (9) which enhances the vasodilation due to the H^+. The resistance decreases appeared to result from active vasodilation since they occurred at constant blood flow and during decreases in transmural pressure. These data agree with studies in which blood pH was decreased locally by acid infusion in other vascular beds (1–8; see introduction) and in skeletal muscle (7, 8). Kontos *et al.* (8), lowered pH by intraarterial infusion of a buffered HCl solution and found a linear relationship between perfusion pressure and venous blood P_{CO_2} and pH in the constant flow perfused, dog gastrocnemius muscle.

The striking increase in perfusion which frequently occurred at extremely low pH's and the subsequent fall in perfusion pressure immediately after stopping the acid infusion is not readily explainable. Since the hematocrit did not usually rise, and frequently red cells were lysed, vasoconstrictor substances liberated from blood cells may be responsible. A clumping phenomenon may also be responsible. This latter explanation is more tenable since the rapid fall in perfusion pressure which regularly occurred immediately after stopping acid infusion suggests that the vessels might have been well dilated during the period perfusion pressure was elevated.

It has been proposed for years that changes in hydrogen ion concentration may account for the increase in blood flow which occurs during exercise or following a period of ischemia. The difficulty in assigning a major role to the hydrogen ion in these phenomena has been that relatively large changes in pH are required to produce changes in resistance of appropriate magnitude (1). However, the recent work of Kontos *et al.* (8), suggest that approximately 65% of the fall in vascular resistance occurring during ischemia can be accounted for by changes in

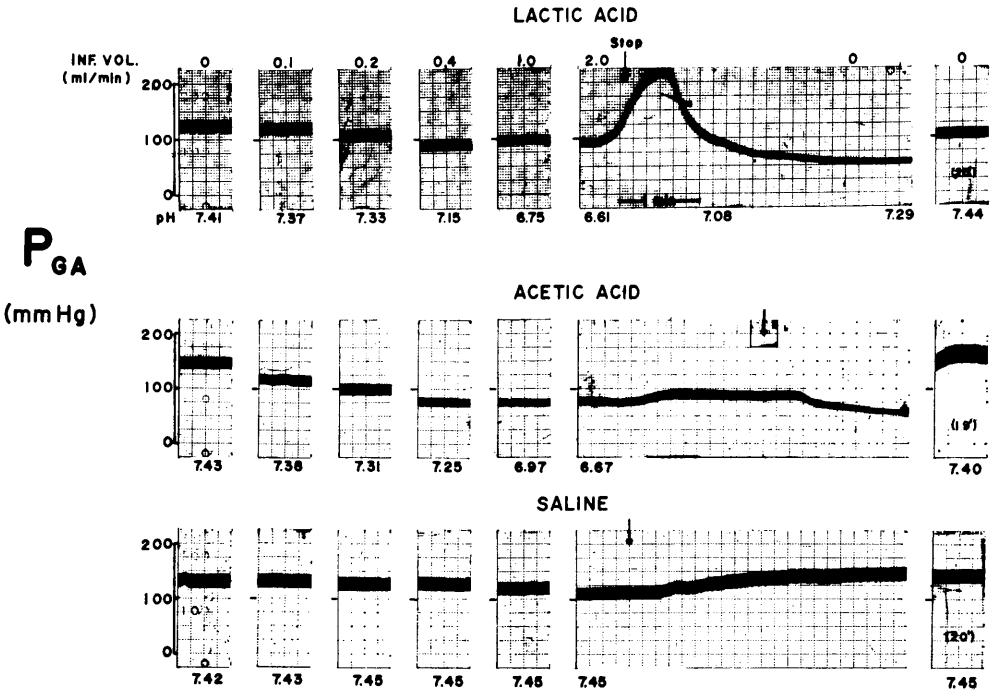

FIG. 2. Representative tracings showing the effects of intraarterial infusion of various acids on gracilis artery perfusion pressure (P_{GA}) and gracilis venous pH in a constant flow perfused gracilis muscle. Values in parenthesis indicate number of min after stopping solution infusion. INF. VOL. = infusion volume of solutions.

pH or P_{CO_2}. However, it should be pointed out that a comparison between the present study and the one of Kontos *et al.* (8) is difficult to make because the experimental conditions were not the same and different skeletal muscle beds were studied. Earlier studies by other investigators do not support the hypothesis that the H^+ is the major factor responsible for the large fall in resistance which occurs during active or reactive hyperemia. For example, blood flow has been noted to rise 100% to 1000% in both dog and human muscle with only a small decrease in venous blood pH (0.03–0.1 unit) during active or reactive hyperemia (1). Examples are the studies of Rudko and Haddy (10), who measured an increase in dog skeletal muscle blood flow of 270% of control with a fall in muscle venous pH of only 0.07 unit, and Ross *et al.* (11), who noted a rise in dog gastrocnemius muscle blood flow of 173% of control during muscle contraction which was associated with a fall in venous blood pH of 0.05 unit.

Data in the current study lend support to the concept that the hydrogen ion is not involved to a major extent, or at least is not the major factor responsible for the increase in flow seen during an increase in muscle metabolism or during ischemic situations. Recall that a decrease in pH by 0.05 unit by lactic acid infusion was associated with a decrease in perfusion pressure and hence vascular resistance of 2% and further decreasing pH by 0.26 unit decreased perfusion pressure by only 18%. Therefore, considering the relatively small changes in pH observed during situations of active or reactive hyperemia, and the large increases in flow and decreases in resistances, it is suggested that hydrogen ion *per se* is involved, but that its effect appears to be too small to account for the substantial decrease in resistance characteristics of active or reactive hyperemia in skeletal muscle.

Summary. Experiments were completed in collateral free, constant flow perfused dog gracilis muscles to determine the effects of local increases in H^+ on skeletal muscle vascular resistance. Blood pH was decreased step-wise by intraarterial infusions of isotonic lactic or acetic acid solutions. A decrease in pH with either of these acids was associated with a step-wise decrease in muscle vascular resistance. While it was clearly demonstrated that hydrogen ion is vasodilator in skeletal muscle, the H^+ effect appears to be too small to account for the substantial decrease in resistance characteristic of active or reactive hyperemia.

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