Influence of Glucose, Insulin, and Potassium on Vulnerability to Ventricular Fibrillation in the Canine Heart

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SUMMARY We studied the influence of glucose (G), insulin (I) and potassium (K+) on vulnerability to ventricular fibrillation (VF) of the nonischemic canine myocardium. Vulnerability was assessed by determining VF and repetitive extrasystole (RE) thresholds with a single stimulus applied to the right ventricular endocardium during the vulnerable period. Electrical testing of the heart was performed before and after 1 and 2 hours of infusing G (10 mg/kg per min) and I (0.025 U/kg per min) with and without K+. Infusion of glucose, insulin, and potassium (GIK) in 11 dogs significantly increased both VF (22%) and RE (33%) thresholds within the 1st hour and only the VF threshold (64%) in the 2nd hour. No significant changes in serum K+ concentration occurred. Spontaneous termination of VF was observed in six dogs during GIK infusion. Glucose and insulin infusion increased both VF and RE thresholds within the 1st (17% and 19%) and 2nd hours (43% and 43%). This occurred despite substantial reductions in serum K+ concentration. Lowering serum K+ by hemodialysis in six dogs decreased both VF and RE thresholds within the 1st (33% and 32%) and 2nd hours (35% and 14%). Restoration of the serum K+ concentration by KCl infusion while maintaining dialysis returned the thresholds to control values. In six dogs, insulin infusion during low K+ dialysis increased the VF and RE thresholds despite a further reduction in serum K+ concentration. We concluded that insulin exerts a protective effect against vulnerability to VF in the normal canine heart. This salutary action is most marked when the drug’s hypokalemic effect is prevented by concomitant K+ infusion.

RECENT interest in so-called polarizing solutions has centered mainly around the possibility of salvaging ischemic myocardium during the inception of acute myocardial infarction. The preceding decade witnessed the introduction of polarizing solutions for the prevention of ventricular arrhythmias and other electrophysiological sequellae of acute myocardial ischemia. It was reasoned that glucose, insulin, and potassium (GIK) would restore intracellular potassium, extruded by the ischemic lesion from myocardial cells into the extracellular compartment. Since potassium is a key ion in membrane polarization, it was assumed that the electrophysiological effects of GIK were mediated by changes in potassium metabolism.

However, the influence of the specific components of polarizing solutions on cardiac electrical properties has not been clearly defined. The present study was undertaken to examine the effect of glucose and insulin with and without potassium supplement on vulnerability to ventricular fibrillation (VF) in the nonischemic canine myocardium. In addition, the effects of hypokalemia on ventricular vulnerability produced by glucose and insulin infusion were compared to those observed during hypokalemia produced by hemodialysis. Thus, the direct effects of glucose and insulin on vulnerability to fibrillation could be separated from those related to alterations in serum potassium levels.

Methods

Mongrel dogs of either sex were anesthetized with alpha-chloralose, 100 mg/kg, and intubated. Artificial ventilation was carried out with a Harvard pump using an O2 supplement to maintain the arterial Po2 at 80–150 mm Hg and arterial pH, PCO2 in the physiological range. Blood pressure was maintained at a mean of 100–120 mm Hg by infusion of saline or donor blood in dogs undergoing hemodialysis. Two bipolar catheters were introduced through the jugular veins and positioned under fluoroscopic control in the apex of the right ventricle. One catheter (USCI, 6F) was used to record the intracavitary ECG and the other (Medtronic 6901) was employed to pace the right ventricle (180 beats/min) and for electrical testing. This catheter was positioned with the distal cathodal electrode lodged in the trabeculae. Heart rate was maintained constant by ventricular pacing throughout the experiment. The pacing stimulus was a rectangular pulse 2 msec in duration and with its amplitude adjusted to ensure consistent capture.

The test stimulus was delivered through the same pacing catheter; appropriate circuitry was incorporated to inhibit the output of the pacemaker for a predetermined and adjustable period following the test pulse. A single stimulus method was used to
determine ventricular fibrillation threshold (VFT) and threshold for repetitive extrasystoles (RET). The test stimulus consisted of rectangular pulse of either 2- or 5-msec duration which was delivered after every 10–15 paced beats. Testing was initiated with a 2-msec stimulus. If no repetitive extrasystoles were evoked at 60 mA or higher, the stimulus duration was changed to 5 msec. The duration was then kept constant for the rest of the experiment. The amplitude and configuration of the test stimulus were calibrated and periodically verified by means of a Tektronics P6021 AC current probe attached to a 5102N Tektronics oscilloscope. Stimulus intervals monitored with a digital timer arranged in parallel with the circuitry were accurate to within ± 1 msec.

The vulnerable period was scanned in 2- or 5-msec increments (depending on the duration of the test stimulus) for 30 msec from the edge of the refractory period. The intensity of the test stimulus was increased in 2-mA steps and the scanning repeated until two or more extrasystoles were evoked by a single stimulus. The lowest threshold at which this repetitive response was observed in two out of three trials was considered to be the RET. The intensity of the stimulus was then increased further in the same step-wise manner until ventricular fibrillation supervened, thus defining the VFT. Defibrillation was promptly accomplished, usually within 15 sec, by a DC capacitor discharge (100–150 W·sec) delivered through copper plates previously placed across the chest. In some instances, VF was allowed to continue for longer periods to determine the occurrence of spontaneous recovery which was noted during certain interventions.

Four groups of dogs were studied:

**Glucose and Insulin**

**With Potassium (GIK)**

In 11 dogs, 200–300 ml of a solution of glucose, regular insulin, and potassium (GIK) were administered intravenously for a period of 2 hours at the rate of 10 mg/kg per min for glucose, 0.025 U/kg per min for insulin, and 0.025 mEq/kg per min for potassium. Electrical testing was performed in the control state, as well as at 1 and 2 hours after the onset of the infusion. Serum potassium and glucose concentrations were determined at the time of electrical testing.

**Without Potassium**

In this group of eight dogs, glucose and insulin were administered as in the first group but without the addition of potassium. The objective was to determine the effects of these two components of the polarizing solution in the absence of potassium and in the presence of the ensuing hypokalemic state. Electrical testing was performed in the control state and at the end of 1 and 2 hours of the infusion. Serum potassium and glucose were determined at the time of electrical testing. In two dogs, KCl was infused at the end of the experiment for a period of 1 hour (at the rate of 0.025 mEq/kg per min) and testing repeated thereafter.

**Hemodialysis**

Two series of experiments were conducted on 12 dogs subjected to hemodialysis. The objective was to study: (1) the effects of alteration in serum potassium on cardiac vulnerability and (2) the action of insulin in the potassium-depleted and repleted dog. Hemodialysis was performed with a Travenol dialyzer. The bath consisted of 120 liters of isotonic fluid wherein the potassium content was monitored and adjusted according to the conditions of the experiment. The flow rate was maintained at 100–150 ml/min and venous pressure at 20–40 mm Hg. The fluid loss during the procedure was estimated and was replaced by isotonic saline to maintain the hematocrit at levels near the control. The dogs were heparinized during the entire procedure. Blood transfusions from donor dogs were employed to support the blood pressure in dogs developing hypotension at the onset of dialysis (four dogs). In the rest of the dogs, priming was accomplished with isotonic saline.

**Dialysis to Produce Potassium Depletion and Repletion**

In this group of six dogs, dialysis was carried out using a potassium free bath for a period of 2 hours. Potassium then was added to the bath and dialysis continued for a period of 2 hours to restore serum potassium to control levels. Electrical testing was performed every hour during potassium depletion and repletion. Serum sodium, potassium, and glucose were determined at the time of testing. The total amount of potassium that was removed during dialysis was estimated from the bath concentrations at the inception and completion of the 2 hours of the depleting dialysis.

**Dialysis Combined with Insulin**

In this group of six dogs, potassium depletion was achieved by dialysis for 2 hours with potassium-free solutions as in the previous group. Electrical testing was carried out at 1 and 2 hours of dialysis. Regular insulin, 10 U/kg, was then given intravenously as a single bolus and repeated two to three times at 5-minute intervals. To avoid hypoglycemia, glucose was added to the dialysis bath to maintain a constant serum concentration of 80 mg/100 ml. Blood glucose levels did not drop below 80 mg% in any of the dogs. Electrical testing was performed at 5-minute intervals for a period of 30 minutes following the administration of insulin. At the end of this period, KCl was added to the dialysis bath to yield a concentration of 6–8 mEq/liter. This restored serum potassium to control levels in all dogs after 1–2 hours of further dialysis. Electrical testing was
Further Observations

The presence or absence of nonsustained ventricular tachycardia (NSVT), defined as more than three successive ventricular responses to one test stimulus, was noted. In addition, the presence or absence of nonsustained ventricular fibrillation (NSVF), defined as self-terminating paroxysm of rapid, irregular, and fractionated succession of ventricular responses to one test stimulus, was also recorded.

Serum sodium, potassium, and glucose were monitored in all dogs. Serum calcium and magnesium were checked in the dialysis group and were found not to vary during the procedure.

Serum osmolarity was determined by freezing point depression (accuracy ± 2 mOsmol/liter).

Since each dog served as its own control, data were analyzed using either Student's *t*-test for paired values or *χ²* analysis with Yate’s correction.

Results

Administration of GIK resulted in an increase in VF threshold in 11 dogs from a control of 45 to 55 mA after 1 hour and 74 mA (*P* < 0.01) after 2 hours of infusion (Fig. 1). Ventricular fibrillation during GIK infusion was preceded in these dogs by episodes of nonsustained ventricular tachycardia (NSVT) or nonsustained ventricular fibrillation (NSVF). Upon eliciting repetitive extrasystoles, increasing the amplitude of the test stimulus resulted in longer runs of self-terminating ventricular tachycardia (NSVT). Whereas in the control state, the maximum number was six repetitive responses to a single test stimulus, during GIK infusion the arrhythmia at times persisted for as long as 20–30 seconds. Six of the dogs exhibited additionally NSVF only during GIK infusion (*P* < 0.05). A further increment in the amplitude of the test stimulus resulted in longer runs of self-terminating ventricular tachycardia (NSVT). Whereas in the control state, the maximum number was six repetitive responses to a single test stimulus, during GIK infusion the arrhythmia at times persisted for as long as 20–30 seconds. Six of the dogs exhibited additionally NSVF only during GIK infusion (*P* < 0.05).

The threshold for inducing repetitive ventricular responses increased during the 1st hour of GIK infusion (*P* < 0.05) and leveled off thereafter, although the VF threshold continued to rise. Plasma potassium and sodium did not change and plasma glucose increased from 104 to 270 mg/100 ml at the end of 2 hours of GIK infusion. No correlation was evident between the magnitude of changes in plasma glucose concentration and the magnitude of changes in the repetitive extrasystole and ventricular fibrillation thresholds.

Infusion of glucose alone in three dogs did not significantly alter the VF threshold at any point during the course of the experiment. Serum osmolality was only slightly increased by 7 ± 2 mOsmol/liter (2.3%) after 2 hours of infusion.

Glucose and Insulin

In this group of eight dogs, infusion of glucose and insulin reduced plasma potassium concentration from a control of 3.3 mEq/liter to 1.9 mEq/liter after 1 hour and to 1.7 mEq/liter after 2 hours of infusion (*P* < 0.001). Comparable changes in coronary sinus blood potassium were noted in three dogs. In spite of the marked lowering in serum potassium level, the VF threshold increased from a control of 35 to 41 mA at 1 hour (not significant) and to 50 mA after 2 hours of infusion (*P* < 0.01), (Fig. 2). NSVT was observed in six of eight dogs after infusion of glucose and insulin and preceded the onset of sustained ventricular fibrillation (*P* < 0.05).

Two dogs were given potassium after infusion of glucose and insulin had been completed. There was a further increase in VFT by 60% in one dog and by 25% in the other. Of note is the fact that NSVF occurred in both of these dogs only after addition of potassium. An example of these changes is presented in Figure 3.
Influence of glucose and insulin infusion on vulnerable period threshold in eight dogs. Glucose and insulin infusion resulted in significant increases in VFT and RET. This occurred despite substantial reductions in serum K⁺ concentration. The abbreviations and symbols are similar to those employed in Figure 1. Values are expressed as means ± SEM. * = P < 0.05 and ** = P < 0.01, respectively, when compared to control.

The increase in RET paralleled the changes in VFT at both 1 and 2 hours of infusion of glucose and insulin. The ratio of RET to VFT was maintained throughout these experiments.

Plasma glucose was elevated to the same extent as in the group receiving GIK. There was no correlation between the magnitude of changes in plasma glucose and those of the electrical indices being tested.

Dialysis

The results from six dogs subjected to hemodialysis are presented in Figure 4. The changes in serum potassium were accompanied by parallel changes in VFT during potassium depletion as well as during repletion. During potassium depletion, the VFT decreased from a control of 40 to 27 mA after 1 hour and to 26 mA after 2 hours of dialysis (P < 0.005). Serum potassium decreased from a control of 3.5 mEq/liter to 2.0 after 1 hour and to 1.9 after 2 hours of dialysis (P < 0.001).

The total potassium loss at 2 hours, determined from the concentration in the dialysate, was 40–80 mEq. A similar amount was restored during the 2-hour dialysis against high potassium bath concentration.

During potassium depletion, ventricular fibrillation once elicited was always sustained and NSVF was not observed. Likewise, NSVT noted during the control state in four dogs did not occur in the depleted state. However NSVT reappeared when the plasma potassium level was restored (Fig. 4). Spontaneous ventricular ectopic beats and couplets
were also noted during potassium depletion induced by hemodialysis.

The changes in RE threshold were parallel to changes in VF threshold but were of lesser magnitude. Thus, the two thresholds were closer together after 2 hours of potassium depletion than in the control or potassium-repleted state.

There were no significant changes in plasma calcium, magnesium, glucose, or sodium during these experiments. The changes in RE and VF threshold reverted to control values when potassium was replenished by hemodialysis. The values after 2 hours were almost identical to the control levels.

Dialysis and Insulin

The data from the six dogs subjected to hemodialysis and then given insulin are presented in Figure 5. In these experiments, potassium depletion by hemodialysis for 2 hours resulted in a reduction of potassium from a control of 3.4 to 2.2 mEq/liter ($P < 0.001$). There was an associated decrease in VF threshold from 36 to 16 mA ($P < 0.001$). After administration of insulin and while dialysis was maintained, plasma potassium concentration was further reduced to 2.0 mEq/liter ($P < 0.01$). However, VF threshold increased significantly to 30 mA ($P < 0.001$). This increase was noted as early as 10 minutes after administration of insulin and was most marked at 20-30 minutes. The value for the VF threshold reported above represents the average of the last two determinations at 20-30 minutes after insulin administration. When serum potassium was raised to control levels by dialysis against high potassium concentration, the VF threshold increased further to 50 mA, a value that is significantly higher than the control ($P < 0.05$) or the post-insulin level ($P < 0.01$). There were no changes in serum calcium, magnesium, sodium, or glucose in these experiments.

NSVT noted during the control period disappeared during hypokalemia and reappeared after insulin administration. When potassium was replenished, both NSVT and NSVF were elicited as in the group receiving GIK. RE threshold decreased during potassium depletion, but the lowering was of a lesser magnitude than the reduction in VF threshold. After either insulin or potassium administration, RE threshold was increased, but the increases were less marked than those of the VF threshold.

Discussion

Despite considerable interest in polarizing solutions, there have been few systematic studies of the effects of this therapeutic modality or its components on VFT in either the nonischemic or ischemic myocardium. Shinohara found a significant increase in VF threshold on coronary occlusion compared to control animals. However, because the polarizing solution was infused 1-2 hours after coronary occlusion, the early period of electrical instability was not studied. Cherbakoff et al. infused 400 ml of 50% dextrose in water + 70-200 U regular insulin over a period of 2 hours in dogs subjected to acute ligation of the left circumflex coronary artery. Potassium concentration was markedly reduced in the coronary sinus.
Ventricular arrhythmias were prevented and the onset of VF was delayed. This protective effect of GIK was ascribed to reduced potassium efflux from ischemic myocardial cells. In these experiments serum potassium concentration in the systemic circulation also was reduced. Since coronary sinus sampling was not restricted to the ischemic zone, changes in coronary sinus venous concentration may have reflected the generalized systemic and myocardial potassium concentration rather than the contents of effluent draining ischemic myocardium. Hiatt and coworkers using large doses of insulin demonstrated a protective effect against ventricular arrhythmias in dogs subjected to circumflex coronary artery ligation. The insulin doses were much higher than those used in polarizing solutions. To safeguard against hypoglycemia, 10% dextrose in water was infused. Serum potassium concentration was not reported, but it must have been reduced significantly because no potassium supplement was administered. They ascribed the protective effects against ventricular arrhythmias to insulin blocking the action of catecholamines.

This possible mechanism was deduced from prior investigations. GIK has been employed to treat patients with acute myocardial infarction and those with congestive heart failure. These clinical studies differ in many respects, including the combination of components, the routes of administration, the duration of therapy, the time after onset of infarction, and the use of ancillary measures. They therefore permit no certain conclusions as to the antiarrhythmic or antifibrillatory effects of GIK.

Our data demonstrate a protective effect of GIK on vulnerability to VF in the nonischemic dog myocardium. The protection also is manifest by an increase in the threshold for repetitive ventricular responses to a single test stimulus, as well as "containment" of the induced arrhythmia, i.e., its nondegeneration to VF or the reversion of VF to sinus rhythm. Such spontaneous reversion in the dog is rare, being observed only once by Wiggers in 400 dogs with VF. Zipes similarly found it to be an uncommon event in over 200 hours of VF in dogs in which the circulation was supported by cardiopulmonary bypass. Its occurrence in 6 of 11 dogs treated with GIK, and the observation of NSVF only under similar conditions in the other groups in this study, indicate therefore that reversal of the fibrillatory process was indeed the result of the experimental intervention. The present study does not provide information as to the possible electrophysiological mechanism for NSVF.

When glucose and insulin were infused in the absence of a potassium supplement, VFT was significantly elevated, although the serum potassium concentration was markedly reduced. This observation, as well as the fact that coronary flow was unimpaired, indicates that a protective effect against VF was exerted by the administered glucose and insulin. Thus, whereas our findings lend support to the observation of Cherbakoff et al. concerning the antiarrhythmic effects of glucose and insulin, their suggested explanation that the protective action derives from correction of cellular potassium depletion induced by ischemia may not be valid. This is so because the effects of GIK on cardiac vulnerability to fibrillation occur in the absence of ischemia. Pathological changes in the nonischemic myocardial segments during coronary occlusion have been demonstrated by histological and biochemical studies and polarizing solutions may improve these better-perfused segments more than the ischemic ones. To date there has been no direct proof that potassium replenishment of ischemic cells takes place under the influence of glucose and insulin with or without potassium supplement.

Although the effects on the VFT of glucose and insulin alone were similar to those of GIK, several differences were noted. The parallel relationship between RE and VF threshold was maintained during glucose and insulin infusion but was modified when potassium was included in the solutions. NSVF was observed only with GIK, suggesting that potassium exerts an additional protective effect by causing reversion of VF to sinus rhythm. Interestingly, a similar observation was made by Shinohara when GIK was administered in the ischemic setting. The defibrillating properties of potassium have been well recognized but at much higher concentrations than those employed in our study.

When hypokalemia was induced by hemodialysis, there was marked reduction in VFT. The threshold for evoking repetitive extrasystoles also was reduced, but to a lesser extent; at times the two thresholds were similar. Thus elicitation of a repetitive extrasystole resulted in VF at the same or only slightly higher stimulus intensity. Salvos of ventricular tachycardia noted in the control state did not occur during the hypokalemia of dialysis but reappeared with potassium repletion; although NSVF was never observed, and VF always was sustained. It has been demonstrated in the isolated perfused heart that a low potassium concentration in the perfusate leads to VF, especially in the presence of calcium. Restoration of potassium to normal levels reverses this tendency to fibrillate.

It is reasonable to question whether the changes in ventricular vulnerability might be due to hyperoncity of the infusate. This possibility does not seem likely, however, for the following reasons: (1) The GIK solution was infused over a slow rate (2 ml/min) and thus was diluted in a comparatively large intravascular volume. In fact, serum osmolarity did not significantly change during the 2-hour period of infusion. (2) When glucose alone was infused in the absence of insulin, no significant change in vulnerability was observed at any time.
during the infusion. (3) Injection of insulin without infusate, however, did result in a substantial alteration in ventricular vulnerability.

There is evidence to suggest that there may be an independent effect of insulin on myocardial vulnerability to fibrillation. The favorable effects of insulin on myocardial function are documented in nonischemic animal models, dogs depleted of potassium by hemodialysis were given large doses of insulin. In these dogs, further reduction of serum potassium was not possible since the large dialyzing bath volume fixed the concentration at the reduced level. It seems likely that significant shifts of potassium intracellularly also were precluded because of the depleted extracellular state. Nonetheless, when insulin was administered, the VF threshold, already reduced from a control of 35 to 15mA by dialysis, rose significantly to near control values (30 mA). Furthermore salvos of repetitive extrasystoles reappeared without degenerating into VF. Such degeneration was the case in the depleted dog without the insulin supplement. Protection of the myocardium by insulin against fractionation and degeneration of a reentrant wave front occurred even in the presence of severe potassium deficiency. Its mode of action must have been other than by augmenting intracellular potassium stores. The effect may be related to mass action effect upon beta-receptor stimulation upon vulnerability of the canine ventricle. Circulation 52: 578-585, 1975.

To determine more precisely the mechanism of insulin's effect on cardiac vulnerability in our nonischemic animal models, dogs depleted of potassium by hemodialysis were given large doses of insulin. In these dogs, further reduction of serum potassium was not possible since the large dialyzing bath volume fixed the concentration at the reduced level. It seems likely that significant shifts of potassium intracellularly also were precluded because of the depleted extracellular state. Nonetheless, when insulin was administered, the VF threshold, already reduced from a control of 35 to 15mA by dialysis, rose significantly to near control values (30 mA). Furthermore salvos of repetitive extrasystoles reappeared without degenerating into VF. Such degeneration was the case in the depleted dog without the insulin supplement. Protection of the myocardium by insulin against fractionation and degeneration of a reentrant wave front occurred even in the presence of severe potassium deficiency. Its mode of action must have been other than by augmenting intracellular potassium stores. The effect may be related to mass action effect upon beta-receptor blockade antagonism or cyclic adenosine 3',5'-monophosphate by insulin. When normokalemia was restored in the presence of insulin, the ventricular fibrillation threshold was elevated further and the effects were identical to those of the GIK infus.

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References


Experimental Evaluation of Streamline Patterns and Separated Flows in a Series of Branching Vessels with Implications for Atherosclerosis and Thrombosis

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SUMMARY Flow conditions in four models representing the aortic bifurcation, iliac bifurcation, and a renal artery branch were investigated at volumetric flow rates corresponding to Reynolds numbers from 1000 to 4000 over the complete range of flow division between daughter vessels. Qualitative flow streamline patterns and quantitative definition of those flow conditions leading to disturbed flow (flow separation) were determined primarily at steady flow with a limited set of pulsatile experiments. Under conditions of no flow separation, common characteristic streamline patterns not parallel to the center lines of parent or daughter tubes were found for all models. These effects were accentuated with increasing Reynolds number. Flow separation was inducible through alteration of flow division between daughter vessels or by an increase in flow rate. Each of the four models had distinct combinations of flow division ratio and flow rate which gave: (1) no flow separation, (2) flow separation at the outside of the right daughter tube, and (3) flow separation at the outside of the left daughter tube. Models representing the renal artery also had regions of simultaneous left- and right-hand separation on the outside of their daughter tubes. The separated flows observed here displayed streamlines forming an open vortex with flows entering and leaving. These regions, which occur only at distinct combinations of flow rate and flow division, may be key centers where platelet aggregates may form, release constituents, and cause vessel injury.
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