J Mol Cell Cardiol 26, 1349–1356 (1994)

Prevention of Extracellular K^+ Inhomogeneity Across the Ischemic Border by Coronary Venous Obstruction in the Dog: Salutary Antiarrhythmic Effects of Enhanced Myocardial Hydration

Alexandros C. Kralios, Michael T. Leonard, Fred L. Anderson and Fany A. Kralios

Department of Veterans Affairs Medical Center, Cardiology Section and Departments of Internal Medicine and Pediatrics, School of Medicine and Nora Eccles Cardiovascular Research and Training Institute, University of Utah, Salt Lake City, UT, USA

(Received 11 August 1993, accepted in revised form 15 April 1994)

A. C. KRALIOS, M. T. LEONARD, F. L. ANDERSON AND F. A. KRALIOS. Prevention of Extracellular K^+ Inhomogeneity Across the Ischemic Border by Coronary Venous Obstruction in the Dog: Salutary Antiarrhythmic Effects of Enhanced Myocardial Hydration. *Journal of Molecular and Cellular Cardiology* (1994) **26**, 1349–1356. Partial coronary sinus obstruction (CSO) in the dog prevents or delays the predictable ventricular fibrillation (VF) of the early phase of acute ischemia, by normalizing regional electrophysiological disparities which presumably reflect inhomogeneous extracellular potassium ($[K^+]_o$) accumulation. To clarify whether CSO indeed affects $[K^+]_o$ inhomogeneity, we determined in 12 chloralose anesthetized dogs the dynamic $[K^+]_o$ changes occurring early during reversible coronary artery occlusion (CAO) involving the mid-left anterior descending branch. These changes were compared to those observed during CAO preceded by CSO sufficient to increase the coronary sinus pressure to 40 mmHg. $[K^+]_o$ was determined using valinomycin coated electrodes implanted within the ischemic (IZ) and the normal (NZ) zones, as well as immediately inside (BZi) and outside (BZo) the visible border. $[K^+]_o$ increased rapidly within the IZ and the BZi reaching plateau 5 min after CAO, at about three-fold control (11.89 ± 1.12 mEq/l). Unexpectedly, $[K^+]_o$ also increased initially outside the border (BZo) but declined after 3 min to a lower level (7.00 ± 0.40 mEq/l), thus creating a steep gradient of up to 5.54 ± 0.20 mEq/l, $P<0.001$) across the visible border. In four trials, the gradient coincided with VF. With CSO preceding CAO, the development of this border zone gradient was entirely prevented. Moreover, $[K^+]_o$ reached a significantly lower and similar level in the IZ, BZi and BZo (9.5 ± 0.89 mEq/l, $P<0.001$) and no VF was observed. Thus the beneficial electrophysiologic and antiarrhythmic effects of CSO in acute ischemia may be explained by $[K^+]_o$ equalization. Prevention of "patchy" $[K^+]_o$ sequestration probably reflects higher dilution, facilitation of diffusion and possibly enhanced lymphatic washout of extracellular solutes mediated through enhanced tissue hydration.

KEY WORDS: Acute ischemia; Ventricular fibrillation; Extracellular water; Edema.

Please address all correspondence to: A. C. Kralios, VA Medical Service, Cardiology Service (111A), 500 Foothill Blvd., Salt Lake City, Utah 84148, USA.

Introduction

Reentrant lethal arrhythmias in the early (Ia) phase of acute ischemia stem from disparate electrophysiologic conditions, developing soon after interruption of myocardial perfusion (Kaplinsky *et al.*, 1979). These conditions were originally linked to elevated extracellular potassium ion concentrations $[K^+]_o$ by Harris *et al.* (1954). Since then the role of $[K^+]_o$ as a contributor to the "arrhythmogenic substrate" has been extensively addressed (Harris, 1966; Hill and Gettes, 1980; Hirche *et al.*,1980; Weiss and Shine, 1981, 1982; Kleber, 1983; David *et al.*, 1988; Pelleg *et al.*, 1989; Knopf *et al.*, 1990; Hariman *et al.*, 1993; Yan *et al.*, 1993). With the development of implantable, K^+ sensitive electrodes (Hill *et al.*, 1978) the concept of disparity of refractoriness, central to the "wavelet" hypothesis of Moe (1962) for reentrant arrhythmias, was considerably strengthened.

$[K^+]_o$ accumulates over time during ischemia exhibiting regional "patchy" sequestration that results in steep gradients within the ischemic and between ischemic and normally perfused regions (Hill and Gettes, 1980; David *et al.*, 1988; Pelleg *et al.*, 1989). Whereas in ischemia the $[K^+]_o$ changes coincide with increased arrhythmia incidence, extraneous K^+ infusions cause significant homogeneous electrophysiological effects but no lethal arrhythmias. This suggests that the key to arrhythmogenesis is not simply the elevation of $[K^+]_o$, but its inhomogeneous "patchy" sequestration which is absent when aqueous perfusates are used. Steep gradients of highly diffusible ions such as K^+, during ischemia indicate discontinuity of extracellular space and formation of diffusion barriers which may simply be indicative of deficient tissue hydration.

We have recently demonstrated that coronary sinus obstruction (CSO) prevents or delays the early VF of acute ischemia (Kralios *et al.*, 1993). We have also found that CSO abolishes the electrophysiological inhomogeneity, appearing soon after coronary flow interruption and commonly attributed to inhomogeneous $[K^+]_o$ accumulation (Kralios *et al.*, 1991).

The purpose of this study was to test the hypothesis that CSO produces its beneficial electrophysiologic/antiarrhythmic effects by abolishing the arrythmogenic $[K^+]_o$ inhomogeneity. The results confirm that CSO exerts a strong "homogenizing" effect on the disparate extracellular space of ischemia, presumably through higher dilution, diffusion and lymphatic washout of arrhythmogenic solutes. They also demonstrate that prevention of $[K^+]_o$ inhomogeneity by physiological manipulations, prevents VF and points to deficient tissue hydration as a basic condition for arrhythmogenesis.

Methods

Twelve fasting mongrel dogs weighing 23.3 to 27.2 kg were anesthetized with chloralose (60 to 100 mg/kg of body weight) and ventilated with a mixture of 97% oxygen and 3% carbon dioxide using a volume respirator. The heart was exposed and suspended in a pericardial cradle via a mid sternotomy, performed under monitoring of systemic arterial pressure, ECG and acid-base balance as described previously (Kralios *et al.*, 1988, 1993). An umbilical tape-in-silicone elastomer snare was placed around the left anterior descending artery, just distal to its first diagonal branch for reversible coronary artery occlusion (CAO). A trial 30-s occlusion was performed to visualize the ischemic border and decide the appropriate placement of K^+ sensitive electrodes.

In this study, we used commercially available pairs of flexible miniature teflon-coated wire K^+ sensitive and reference electrodes (Biosensors Inc., Flanders, NJ, USA) prepared according to the technique described by Hill and Gettes (1978). These electrodes employ a highly selective valinomycin membrane, and display Nernstian response to K^+ in a wide pH range at body temperature. Each pair can be inserted in the desired region and depth of myocardium through an 18 gauge hypodermic needle with minimal trauma. After being hydrated for 24 h, the electrodes were tested for linearity using 3, 5 and 10 mEq/l $[K^+]_o$ solutions of KCl adjusted with NaCl for total ionic strength of 0.150 M at 37°C. The depth of electrode placement was set at 5 mm for all electrodes. This was accomplished by sliding a piece of silicone elastomer tubing over the hypodermic needle so that its tip could not advance further than 5 mm for the epicardial surface.

The electrodes were connected to a four channel, high impedance differential amplifier (Biosensor, Inc. Model DAPF-4) and the output of each channel was connected to the input of Gould DC amplifiers. Calibrated signals were recorded on a strip chart recorder (model 2000 ES Gould Inc.) and to a Macintosh II-fx computer via a MacLab head stage and Chart application. Calibration data were plotted semilogarithmically and only those electrodes with a slope of approximately 60 mV per log K^+ activity and rapid response were inserted. Four pairs of electrodes were used in each experiment. Two were placed 5 mm apart across the visible border, i.e.

immediately inside (BZi) and outside (BZo) the border zone of ischemia. The other two were placed 15 mm away from each of the previous, one towards the center of the ischemic zone (IZ) and the other in the normal zone (NZ) so that all electrodes were aligned perpendicular to the border. Six of the original 48 electrode pairs were replaced during data collection. Four pairs were replaced in three animals with VF during CAO because they were inadvertently pulled out during resuscitation. In addition, another two pairs were replaced because of erratic signals. Three of the pairs were located in the IZ, two in the BZi and one in the NZ.

After heparinization, the coronary sinus was canulated and its outlet occluded around the cannula tip with a silk suture as described in detail previously (Kralios *et al.*, 1988). The distal end of the cannula was connected to the right atrial appendage for continuous drainage. The mean coronary sinus pressure was monitored in the cannula tip through a thin polyethylene tube and could be adjusted to the desirable 40 mmHg level by tightening a screw-clamp placed around the cannula. After electrode placement, a prolonged 20-min CSO trial was performed in six animals to test for potential time-related hemodynamic, rhythm, electrocardiographic and $[K^+]_o$ effects. Two 6-min ischemia trials, one with CAO and the other with CAO + CSO, were peroformed in random order in each of the 12 animals under continuous recordings of a Y of the lead ECG, systemic and coronary sinus pressure and $[K^+]_o$ signals. The trials were separated by at least 20 min of normal perfusion and their randomized order turned out so that the CAO trial was first in seven experiments and the CAO + CSO trial was first in five. If VF occurred during a trial, cardiac resuscitation with cardiac massage for about 1 min was performed, followed by defibrillation with a 5 J DC shock. The trial was repeated after about 15 min when hemodynamics rhythm and ECG returned to baseline. At the end of the experiment, the condition, position and relative distance of electrodes were verified.

Data analysis

$[K^+]_o$ data were derived at 1 min intervals for each site and type of trial, in each animal. $[K^+]_o$ gradients were calculated in each experiment by dividing the $[K^+]_o$ differences by the interelectrode distance. Then, they were grouped and analysed using analysis of variance. Best fit curves were applied on the mean values over time. Reported data are mean ± S.E.

Results

Prolonged 20-min CSO trials, conducted in six experiments, did not produce any hemodynamic, electrocardiographic or rhythm abnormality. $[K^+]_o$ did not change significantly in any of the four zones. In the CAO trials, $[K^+]_o$ increased significantly within the IZ and BZi reaching a similar plateau after 5 min, at 11.89 ± 1.12 mEq/l. The data over time, fit a third order polynomial curve ($[K^+]_o = 5.45 - 2.42*\text{min} + 1.11*\text{min}^2 + 0.09*\text{min}^3$, $r = 0.998$, Fig. 1). During the first 3 min, $[K^+]_o$ increased not only in the IZ and BZi but also in the BZo up to 7.88 ± 0.48 mEq/l. Thereafter, while $[K^+]_o$ continued to increase in the IZ and BZi, it declined to 7.00 ± 0.40 mEq/l in the BZo and remained unchanged thereafter. Therefore, a substantial and sustained $[K^+]_o$ gradient of 5.54 ± 0.20 mEq/l ($P<0.001$) developed across the visible ischemic border following 3 min of CAO. A modest, but progressive and statistically significant $[K^+]_o$ increase, from 3.56 ± 0.23 to 5.01 ± 0.31 mEq/l ($P<0.005$), was also identified in the NZ.

In the trials where CAO was preceded by CSO, the course of the $[K^+]_o$ changes in the four zones was significantly altered (Fig. 2). First, no significant $[K^+]_o$ gradient developed across the visible border at any time during the 6-min trials. Second, although the time course of the $[K^+]_o$ increase in the IZ was similar to that observed during CAO, at the ischemic border (BZi and BZo), it followed a second order polynomial ($[K^+]_o = 1.79 + 2.47*\text{min} - 0.19*\text{min}^2$, $r = 0.993$). Third, the $[K^+]_o$ plateau reached at 6 min was substantially lower, for both the IZ and BZi, by 2.81 ± 0.39 and 2.02 ± 0.36 mEq/l respectively ($P<0.001$) and higher for the BZo by 2.70 ± 0.47 mEq/l, ($P<0.001$) as compared to that observed during CAO. Fourth, the $[K^+]_o$ increase in the IZ, lagged behind the increase at the border sites, so that for the initial 3 min it was significantly lower by 2.1 ± 0.32 mEq/l ($P<0.005$). Fifth, the $[K^+]_o$ in the NZ increased to 5.54 ± 0.20 mEq/l for the initial 3 min and then, declined gradually to control levels, unlike the sustained above 5 mEq/l elevation following CAO.

The calculated $[K^+]_o$ gradients were derived by dividing the differences between $[K^+]_o$ values obtained in the four sites by the interelectrode distance. The average values showed marked changes among regions following CSO. Figure 3 illustrates that the mean gradient developing across the border zone (BZi–BZo) follows a third order polynomial ($\Delta\ [K^+]_o$ mEq/l/mm $= 1.61 - 0.41*\text{min} + 0.15*\text{min}^2 - 0.0112*\text{min}^3$, $r = 0.983$), becoming positive

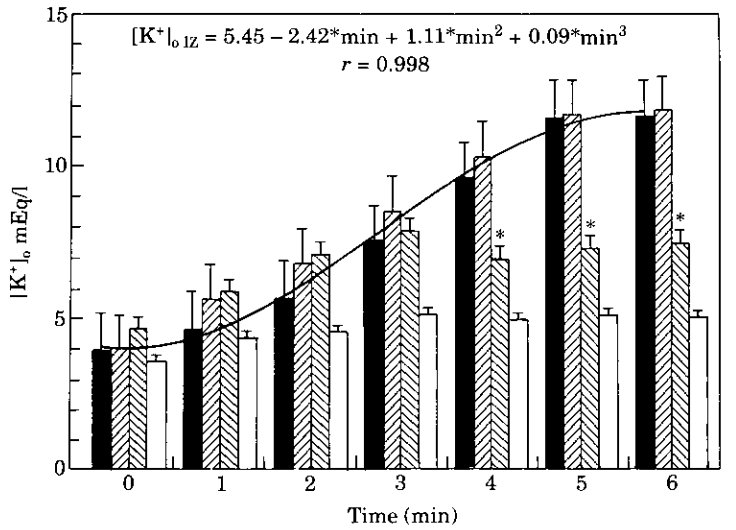

Figure 1 Time course of $[K^+]_o$ during the first 6 min of ischemia in 12 experiments (mean ± S.E.). (■) Ischemic zone site. (□) Normal zone site. BZi (▨) and BZo (▧), sites 5 mm apart, inside (BZi) and outside (BZo) the visible ischemic border. A significant $[K^+]_o$ gradient develops across the ischemic border after 3 min of ischemia (*$P<0.001$). (See text).

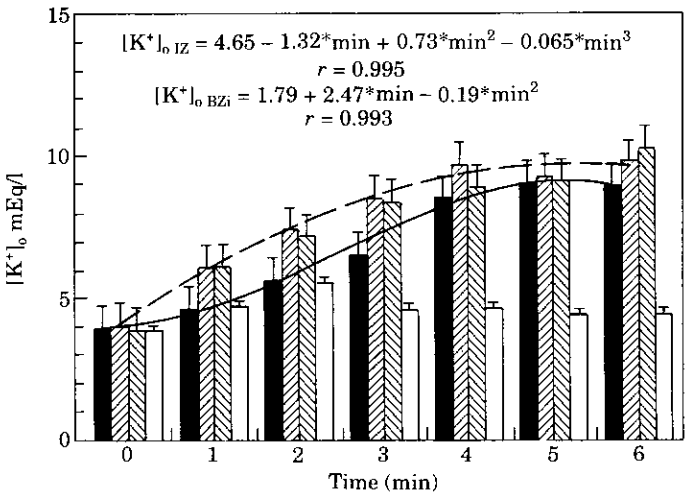

Figure 2 Time course of $[K^+]_o$ during the first 6 min of ischemia preceded by coronary sinus obstruction (CSO) in 12 experiments (mean ± S.E.). Site abbreviations are the same as in Figure 1. No significant $[K^+]_o$ gradient developed across the visible border of ischemia (BZi–BZo). (See text).

3 min after CAO and reaching 0.76 ± 0.14 mEq/l/mm at 6 min. The averages of the rest of the gradients did not exceed 0.20 mEq/l/mm and most often were not statistically significant. With CAO + CSO the mean BZi–BZo gradient became progressively negative (Fig. 4), whereas the gradient within the normally perfused region (BZo–NZ) appeared to increase progressively. Nevertheless, with CAO + CSO the magnitude of all gradients never exceeded 0.4 ± 0.04 mEq/l/mm.

VF limiting the time of data collection appeared only during CAO trials at the 3rd min in one animal and the 4th min in three others. The trial order was first in two VF occurrences and second in the other two. No VF occurred during combined CAO + CSO trials.

Discussion

In the present study we used sets of four strategically placed, K^+ sensitive electrodes to examine whether the recently identified protective effect of CSO from the early VF of acute ischemia (Kralios *et al.*, 1993)

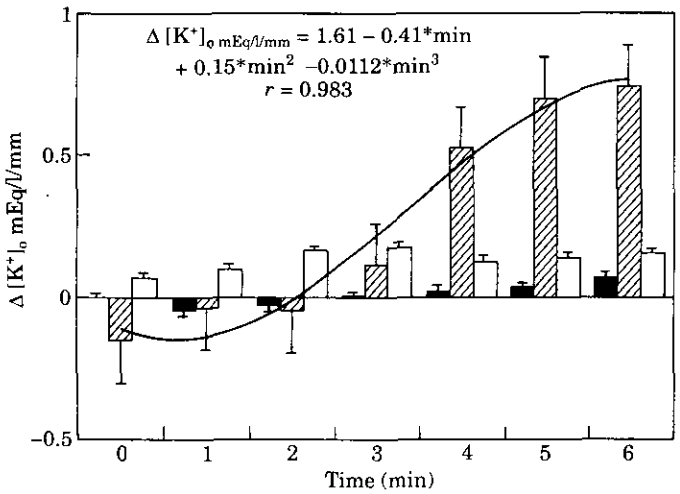

Figure 3 Estimates of $[K^+]_o$ gradients during the first 6 min of ischemia in 12 experiments. Gradients among sites were derived by dividing the difference of $[K^+]_o$ values by the interelectrode distance (mean ± S.E.). Gradients: (■) IZ–BZi, (▨) BZi–BZo, (□) BZo–NZ. A significant gradient develops over time across the visible border of ischemia.

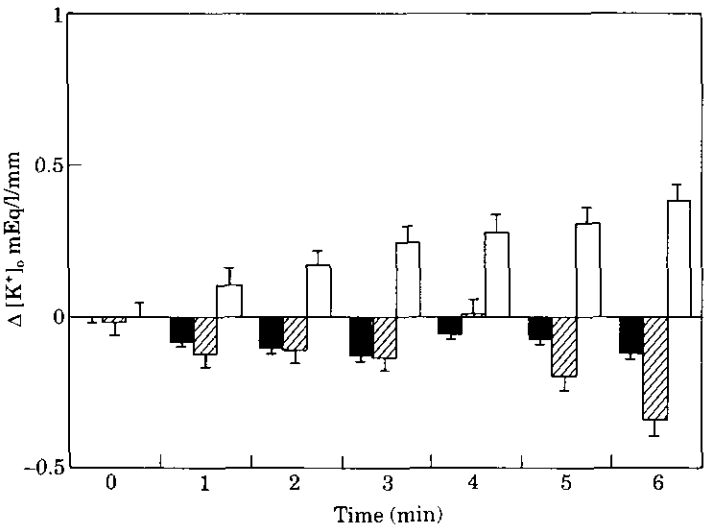

Figure 4 Estimates of $[K^+]_o$ gradients during the first 6 min of ischemia (CAO) preceded by coronary sinus obstruction (CSO) in 12 experiments. Gradients as in Figure 3. The small negative BZi–BZo gradient indicates $[K^+]_o$ diffusion past the visible border of ischemia.

involves correction of the inhomogeneous extracellular K^+ accumulation across the ischemic border (Pelleg *et al.*, 1989).

Whereas cellular hypoxia results in K^+ loss, it is the extracellular ($[K^+]_o$) accumulation which is associated with electrophysiologic effects (Yan *et al.*, 1993). Furthermore, whereas the extracellular accumulation during ischemia can be explained by lack of washout, (Harris *et al.*, 1954; Hill and Gettes, 1980; Hirche *et al.*, 1980) the reasons for its characteristic inhomogeneous distribution are unclear. Yet, it is this "patchy" $[K^+]_o$ distribution which is associated with and probably causative of the electrophysiological inhomogeneity that lead to the early arrhythmias (Pelleg *et al.*, 1989).

Since the functional changes occurring during the first 10 min of ischemia are completely reversible with reperfusion, repetitive trials performed in the same animal in random order yield comparable results, if separated by adequate reperfusion time (Clusin *et al.*, 1982; Kralios *et al.*, 1993). In this study, we limited the interruptions of coronary perfusion to brief 6-min periods, in order to attain the time of the peak incidence of phase Ia arrhythmias which characteristically begin after the first two min of ischemia, reach peak incidence at

5–6 min and decline thereafter (Meesmann *et al.*, 1978; Kaplinsky *et al.*, 1979; Clusin *et al.*, 1982; Kralios *et al.*, 1993; Muller and Kralios, 1993). Also, the 6-min time periods would be adequate to observe the rapid rate of $[K^+]_o$ rise that appears to parallel the peak incidence of phase Ia arrhythmias, while also reaching the plateau phase of $[K^+]_o$ accumulation (Kleber, 1984; Pelleg *et al.*, 1989), without prolonging ischemia unnecessarily.

For the same reason deliberate "ischemic preconditioning" known to influence $[K^+]_o$ accumulation and electrophysiologic changes (Fleet *et al.*, 1985) and to inhibit arrhythmias in subsequent ischemic episodes was not attempted. Nevertheless preconditioning was most probably and to a large extent accomplished not only by inadvertent coronary artery occlusions during the placement of pericoronary snares but also by the first deliberate CAO used to visualize the ischemic border. Furthermore, any potential systemic influence was counteracted by the balanced outcome of the randomization of the order of the trials.

As expected, the brief periods of ischemia in this study were marked by the characteristic rapid $[K^+]_o$ accumulation, that reached "plateau" at threefold control level after 5 min. Although individual values varied considerably, especially in the BZi, the course and magnitude of averaged values appeared similar for the central ischemic region (IZ), as well as the region near the visible border of ischemia (BZi) (Fig. 1). The apparent variance of this finding from those reported by Hariman *et al.* (1993) in the canine heart and Coronel *et al.* (1988) and Johnson *et al.* (1991) in the porcine heart, is presently unclear. It is possible that the initially visualized ischemic border may have shifted during the experiment so that some electrodes were sampling physiologically similar zones. Furthermore, the number of electrodes remaining after VF during CAO might have been insufficient to permit clear definition between zones.

Unexpectedly, the $[K^+]_o$ accumulation immediately outside the visible border (BZo), was also similar to that of the ischemic region for the first 3 min and only thereafter declined to a lower level. David *et al.* (1988) have described similar biphasic patterns of $[K^+]_o$ accumulation as a characteristic marker of the "border zone". Such findings indicate an initial free $[K^+]_o$ diffusion across the ischemic border, which only later becomes a diffusion barrier. The $[K^+]_o$ decline in the BZo accounted for part of the steep $[K^+]_o$ gradient developing across the border zone, after the 3rd min (Fig. 1). Since the $[K^+]_o$ differences between electrodes only a few millimeters apart across the ischemic border averaged about 4.5 mEq/l, they appear indicative of gradients potentially as high as 1 mEq/l/mm.

The rapid development and timing of these border zone gradients during CAO and particularly their elimination by simultaneous CSO are the main findings of this study (Figs 1 and 2). Conceptually, the onset of phase Ia arrhythmias after the second minute of ischemia was expected to coincide with the development of border zone gradients and the arrhythmia incidence to peak at the time of maximum gradient (Kaplinsky *et al.*, 1979; Kralios *et al.*, 1993). Indeed, all four incidents of VF, in this study, occurred after the 3rd min of CAO when $[K^+]_o$ gradients were at maximum. In contrast, during combined CAO + CSO, VF was prevented despite adequate rapidity ($d[K^+]_o/dt$) and magnitude of $[K^+]_o$ accumulation.

While prevention of VF by CSO was expected in accordance to recent findings (Kralios *et al.*, 1993), elimination of the gradient was anticipated only as a result of an overall $[K^+]_o$ elevation and equalization, reflecting retention of potassium ion rich, coronary venous effluent (Harris, 1966). Indeed, Curtis and Hearse (1989) demonstrated that ischemic arrhythmias in perfused rat hearts could be inhibited by increasing K^+ concentration in the perfusate. Moreover, Zipes *et al.*, (1975) were able to defibrillate canine hearts *in situ* by intracoronary KCl injections. In contrast to this notion, our findings with CSO indicate that $[K^+]_o$ levels were lower than during CAO and a progressively increasing reverse gradient appeared across the border as a mirror of the gradient from the border to the normal zone (Figs 3 and 4). These changes indicate that CSO sustains free $[K^+]_o$ diffusion across the border from the ischemic to the normal myocardium, from where it is washed out by the compensatory increase of coronary blood flow (Kralios *et al.*, 1976).

Antiarrhythmic effects due to attenuation of $[K^+]_o$ accumulation during combined CAO + CSO cannot be excluded. However, since $[K^+]_o$ levels were similar during the phase of rapid rise for both trials and VF was prevented when $[K^+]_o$ gradients were eliminated, it is reasonable to assume that the steepness of gradients and the degree of the resulting inhomogeneity are perhaps the critical factors in lethal arrhythmias.

In the absence of increased $[K^+]_o$ retention with CSO and since tissue oxygenation and exit of intracellular K^+ from the hypoxic cells are unlikely to be affected by CSO, the identified CSO effect could be due to correction of an extracellular myocardial hydration deficit (Kralios *et al.*, 1993). In acute ischemia water is shifting into the anaerobically metabolizing and thereby osmotically loaded myo-

cytes whereas the intercellular clefts and microvasculature are obliterated due to cell swelling (Kleber, 1984; Knopf *et al.*, 1990). Therefore, the volume of extracellular water-solvent may become deficient and contribute to dilutional amplification of solute concentrations. Although similar changes in the concentrations of a variety of other micromolecular electroactive or inert, ionic and even gaseous factors of cellular origin may be explained along this line, inhomogeneity in the continuum of extracellular environment can only signify space discontinuity. We speculate that critical reduction of extracellular fluid volume may promote formation of diffusion barriers respresenting collapsed parts of the extracellular space.

In contrast with CSO, sufficient hydration may restore hydraulic continuity, expand both the interstitial and intravascular fluid volumes, correct dilutional amplifications and promote diffusional equalization of solutes, while enhancing fluid transport through the interstitial space as well as through blood vessels and lymphatics. Such events are expected to preserve the homogeneity of extracellular environment. In regard to vascular transport, Laine (1991) has shown that in intact hearts, lymphatic flow increases in response to CSO. Whether this mechanism is still operative after interruption of coronary arterial perfusion is unclear.

Collateral flow to the ischemic area could also limit the production and improve the extracellular washout of arrhythmiogenic solutes, but as Hariman *et al.* (1993) have recently shown, the inadequacy of this flow tends to exacerbate the $[K^+]_o$ inhomogeneity. Although as shown previously (Kralios *et al.*, 1988), partial CSO does not affect coronary blood flow, it is unclear whether it can still modify collateral flow. Similarly, retrograde flow with coronary venous hypertension, a notion advanced half a century ago by Beck and Mako (1941) and Beck and Leighninger (1955) as a way of salvaging ischemic myocardium, results in inadequate flow rate and thus would also tend to increase the $[K^+]_o$ inhomogeneity. Finally, enhancement of tissue hydration by CSO may have prevented the diminution of extracellular space during ischemia which increases extracellular resistance and reduces conduction velocity (Kleber and Riegger, 1987; Kleber *et al.*, 1987; Cascio *et al.*, 1990).

In conclusion we found that CSO preceding CAO, facilitates $[K^+]_o$ dispersion, which decreases electrophysiologic disparities and thereby appears to prevent reentrant arrhythmias. The findings of this study confirm for the first time that elimination of $[K^+]_o$ disparities is possible not only by pharmacological but also physiological interventions and has remarkable salutary antiarrhythmic effects. The findings are consistent with the concept of extracellular hydration deficit, developing during early ischemia and corrected by CSO. Presumably, this deficit promotes dynamic formation of reversible diffusion barriers and fractionation of the extracellular space into isolated compartments, which enable sequestration of solutes including K^+. The disparity of solute concentrations may be the necessary and sufficient condition mandating the disparate electrophysiologic tissue characteristics of the arrhythmiogenic substrate. Accordingly, extracellular myocardial hydration may be central to the arrhythmiogenesis of acute ischemia.

References

Beck CS, Mako AE, 1941. Venous stasis in the coronary circulation. *Am Heart J* 21: 767–779.

Beck CS, Leighninger DS, 1955. Scientific basis for the surgical treatment of coronary artery disease.

Cascio WE, Gan-Xin Y, Kleber AG, 1990. Passive electrical properties, mechanical activity and extracellular potassium in arterially perfused and ischemic rabbit ventricular muscle. Effects of calcium entry blockade or hypocalcemia. *Circ Res* 66: 1461–1473.

Clusin WT, Bristow MR, Baim DS, Schroeder JS, Jaillon P, Brett P, Harrison DC, 1982. The effect of diltiazem and reduced serum ionized calcium on ischemic ventricular fibrillation in the dog. *Circ Res* 50: 518–526.

Coronel R, Fiolet JWT, Wilms-Schopman FJG, Schaapherder AFM, Johnson TA, Gettes LS, Janse MJ, 1988. Distribution of extracellular potassium and its relation to electrophysiologic changes during acute myocardial ischemia in the isolated perfused porcine heart. *Circulation* 5: 1125–1138.

Curtis MJ, Hearse DJ, 1989. Ischemia-induced and reperfusion-induced arrhythmias differ in their sensitivity to potassium: implications for mechanisms of initiation and maintenance of ventricular fibrillation. *J Mol Cell Cardiol* 21: 21–40.

David D, Michelson EL, Naito M, Dreifus LS, 1988. Extracellular potassium dynamics in the border zone during acute myocardial ischemia in a canine model. *J Am Coll Cardiol* 11: 422–430.

Fleet WF, Johnson TA, Graebner CA, Gettes LS, 1985. Effect of serial brief ischemic episodes on extracellular K^+, pH and activation in the pig. *Circulation* 4: 922–932.

Hariman RJ, Louie EK, Krahmer RL, Euler D, Bremner SM, Hwang MH, Ferguson JL, Loeb HS, 1993. Regional changes in blood flow, extracellular potassium and conduction during myocardial ischemia and reperfusion. *J Am Coll Cardiol* 21: 798–808.

Harris AS, Bisteni A, Rund RA, Brigham JC, Firestone JA, 1954. Excitatory factors in ventricular tachycardia resulting from myocardial ischemia: potassium a major excitant. *Science* 119: 200–203.

Harris AS, 1966. Potassium and experimental coronary artery occlusion. *Am Heart J* 76: 797–802.

HILL JL, GETTES LS, LYNCH MR, HEBERT NC, 1978. Flexible valinomycin electrodes for on-line determination of intravascular and myocardial K^+. *Am J Physiol* **235**: H455–H459.

HILL JL, GETTES LS, 1980. Effect of acute coronary artery occlusion on local myocardial extracellular K^+ activity in the swine. *Circulation* **61**: 768–778.

HIRCHE HJ, FRANZ CHR, BOX L, BISIG R, SCRAMM M, 1980. Myocardial extracellular K^+ and H^+ increase and noradrenaline release as possible cause of early arrhythmias following acute coronary artery occlusion in pigs. *J Mol Cell Cardiol* **12**: 579–593.

JOHNSON TA, ENGLE CL, BOYD LM, KOCH GG, GWINN M, GETTES LS, 1991. Magnitude and time course of extracellular potassium inhomogeneities during acute ischemia in pigs. Effect of verapamil *Circulation* **83**: 622–634.

KAPLINSKY E, OGAWA S, BALKE W, DREIFUS LS, 1979. Two periods of early ventricular arrhythmia in the canine acute myocardial infarction model. *Circulation* **60**: 397–403.

KLEBER AG, 1983. Resting membrane potential, extracellular potassium activity and intracellular sodium activity during acute global ischemia in isolated perfused guinea pig hearts. *Circ Res* **52**: 442–450.

KLEBER AG, 1984. Extracellular potassium accumulation in acute myocardial ischemia. *J Mol Cell Cardiol* **16**: 389–394.

KLEBER AG, RIEGGER CB, 1987. Electrical constants of arterially perfused rabbit papillary muscle. *J Physiol* [Lond] **385**: 307–324.

KLEBER AG, RIEGGER CB, JANSE MJ, 1987. Electrical uncoupling and increase of extracellular resistance after induction of ischemia in isolated arterially perfused rabbit papillary muscle. *Circ Res* **61**: 271–279.

KNOPF H, THEISING R, MOON CH, HIRCHE HJ, 1990. Continuous determination of extracellular space and changes of K^+, Na^+, Ca^{2+}, and H^+ during global ischemia in isolated rat hearts. *J Mol Cell Cardiol* **22**: 1259–1272.

KRALIOS AC, TSAGARIS TJ, KUIDA H, 1976. Myocardial oxygen consumption after major coronary artery occlusion in anesthetized dogs with constant workload. *Cardiovasc Res* **10**: 642–649

KRALIOS AC, NAPPI JM, TSAGARIS TJ, KRALIOS FA, KUIDA H, 1988. Paradoxical increase of ventricular fibrillation threshold in response to coronary sinus obstruction. *Am Heart J* **115**: 334–340.

KRALIOS AC, KRALIOS FA, ANDERSON FL, 1991. Coronary venous stasis abolishes the arrhythmiogenic substrate of ischemia (Abstr.). *J Am Coll Cardiol* **17**: 247A.

KRALIOS AC, ANDERSON FL, KRALIOS FA, 1993. Protective effect of coronary sinus obstruction from primary ischemia-induced ventricular fibrillation in the dog. *Am Heart J* **125**: 987–995.

LAINE GA, ALLEN SJ, 1991. Left ventricular myocardial edema. Lymph flow, interstitial fibrosis and cardiac function. *Circ Res* **68**: 1713–1721.

MEESMANN W, WIEGAND V, MENKEN U, KOMHARD W, REHWADE U, 1978. Early mortality due to ventricular fibrillation and the vulnerability of the heart following acute experimental coronary artery occlusion: possible mechanism and pharmacologic prophylaxis. In: *The Arterial System, Dynamics, Control, Theory and Relation*. Springer, Heidelberg, pp275–284.

MOE GK, 1962. On the multiple wavelet hypothesis of artrial fibrillation. *Arch Int Pharmacodyn Ther* **140**: 183–188.

MULLER CA, KRALIOS AC, 1993. Synergistic effects of hemodynamic stabilization and B blockade on ventricular fibrillation threshold in coronary artery ligated pigs. *Cardiovasc Res* **27**: 274–278.

PELLEG A, MITAMURA H, PRICE R, KAPLINSKY E, MENDUKE H, DREIFUS LS, MICHELSON EL, 1989. Extracellular potassium ion dynamics and ventricular arrhythmias in the canine heart. *J Am Coll Cardiol* **13**: 941–950.

WEISS J, SHINE KI, 1981. Extracellular potassium accumulation during myocardial ischemia: Implications for arrhythmogenesis. *J Mol Cell Cardiol* **13**: 699–704.

WEISS J, SHINE KI, 1982. K^+ accumulation and electrophysiologic alterations during early myocardial ischemia. *Am J Physiol* **243**: H318–327.

YAN GX, YAMADA KA, KLEBER AG, MCHOWAT J, CORR PB, 1993. Dissociation between cellular K^+ loss, reduction in repolarization time, and tissue ATP levels during myocardial hypoxia and ischemia. *Circ Res* **72**: 560–570.

ZIPES DP, FISHER J, KING RM, NICOLL A, JOLLY WW, 1975. Termination of ventricular fibrillation in dogs by depolarizing critical amount of myocardium. *Am J Cardiol* **36**: 37–44.