Supporting Methods

Study Protocol. The experimental protocol has been described in detail elsewhere.(1) Briefly, pigs (20-25 kg) of either sex were anesthetized and maintained under physiologic conditions. The heart was isolated and perfused at a constant flow of 220 ml/min with 37 ± 1°C Tyrode's solution and gassed with 95% O_2, 5% CO_2. An ECG was recorded via epicardial electrodes on the right ventricle (RV) and left ventricle (LV) with the ground on the aortic root. The ECG was monitored continuously and saved to disk for each episode (total duration of ECG saved to disk was 8-20 s, beginning 2 s before shock application). A catheter with a 34-mm platinum-coated titanium coil defibrillation electrode (Guidant) was inserted into the RV apex. A titanium mesh electrode (2.5-cm diameter) was sutured to the right atrium. Biphasic, truncated exponential shocks (6/4 ms) were delivered to the RV coil (cathodal, first phase) and the right atrial mesh electrode (anodal, first phase) from a defibrillator (Ventritex HVS-02). VF was induced by delivering 60-Hz alternating current to the electrode at the catheter tip, and defibrillation shocks were applied 10 s after initiation. Shock strengths were incremented from 100 to 900 V in 100-V increments to minimize the effects of tissue damage at high shock strengths. Ten experiments were attempted, but the complete protocol was completed in only five hearts. Each defibrillation attempt was classified as success or failure based on the absence or presence of arrhythmic activity from the ECG and optical maps 2 s after the shock. Rescue shocks of 20-30 Joules were required for all failed attempts and 2 min were allowed to elapse between VF episodes. Of the 45 defibrillation shocks, four were excluded from the analysis because the shock was not applied during the optical recording because of equipment malfunction.

Optical Mapping. Diacetyl monoxime was added to the perfusate to stop cardiac motion; the initial concentration of 10 mM/liter was not sufficient to eliminate motion in any of the hearts so a final concentration of 20 mM/liter was used. Although, the use of diacetyl monoxime **(**DAM) is expected to shorten the action potential duration during pacing, we have no reason to expect this fact to affect our findings. The hearts were stained with 1-[-3-sulfonatoproply]-4-[β-[[2-di-n-butylamino)-6-napthyl]vinyl]pyridinium (di-4-ANEPPS) (10.4 μM/liter, Molecular Probes), infused via the aortic root cannula. Transmembrane activity from the anterior and posterior epicardial surfaces was recorded from 0.5 s before until 2.5 s after each shock by two charge-coupled device cameras (MiCam01, Sci-Media) each with 64 × 96 pixels and aquiring images at 250 frames per s (1). Each pixel corresponded to an epicardial area of 0.66 ± 0.14 mm^2. The fluorescence signals were filtered by using a 5-point median temporal filter followed by a 3 × 3 × 3 boxcar spatio-temporal filter. In contrast to our previous study (1), a spatio-temporal filter was applied because we have found that phase map analysis is improved by smoothing repolarization. All recordings were normalized such that the minimum and maximum preshock values, and thus the action potential amplitude during VF for all sites, were identical. Movies of raw fluorescence, filtered fluorescence, and phase are provided in Movies 1-4. Until it is possible to record transmembrane potential from many sites in the beating human heart, we are limited to animal studies in this regard; many issues remain to be resolved such as the effects of disease, species variations, heart isolation, diacetyl monoxime, etc.

Data Analysis. The fluorescence signal at each site, $F(t)$, was converted to a phase signal, $\theta(t)$, as

$$\theta(t) = \arctan[F(t+\tau) - F_{50}, F(t) - F_{50}] \ , \qquad \textbf{[1]}$$

where F_{50} was the 50% level of $F(t)$ and τ was equal to 12 ms (2). F_{50} was chosen to be consistent with an earlier study (1) and τ is smaller than used previously to minimize the time interval of Eq. **1** because here we analyze events occurring during the short duration (10 ms) shock. Phase differences were computed as $\Delta\theta = \theta(t+\tau) - \theta(t)$; this corresponds to a total time interval of 24 ms (see Eq. **1**), which allowed us to study dynamics on the time scale of the duration of the shock (10 ms). PRCs were qualitatively similar when $\Delta\theta$ was computed with values separated by 8-24 ms; clearly as this interval approached half a cycle (~45 ms) the range of $\Delta\theta$ was 2π (one cycle). The PRCs for VF (no external stimulus) were not flat because of the difference between state space and time encoded phase(3). Because $\tau > 0$, phase decreases during an action potential, therefore $\Delta\theta < 0$ indicates a phase advance, whereas $\Delta\theta > 0$ indicates a phase delay. However, because of the cyclical nature of θ , $|\Delta\theta|$ is restricted to values $< \pi$, such that if $\Delta\theta$ is more negative than $-\pi$ it is actually a phase delay, and vice versa. We use the term PRC here to refer to graphs of $\Delta\theta$ versus θ (t).

The dynamic spatial patterns of transmembrane potential, $F(x,y,t)$, were depicted as either phase maps, $\theta(x,y,t)$, or isochrone maps (2, 4, 5). One of the main advantages of our phase mapping technique is its spatial (< 2 mm^2) and temporal (< 12 ms) localization; these scales (postsignal processing) are much smaller than the relevant dynamics (spiral wave core area is ~30 mm^2 and period is ~90 ms) (1, 6). The sites where all phase values converge, specifically the condition

$$\oint \nabla \theta \cdot d\vec{\ell} \neq 0 \qquad \textbf{[2]}$$

denotes a PS where the line integral is along any closed curve, and the sign of this integral indicates its chirality. Although all PSs do not represent complete reentrant pathways, a PS is always present for complete reentry (rotor); thus, the number of phase singularities at any instant is greater than or equal to the number of reentrant waves (2). An isochrone map depicts the position of the wavefront(s) as it propagates during one beat. First, the depolarization time was computed for each site as the time when $\theta = +\pi/2$ (this corresponds to the time the fluorescence signal crosses the 50% level during depolarization). Second, contour maps were generated from the spatial distribution of depolarization times.

1. Chattipakorn, N., Banville, I., Gray, R. A. & Ideker, R. E. (2001) *Circulation* **104,** 1313-1319.

2. Gray, R. A., Pertsov, A. M. & Jalife, J. (1998) *Nature* **392,** 75-78.

3. Oprisan, S. A., Thirumalai, V. & Canavier, C. C. (2003) *Biophys. J.* **84,** 2919-2928.

4. Gray, R. A. & Banville, I. (2002) in *Quantitative Cardiac Electrophysiology*, eds. Cabo, C. & Rosenbaum, D. S. (Dekker, New York), pp. 623-659.

5. Banville, I., Chattipakorn, N. & Gray, R. A. (2004) *J. Cardiovasc. Electrophysiol.* **15,** 455-463.

6. Pertsov, A. M., Davidenko, J. M., Salomonsz, R., Baxter, W. T. & Jalife, J. (1993) *Circ. Res.* **72,** 631-650.