Genesis of the monophasic action potential: role of interstitial resistance and boundary gradients

Joseph V. Tranquillo, Michael R. Franz, Björn C. Knollmann, Alexandra P. Henriquez, Doris A. Taylor, and Craig S. Henriquez

1Department of Biomedical Engineering, Duke University, Durham 27708; 2Georgetown University and Veterans Affairs Medical Centers, Washington, District of Columbia 20007; and 3North Carolina Supercomputing Center, Research Triangle Park, North Carolina 27709

Submitted 26 August 2003; accepted in final form 1 December 2003

The extracellular potential at the site of contact and the adjacent myocardium. During plateau, these sources dominate the surrounding sources and have monophasic time courses that follow the underlying TAPs. While the extracellular potentials above the electrode generated by this model are monophasic, they lacked features seen in recorded MAPs such as an initial negative, stable resting potential, and a clear upstroke. In addition, the magnitudes of the MAPs were at least an order of magnitude smaller than those reported experimentally. Henriquez and Papazoglou (12) developed a model of the MAP in which the contact pressure was assumed to open nonspecific stretch (pressure)-sensitive ion channels that depolarized the membrane. This model accounted for the negative resting potential, as well as the upstroke notch often seen in clinical MAPs, but again underestimated the amplitude of the signal. Finally, Vigmond and Leon (35) created a three-dimensional (3-D) model with a blood bath and the stretch-sensitive current developed by Henriquez and Papazoglou (12) to investigate MAP genesis. In contrast to previous studies, this investigation explored how the various parameters governing the size, depth, and magnitude of the depolarization in the electrode region affected the shape of the MAP signal. While accounting for many features of the MAP time course, the model of Vigmond and Leon produced very large MAP signals with amplitudes that approached that of the transmembrane potential. Whereas MAP signals from contact pressure are generally large, they are rarely >70 mV.

All of the aforementioned models had to make assumptions about the membrane behavior and tissue properties under the contact electrode. In a recent experimental study on a Langendorff-perfused mouse heart, the depolarization due to contact pressure was measured for the first time. The measurements showed that the application of pressure depolarizes the ventricular myocytes beneath the electrode to intracellular potentials of approximately −23 mV, giving rise to an extracellular MAP signal with an amplitude of 15–19 mV (with two-thirds of the potential <0 mV) (22). Spatial mapping near the contact region showed that the depolarization is very local. The intracellular potentials were found to rapidly return to normal resting values of approximately −78 mV within a short distance (~100–200 μm) away from the electrode contact site (22). Models have shown that the potential gradient at the electrode border is critical to producing a

EXTRACELLULARLY RECORDED SIGNALS, produced by applying a point mechanical load to the myocardium, have been shown to resemble the underlying transmembrane action potential (TAP) and thus have been used to gain information about the electrophysiology of cardiac muscle when intracellular microelectrode recordings are not possible or too challenging to perform. In the beating heart, these extracellular signals, generally termed monophasic action potentials (MAPs), are widely used to measure the effects of drugs and exogenous stimulation and to gain insight into the cellular origin of arrhythmias (7). Whereas MAPs and TAPs have been compared in several studies (9, 14, 15), the biophysical connection between the two signals is not completely understood (7, 18, 26, 36).

Several biophysical models have been created to elucidate the biophysical basis of the MAP produced by contact pressure. Malden and Henriquez (24, 25) and later Trayanova et al. (34), building on the early work of Hirsch et al. (13), modeled the region under the electrode as passive and not capable of generating an action potential. These models showed that large current sources form at the periphery of the inhomogeneity due to the large potential gradient created between the region of contact and the adjacent myocardium. During plateau, these sources dominate the surrounding sources and have monophasic time courses that follow the underlying TAPs. While the extracellular potentials above the electrode generated by this model are monophasic, they lacked features seen in recorded MAPs such as an initial negative, stable resting potential, and a clear upstroke. In addition, the magnitudes of the MAPs were at least an order of magnitude smaller than those reported experimentally. Henriquez and Papazoglou (12) developed a model of the MAP in which the contact pressure was assumed to open nonspecific stretch (pressure)-sensitive ion channels that depolarized the membrane. This model accounted for the negative resting potential, as well as the upstroke notch often seen in clinical MAPs, but again underestimated the amplitude of the signal. Finally, Vigmond and Leon (35) created a three-dimensional (3-D) model with a blood bath and the stretch-sensitive current developed by Henriquez and Papazoglou (12) to investigate MAP genesis. In contrast to previous studies, this investigation explored how the various parameters governing the size, depth, and magnitude of the depolarization in the electrode region affected the shape of the MAP signal. While accounting for many features of the MAP time course, the model of Vigmond and Leon produced very large MAP signals with amplitudes that approached that of the transmembrane potential. Whereas MAP signals from contact pressure are generally large, they are rarely >70 mV.

All of the aforementioned models had to make assumptions about the membrane behavior and tissue properties under the contact electrode. In a recent experimental study on a Langendorff-perfused mouse heart, the depolarization due to contact pressure was measured for the first time. The measurements showed that the application of pressure depolarizes the ventricular myocytes beneath the electrode to intracellular potentials of approximately −23 mV, giving rise to an extracellular MAP signal with an amplitude of 15–19 mV (with two-thirds of the potential <0 mV) (22). Spatial mapping near the contact region showed that the depolarization is very local. The intracellular potentials were found to rapidly return to normal resting values of approximately −78 mV within a short distance (~100–200 μm) away from the electrode contact site (22). Models have shown that the potential gradient at the electrode border is critical to producing a

H1370
MAP signal with a time course that follows the TAP. We hypothesize that the application of pressure restricts the current flow at the electrode site by altering the local interstitial resistance, and this restriction affects the degree of local depolarization and the magnitude of the MAP. To test this hypothesis, we have constructed a 3-D bidomain model of a thin layer of superfused mouse myocardium using data from the aforementioned experimental study to compute the extracellular potentials in the presence and absence of a pressure-induced depolarization for different tissue properties. The results show that the magnitude and the time course of the MAP are reproduced only for certain combinations of local or global intracellular and interstitial resistances that form a resting tissue length constant that is smaller than that required to match the wave speed. The results suggest that the application of pressure not only causes local depolarization but also changes local tissue properties, likely due to compression of the interstitial space.

METHODS

To investigate both extracellular and transmembrane potentials, a 3-D anisotropic bidomain model of a thin layer of myocardium (0.5 × 0.5 × 0.05 cm) adjoining an extensive upper bath (0.5 × 0.5 × 0.5 cm) and lower bath (0.5 × 0.5 × 0.05 cm) was created (Fig. 1A). The bidomain formulation of Roth (30) was used to solve for the transmembrane voltage ($V_{tm}$) and interstitial voltage ($V_{e}$)

\[
\nabla \cdot \left( \mathbf{D}_{i} \nabla V_{m} \right) + \nabla \cdot \left( \mathbf{D}_{e} \nabla V_{e} \right) = -\beta \left( C_{m} \frac{\partial V_{m}}{\partial t} + I_{m} \right)
\]

\[V_{m} = \phi_{i} - \phi_{e} \tag{1}\]

where $\beta$ (cm$^{-1}$) is the surface area-to-volume ratio, $\nabla$ is the gradient, $C_{m}$ (µF/cm$^2$) is the membrane capacitance, $I_{m}$ (µA/cm$^2$) is the sum of ionic fluxes, and $D_{i}$ and $D_{e}$ (mS/cm) are the bidomain conductivity tensors (3) in the intracellular (i) and interstitial (e) spaces, respectively. For simplicity, the tensors are assumed to be aligned with the coordinate axes. In this case, the bidomain tensors reduce to the scalar bidomain conductivities, $g_{i,e}$, $g_{i,e}^{+}$, and $g_{i,e}^{-}$ over a computational voxel, which is composed of both intracellular and interstitial domains

\[g_{i} = \psi \sigma_{i} \]

\[g_{e} = \psi \sigma_{e} \tag{2}\]

The values of $\psi_{i,e}$ are the ratio of intracellular and extracellular volume to the total computational volume (11). Because the dimension along the fiber ($L$) is the same for both domains the ratios $\psi_{i,e}$ are defined by the ratios of cross-sectional area ($A_{i}/A_{tot}$ and $A_{e}/A_{tot}$, as shown in Fig. 2).

To simulate a pressure-induced extracellular potential (a MAP), we assume the properties directly under the electrode region change. First, we assume the applied pressure opens stretch-sensitive channels to a specified depth, referred to as the depolarized depth. Namely, this depth is set to 50 µm. The stretch (pressure) sensitive channels are modeled as an nonspecific, linear current, $I_{press}$, added to $I_{ion}$ in the region of the electrode

\[I_{press} = g_{press}(V_{m} - E_{press}) \tag{3}\]

where $E_{press}$ is the reversal potential and $g_{press}$ is the maximum channel conductance (6, 12). Nominal values ($E_{press} = -6$ mV and $g_{press} = 2$ mS/cm$^2$), were chosen to be in the range of experimental measurements ($E_{press} = -6$ mV, $g_{press} = 2.32$ mS/cm$^2$) on stretch channels in rat cardiomyocytes obtained by Zeng et al. (37).

We also assume that the application of pressure could locally compress the interstitial space surrounding cells by causing the interstitial fluid to diffuse to surrounding tissue (10, 33, 38), whereas the intracellular volume remains constant as pressure is applied (due to the rigidity of the lipid membrane). Such effects of compression on

Fig. 1. A: three-dimensional (3-D) model used for the nominal case showing the tissue bounded by the upper and lower baths. B: X-Y plane at the boundary between the tissue and upper bath showing the electrode region. Dashed lines indicate where extracellular recordings were made. C: X-Z plane through the electrode region. D: simulated mouse action potential with lines of 90%, 50%, and 30% repolarization is shown as well as the total amplitude of the monophasic action potential (MAPA) or transmembrane action potential (TAPA) signal.

AJP-Heart Circ Physiol • VOL 286 • APRIL 2004 • www.ajpheart.org
The insulating shaft of the electrode is represented by reducing the bath conductivity to zero in a column above the electrode region in the upper bath. No-flux boundary conditions were assumed at the external faces of the tissue and bath. At tissue/bath boundaries continuity of interstitial and extracellular potentials and continuity of normal interstitial and extracellular currents were assumed (11).

Because the instantaneous change of properties under the contact electrode region produces a suprathreshold depolarization large enough to initiate a waveform, time \( t = 0 \) was defined to occur 200 ms after the initial waveform to ensure that tissue reached a steady state (12). Wavefronts were then initiated either along or across fibers by applying a transmembrane current stimulus of 1.000 \( \mu \text{A/cm}^2 \) for 0.5 ms along an entire face of the tissue, as indicated by the shaded region in Fig. 1A.

The time traces of the surface \( V_m \) and \( \phi_e \) were obtained at surface sites along the dashed lines indicated in Fig. 1B (along the \( x \)- and \( y \)-axes) and along a line into the tissue depth directly below the electrode region (along the \( z \)-axis of Fig. 1C). The surface interstitial signals, \( \phi_i \), are referenced to a point located 0.4 cm into the upper bath, at the boundary of the insulating electrode shaft.

The shape of MAPs and TAPs were characterized using several measures (shown in Fig. 1D). The MAP amplitude is reported as three numbers: the total amplitude (MAPA) and the minimum and maximum voltages (appearing in parentheses). The MAP minimum was defined as the absolute baseline voltage (discounting any negative deflections due to the notch). The MAP duration (MAPD) is also reported as three numbers: MAPD30, MAPD50, and MAPD90 at 30% repolarization, 50% repolarization, and 90% repolarization, respectively. The difference between the MAPD30, MAPD50, and MAPD90 and the surface TAPD30, TAPD50, and TAPD90 far from the electrode (remote) were also calculated (appearing in parentheses). The maximum rate of rise (\( V_{\text{max}} \)) was computed using a forward difference on the upstroke. The rise time (RT) is defined as the time of the first deflection from baseline to the peak of the MAP.

Experiments have shown a rapid return to rest near the site of depolarization. Jack et al. (16) have shown that the spatial distribution of potentials resulting from a point source perturbation in a multidimensional monodomain is described by a modified Bessel function of the second kind \( (K_0) \) (16). No analytic solution exists for the spatial distribution of potentials in an unequal anisotropic bidomain although the solution should approach a modified Bessel function in the limit of equal anisotropy. We therefore fit the simulated diastolic intracellular potential distribution to

\[
\phi_d(r) = c_1 K_0(r c_2) + c_2
\]

where \( c_1 \) and \( c_2 \) are constants, \( r \) is the distance from the electrode edge, and \( \gamma \) is the estimate of the theoretical one-dimensional (1-D) length constant given by

\[
\lambda = \frac{s g_e}{\beta G_m(g_0 + g_e)}
\]

In all simulations, the parameter \( \gamma \) was calculated along and across fibers as well as transmurally and compared with the theoretical 1-D length constant. Note that the Bessel function falls off considerably faster than an exponential with the same length constant.

Spatial discretization was performed using a vertex-centered finite volume method with a space step of 50 \( \mu \text{m} \) (28). The forward Euler method was used to solve the parabolic equation in the coupled bidomain equation (Eq. 1) with a time step of 2 \( \mu \text{s} \), and a Jacobi preconditioned generalized minimal residual algorithm solver (31) was used to solve the elliptic equation. All simulations were performed on up to 32 processors on the IBM SP at the North Carolina Supercomputing Center using a simulation package, CARDIO-WAVE, designed for parallel computation (29).

A series of simulations were performed to determine a set of tissue properties that would approximately match the experimental values of...
the local depolarization due to contact pressure, MAPA, and the conduction velocity in murine cardiac tissue. In all simulations, the pressure-sensitive channels were assumed to have a conductance and reversal potential \( g_{\text{ress}} = 2 \text{ mS/cm}^2 \) and \( E_{\text{ress}} = -6 \text{ mV} \) based on the experimental studies of Zeng et al. (37). In the first simulation, the bidomain conductivities were assumed to be uniform throughout the tissue, with \( \Psi_i \) and \( \Psi_e \) in Eq. 2. In the second simulation, the interstitial bidomain conductivities were uniformly reduced (\( \Psi_i = 0.1 \)) while the intracellular bidomain conductivity was unchanged (\( \Psi_e = 0.7 \)), reducing the resting tissue length constant. This case corresponds to a uniform compression of the interstitial space. In the third simulation, the interstitial bidomain conductivity was the same as the first case (\( \Psi_i = 0.3 \)), but the intracellular conductance was reduced to give the same length constant as in the second simulation (\( \Psi_e = 0.2 \)). In the final simulation, the bidomain conductivities were the same as the first simulation (\( \Psi_i = 0.7 \) and \( \Psi_e = 0.1 \)), except in the electrode region where \( \Psi_{\text{elec}} = 0.1 \). This simulation is an attempt to model local compression of the interstitial space under the electrode and produce a resting tissue length constant near the electrode that is similar to that obtained for the second and third simulations. The tissue properties used in this last simulation are given in the Appendix and are used to produce what is later referred to as the nominal case.

A parameter variation study was performed, using the bidomain conductivities in the nominal case, to investigate how local changes in the electrode region impact the MAP timecourse and amplitude. The degree of compression explored by varying from \( \Psi_{\text{elec}} \) from 0.3 (no compression) to 0.02. To investigate the impact of transmural wall thickness, the tissue preparation was varied from a thickness of 250 to 2,000 \( \mu \text{m} \). The diameter of the contact MAP electrode varies depending on the region of contact pressure and at a distant site, 1,250 \( \mu \text{m} \) from the MAP electrode, were computed. The simulated potential at the pressure region displays many characteristics seen in experimental MAP recordings, including a notch coinciding with the activation deflection at the reference electrode, and an offset such that slightly more than two-thirds of the signal is \( -0 \text{ mV} \). The distant electrogram is biphasic, as expected from a planar front. Figure 4 shows the simulated MAPs, and the corresponding intracellular and transmembrane potentials, and the distant electrogram for the four sets of conductivities discussed above.

For the first simulation set (Fig. 4A), the propagation velocity of the front was 78 cm/s, consistent with that reported in mouse myocardium along fibers (5, 32). The MAPA was 6.1 mV, significantly smaller than experimental values, while the magnitude of the distant electrogram (4.0 mV) was consistent with experimental values. For the second simulation set (Fig. 4B), the MAPA increased to 21.2 mV, a value closer to that measured. However, the remote electrogram magnitude was larger (8.9 mV) and the conduction velocity was considerably smaller (56 cm/s) than measured values. Recall that the third set of conductivities (Fig. 4C) produced nearly the same local depolarization and fall offs as in the second set. As a result, the conduction velocity was approximately the same (55 cm/s). The MAPA (6.1 mV) and the magnitude of the remote electrogram (8.2 mV), however, were similar to that obtained using the first set. Finally, for the fourth set, corresponding to local compression (Fig. 4D), the conduction velocity was 78 cm/s, the MAPA was 20.9 mV, and the magnitude of remote electrogram was 3.9 mV (Table 1).

Transmembrane currents at pressure site during wavefront propagation. As shown in Fig. 3, the electrode region is more depolarized than the surrounding tissue, giving rise to the spatial gradient in transmembrane potential. Because the transmembrane current is proportional to the divergence of the

MODEL OF MAP GENESIS

H1373

AJP-Heart Circ Physiol • VOL 286 • APRIL 2004 • www.ajpheart.org
transmembrane potential gradient, the magnitude of the transmembrane current at the electrode edge is large and negative at diastole. Figure 5 shows the spatial profile of the transmembrane current near the electrode region in the absence (Fig. 5A) and presence of local compression (Fig. 5B). Recall that both the degree of local depolarization and the MAPA for the no compression case are markedly less than that obtained for the compression case. The spatial fall offs are, however, approximately the same. In both cases, when the wavefront passes by the electrode region, the transmembrane potential is more positive than that observed under the electrode, and the transmembrane current at the electrode edge switches polarity and becomes positive (Fig. 5, A-II and B-II). As the surrounding tissue repolarizes, the transmembrane current at the boundary becomes less and less positive until it switches polarity again and returns to rest (Fig. 5, A-III,IV and B-III,IV).

Fig. 3. The simulated fall off in resting interstitial voltage ($\phi_i$; solid lines), intracellular voltage ($\phi_c$; dashed lines), and transmembrane voltage ($V_m$; dashed-dotted lines) in space along the x (left), y (middle), and z (right) axes, where 0 $\mu$m corresponds to the center of the electrode region. Only positive distances are shown due to symmetry about the electrode region. A: diastolic fall offs for uniform conductivities ($\Psi_e = 0.3$). B: diastolic fall offs when interstitial conductivities are uniformly reduced ($\Psi_e = 0.1$) in response to pressure. C: diastolic fall offs when $\Psi_i = 0.2$. D: diastolic fall offs when interstitial conductivities are locally reduced in response to pressure.
The transmembrane currents at the electrode edge follow the TAP time course (Fig. 6, A-II and B-II); however, 100 μm from the electrode edge the transmembrane currents are biphasic (Fig. 6, A-IV and B-IV). Consequently, the extracellular potential near the electrode region is monophasic and rapidly becomes biphasic away from the electrode region. In both the no compression and compression cases ($\Psi_{elec}$ = 0.1 and $\Psi_{elec}$ = 0.3), transmembrane current is similar in magnitude adjacent and remote from the electrode site. The MAPAs at the pressure region for the two cases are markedly different, whereas the remote electrograms have the same magnitudes.

Parameters affecting MAP/TAP correspondence. The MAP signal is assumed to correlate with the underlying transmembrane potential that would arise in the absence of the contact pressure. The pressure-induced depolarization, however, is expected to alter the characteristics of the TAP near the electrode region. With the use of the fourth set of conductivities (see APPENDIX), comparisons were made between the MAP

---

**Fig. 4.** A comparison of $\phi_e$ (solid lines), $\phi_i$ (dashed lines), and $V_m$ (dashed-dotted lines) under the electrode with (left) and without pressure (right) for uniform conductivities (A), globally reduced interstitial conductivities (B), decreased intracellular conductivities ($\Psi_{i} = 0.2$) (C), and locally reduced interstitial conductivities (D).
reduced interstitial conductivities. V

TAP elec , which has both reduced amplitude and reduced isolated whole cell preparation, Fig. 5.

Comparison of potentials, conduction velocities, and length constants for four simulation sets

<table>
<thead>
<tr>
<th></th>
<th>CV, cm/s</th>
<th>EGA, mV</th>
<th>MAPA, mV</th>
<th>V(_{\text{m}}) (rest), mV</th>
<th>(\varphi_e) (rest), mV</th>
<th>(V_{\text{max}}), V/s</th>
<th>(\gamma_e^{x,y,z}), (\mu)</th>
<th>(\lambda_e^{x,y,z}), (\mu)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(\psi_e = 0.3)</td>
<td>78/32</td>
<td>4.0</td>
<td>6.1</td>
<td>36.7</td>
<td>40.7</td>
<td>5.2</td>
<td>773/254/237</td>
<td>720/290/290</td>
</tr>
<tr>
<td>Global (\psi_e = 0.1)</td>
<td>56/25</td>
<td>8.9</td>
<td>21.2</td>
<td>14.0</td>
<td>26.6</td>
<td>8.8</td>
<td>581/188/208</td>
<td>500/240/240</td>
</tr>
<tr>
<td>Decreased (g_i)</td>
<td>55/25</td>
<td>8.2</td>
<td>6.0</td>
<td>22.4</td>
<td>26.5</td>
<td>4.9</td>
<td>584/190/216</td>
<td>500/240/240</td>
</tr>
<tr>
<td>Local (\psi_e = 0.1)</td>
<td>78/31</td>
<td>3.9</td>
<td>20.9</td>
<td>12.8</td>
<td>26.8</td>
<td>9.0</td>
<td>735/231/224</td>
<td>720/290/290</td>
</tr>
<tr>
<td>Experimental</td>
<td>60–80/20–40</td>
<td>4.0</td>
<td>15–19</td>
<td>23.8</td>
<td>26.5</td>
<td>9.2</td>
<td>5.4–9.2</td>
<td></td>
</tr>
</tbody>
</table>

Comparison of the monophasic action potential (MAP), electrogram amplitude (EGA), conduction velocity (CV) along and across the fiber direction, and length constants for the four simulations with variable tissue properties is shown. \(V_{\text{max}}\), maximum upstroke of the MAP; \(V_{\text{m}}\), transmembrane voltage; \(\varphi_e\), interstitial voltage; MAPA, total MAP amplitude; \(\gamma_e^{x,y,z}\), estimated theoretical length constants; \(\lambda_e^{x,y,z}\), unperturbed one-dimensional length constants.

and 1) the intrinsic, uncoupled \(V_{\text{m}}\) as would be recorded by an isolated whole cell preparation, 2) \(V_{\text{m}}\) on the surface far from the recording site (remote) when loaded by the tissue and baths, and 3) \(V_{\text{m}}\) at the edge of the electrode (TAP\textsuperscript{elec}). Note that \(V_{\text{m}}\) at the remote site was identical to that which would be obtained under the electrode region in the absence of applied pressure. Table 2 compares these four simulated \(V_{\text{m}}\) signals with \(\varphi_e\). The timecourse of the MAP most closely resembles TAP\textsuperscript{elec}, which has both reduced amplitude and reduced \(V_{\text{max}}\) compared with the remote TAP.

A parameter variation study was performed to investigate how changes local to the electrode region impact the time course of the MAP. Table 3 shows that increasing the degree of compression of interstitial space by decreasing \(\Psi_{\text{elec}}\) increases MAPA and \(V_{\text{max}}\) while not significantly altering MAPD or RT. More specifically, decreasing \(\Psi_{\text{elec}}\) from 0.3 to 0.02 increased MAPA by 25.2 mV. The decrease in \(\Psi_{\text{elec}}\) constrains the local length constants from \(\lambda^x = 773\) \(\mu\)m, \(\lambda^y = 254\) \(\mu\)m, \(\lambda^z = 237\) \(\mu\)m to \(\lambda^x = 675\) \(\mu\)m, \(\lambda^y = 207\) \(\mu\)m, and \(\lambda^z = 208\) \(\mu\)m. An increase in tissue thickness from 250 to 2,000 \(\mu\)m produced only a modest 5-mV increase in MAPA. Beyond this thickness, the change in MAPA was negligible. Increasing the depth of depolarization from 50 to 250 \(\mu\)m reduced MAPD, in particular MAPD\textsubscript{90}, by 11.7 ms. The MAPA increased to 30.4 mV with increasing depolarization depth up to 100 \(\mu\)m but then decreased to 27.3 mV beyond this depth. Experimental studies have shown that increasing the electrode size resulted in smaller MAPA, \(V_{\text{max}}\), and more distorted MAPD/TAPD correspondence (7). Increasing the size of the contact region in the model from 250 to 600 \(\mu\)m decreased MAPA and \(V_{\text{max}}\) by 54% and 73%, respectively, whereas the difference between MAPD\textsubscript{90} and TAPD\textsubscript{90} increased from 0.6 to 4.2 ms.

In a partial bidomain, Vigmond and Leon (35) found that changes in the conductance and reversal potential of the pressure-sensitive channels had a small effect on the MAP. This was also found to be the case for the complete bidomain model. Changing \(E_{\text{press}}\) alters the diastolic depolarization under the electrode and therefore the direct current offset of the MAP but does not significantly affect other MAP properties. Increasing the magnitude of \(g_{\text{press}}\) from 2 to 4 mS/cm\textsuperscript{2} increases MAPA by 3.3 mV and \(V_{\text{max}}\) from 9.0 to 9.8 V/s and slightly decreases the fitted 1-D length constant from \(\lambda^x = 735\) \(\mu\)m, \(\lambda^y = 231\) \(\mu\)m, and \(\lambda^z = 224\) \(\mu\)m to \(\lambda^x = 719\) \(\mu\)m, \(\lambda^y = 220\) \(\mu\)m, and \(\lambda^z = 221\) \(\mu\)m. Finally, initiating a wavefront in the slow direction (across fibers) reduces \(V_{\text{max}}\) from 9.0 to 4.7 V/s, increases RT from 5.9 to 8.2 ms, and increases MAPD\textsubscript{90} to 80.2 ms.

Effect of bath and shaft. Previous models of MAP genesis have suggested that the bath has a negligible effect on MAPA (35). To investigate the impact of the bath on MAPA in the full bidomain model, a planar wavefront was launched along and across fibers in the absence of the upper bath. In this case, the reference electrode was placed at the far lower corner of the tissue. The two MAPs computed in the absence of a bath are compared with the nominal MAPA. The MAPA along and across fibers were found to be 55.6 and 30.1 mV, respectively, significantly larger than in the presence of a bath. The presence of a shaft also had a significant impact. When the shaft was not included, the values for the MAPA were markedly reduced to

Fig. 5. A: transmembrane current in space (\(I_m\)) along the x-axis near the electrode at \(t = 0\) ms (I), \(t = 8\) ms (II), \(t = 40\) ms (III), and \(t = 60\) ms (IV) for uniform conductivities (\(\Psi_e = 0.3\)). B: \(I_m\) along the x-axis near the electrode at \(t = 0\) ms (I), \(t = 8\) ms (II), \(t = 40\) ms (III), and \(t = 60\) ms (IV) for locally reduced interstitial conductivities.
takeoff potential of under the MAP electrode was 28.0 mV peak to peak with a takeoff potential of 99.2 mV with a takeoff potential of remote intracellular potentials had a peak to peak amplitude of 134.7 V/s. The experimental MAP recorded in vitro (Fig. 7) had a MAPA of 18.4 mV and experimental intracellular potential 0.2 mm away from the electrode (Fig. 7A) has a peak-to-peak amplitude of 31.8 mV, a takeoff potential of −23 mV, and V\textsubscript{max} = 20.4 V/s. The simulated intracellular potential directly under the MAP electrode was 28.0 mV peak to peak with a takeoff potential of −26.8 mV and V\textsubscript{max} = 19.8 V/s. The experimental intracellular potential 0.2 mm away from the electrode (Fig. 7B) was 97.2 mV peak to peak with a takeoff potential of −76 mV and V\textsubscript{max} = 128 V/s. The simulated remote intracellular potentials had a peak to peak amplitude of 99.2 mV with a takeoff potential of −74.5 mV and V\textsubscript{max} = 134.7 V/s. The experimental MAP recorded in vitro (Fig. 7C) had a MAPA of 18.4 mV and V\textsubscript{max} = 8.6 V/s, which are within the ranges reported by Knollmann et al. (21) (MAPA = 15–19 mV, V\textsubscript{max} = 5.4–9.2 V/s) and similar to those of Danik et al. (4) (V\textsubscript{max} = 3.3–15.1 V/s). The simulated MAPA was 20.9 mV and V\textsubscript{max} = 9.0 V/s. While the conduction velocities were not directly measured in the study described in Knollmann et al., other investigations have shown that they range between 60 and 80 cm/s along fibers and between 20 and 40 cm/s across fibers (4, 33). The simulated conduction velocity along fibers was 78 and 31 cm/s across fibers. Finally, the magnitude of remote electrogram (3.9 mV) compared well with the experimentally recorded electrograms (~4.0 mV).

The only significant difference between the simulated and experimental recordings was the MAPD. The difference of −12 ms, however, is consistent with the differences between the APD of the Pandit ionic dynamics used for the model (55.7 ms) and the APD of the mouse used in the experiments (43.0 ms) (20). Note that in both the simulated and experimental situations, the MAPD is within 1 ms of the underlying TAPD.

**DISCUSSION**

It has long been postulated that the MAP relies on a local depolarization under the electrode (7). Our recent experimental study using a Langendorff-perfused mouse heart produced the first direct evidence that the region under pressure is depolarized at diastole and electronically follows the time course of the action potential as a wavefront passes by (22).

### Table 2. Comparison of MAP with transmembrane potential

<table>
<thead>
<tr>
<th>MAPA, mV</th>
<th>MAPD\textsubscript{30%} ms</th>
<th>MAPD\textsubscript{50%} ms</th>
<th>MAPD\textsubscript{90%} ms</th>
<th>V\textsubscript{max}, V/s</th>
<th>RT, ms</th>
</tr>
</thead>
<tbody>
<tr>
<td>MAP</td>
<td>15.8 (−14.0/6.9)</td>
<td>29.2 (−25.5/22.2)</td>
<td>43.0 (−38.4/16.9)</td>
<td>19.8 (−15.1/3.3)</td>
<td>5.9</td>
</tr>
<tr>
<td>Intrinsic V\textsubscript{m}</td>
<td>118.6 (−80.4/38.2)</td>
<td>21.1 (−27.5/12.6)</td>
<td>55.7 (−37.9/14.9)</td>
<td>194.5 (−154.2/41.1)</td>
<td>5.1</td>
</tr>
<tr>
<td>Remote V\textsubscript{m}</td>
<td>99.2 (−74.5/24.7)</td>
<td>12.6 (−29.4/18.9)</td>
<td>76.6 (−29.4/18.9)</td>
<td>134.7 (−29.4/18.9)</td>
<td>6.2</td>
</tr>
<tr>
<td>Local V\textsubscript{m}</td>
<td>7.1 (−12.8/−5.7)</td>
<td>20.0 (−31.4/20.0)</td>
<td>77.0 (−31.4/20.0)</td>
<td>6.3 (−6.3/3.3)</td>
<td>6.3</td>
</tr>
<tr>
<td>Local φ\textsubscript{1}</td>
<td>28.0 (−26.8/1.2)</td>
<td>20.6 (−31.0/20.6)</td>
<td>77.2 (−31.0/20.6)</td>
<td>19.8 (−19.8/6.5)</td>
<td>6.5</td>
</tr>
</tbody>
</table>

MAP characteristics were compared with the intrinsic V\textsubscript{m}, V\textsubscript{m} far from the electrode (remote), and V\textsubscript{m} directly under the electrode (local). MAPD\textsubscript{30\%}, MAPD\textsubscript{50\%}, MAPD\textsubscript{90\%}, times of 30%, 50%, and 90% repolarization of the MAP; RT, rise time.
opens nonspecific stretch-activated channels that locally depolarize the membrane potential under the electrode. Both the experiment and the simulations show that at the edge of the electrode region the fall off of the diastolic intracellular potential is rapid, returning to rest values within 100–200 μm. In our bidomain model, the distribution near the electrode at diastole was found to fall off rapidly and was fit best by a modified Bessel function, according to Eq. 5, consistent with theoretical predictions in multidimensional monodomain tissue (16). The voltage gradient created between the depolarized electrode region and surrounding myocardium gives rise to monophasic current sources that follow the TAP. The extracellular potential

\[
\psi_e^{\text{lec}} \text{, ratio of extracellular volume to the electrode region. Nominal parameters were used while } \psi_e^{\text{lec}} \text{ was varied from 0.3 to 0.02.}
\]

<table>
<thead>
<tr>
<th>(\psi_e^{\text{lec}})</th>
<th>MAPA, mV</th>
<th>MAPD50, ms</th>
<th>MAPD30, ms</th>
<th>(V_{\text{max}}), V/s</th>
<th>(\gamma^{\text{lec}}, \mu\text{m})</th>
<th>RT, ms</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.3</td>
<td>6.1 (−4.0/2.1)</td>
<td>77.3 (0.7)</td>
<td>21.7 (2.8)</td>
<td>5.2</td>
<td>773/254/237</td>
<td>5.9</td>
</tr>
<tr>
<td>0.2</td>
<td>12.6 (−7.2/5.4)</td>
<td>77.3 (0.7)</td>
<td>21.3 (2.4)</td>
<td>7.0</td>
<td>762/248/230</td>
<td>5.9</td>
</tr>
<tr>
<td>0.1</td>
<td>20.9 (−14.0/6.9)</td>
<td>77.2 (0.6)</td>
<td>21.1 (2.2)</td>
<td>9.0</td>
<td>735/231/224</td>
<td>5.9</td>
</tr>
<tr>
<td>0.05</td>
<td>29.9 (−17.1/12.8)</td>
<td>76.7 (0.1)</td>
<td>20.5 (1.6)</td>
<td>12.6</td>
<td>711/110/221</td>
<td>6.0</td>
</tr>
<tr>
<td>0.02</td>
<td>31.3 (−17.7/13.6)</td>
<td>76.7 (0.1)</td>
<td>20.4 (1.5)</td>
<td>14.3</td>
<td>675/207/208</td>
<td>6.1</td>
</tr>
</tbody>
</table>

Table 3. Effects of \(\psi_e^{\text{lec}}\) on MAP

Fig. 7. Comparison between representative experimental MAP and TAP measurements in a mouse heart (A–C) to the MAP and \(\phi_t\) measurements of the model (D–F). The experimental values are for the center signals. A: experimental TAP recorded directly under the MAP electrode (31.8 mV peak to peak, takeoff potential of −23 mV, \(V_{\text{max}} = 20.4\) V/s). B: experimental TAP recorded far from the MAP recording site (97.2 mV peak to peak, takeoff potential of −23 mV, \(V_{\text{max}} = 128\) V/s). C: example of an experimental MAP (MAPA = 18.4 mV and \(V_{\text{max}} = 8.6\) V/s). Note the presence of negative deflection from baseline (notch) before the upstroke. D: simulated \(\phi_t\) under the MAP electrode (28.0 mV peak to peak, takeoff potential of −26.8 mV, \(V_{\text{max}} = 19.8\) V/s). E: simulated \(\phi_t\) far from the MAP recording site (99.2 mV peak to peak, takeoff potential of −74.5 mV, \(V_{\text{max}} = 134.7\) V/s). F: simulated MAP (20.9 mV, \(V_{\text{max}} = 9.0\) V/s).
near the electrode is therefore monophasic. The emergence of monophasic currents at the boundary of the electrode due to the gradient in potential established by the local pressure is consistent with the original hypothesis of MAP genesis postulated by Franz (7, 8). Previous models, however, have not adequately accounted for MAPA and $V_{max}$ observed experimentally (13, 34, 35). The simulation studies performed here suggest that the MAPA is only reproduced when it assumed tally (13, 34, 35). The simulation studies performed here suggest that the MAPA is only reproduced when it assumed tally (13, 34, 35). The simulation studies performed here suggest that the MAPA is only reproduced when it assumed tally (13, 34, 35). The simulation studies performed here suggest that the MAPA is only reproduced when it assumed tally (13, 34, 35).

**Interstitial space modulates MAPA and $V_{max}$.** As shown in Table 3, if a constriction of interstitial space is not assumed ($\Psi$), the MAPA = 6.1 mV and $V_{max}$ are significantly less than the values experimentally recorded in mice (MAPA = 15–19 mV and $V_{max}$ = 5.4–9.2 V/s) (21). In addition, the absence of compression leads to a much smaller local depolarization than observed experimentally. With compression $\Psi_{elec}$ = 0.1, the length constant is decreased, allowing for a greater depolarization under the electrode (within 4 mV of the experimental intracellular resting potential). The consequence of a greater depolarization is a larger voltage gradient leading to a slight increase in the magnitude current source at the electrode boundary (Figs. 5 and 6). The change in current source magnitude, however, is small compared with the large change in MAPA. It should be noted that altering $\Psi_{elec}$ affects the local tissue length constant through a change in $g_{i}^{\lambda;c}$. To determine whether the interstitial resistance or whether the voltage gradient was the primary modulator of MAPA, another model was constructed, in which $g_{i}^{\lambda;c}$ was unchanged but $g_{i}^{\lambda;c}$ was reduced to reduce the tissue length constant (and therefore $\gamma$). The simulated signals in Fig. 3, B and C, have a similar magnitude of intracellular depolarization and $\gamma$, however, the reduced $g_{i}^{\lambda;c}$ case has an MAPA = 6 mV, compared with the nominal MAPA of 20.9 mV. Clearly, MAPA is most strongly modulated by interstitial resistance $g_{i}^{\lambda;c}$ although the local tissue length constant also plays a role.

It is clear that altering $\Psi_{elec}$ has an effect on MAPA, but there are no direct experimental data to support the possible compression in cardiac tissue. Compression has been reported to cause interstitial fluid to redistribute in fibrosarcomas but only when the applied pressure is above a threshold (38). Grodzinsky et al. (10) have also shown that pressure applied to connective tissue causes a change in passive electrical properties via streaming potentials. Furthermore, we propose that an increase in cellular “packing,” resulting from the constriction of interstitial space, does not reverse within the short time frame of one cardiac cycle. This persistence of the mechanical effect on the membrane and tissue response may be at least partly responsible for the stability of the MAP signal in a vigorously beating heart (7). Future experimental studies on cardiac tissue are needed to test the hypothesis suggested by the model that the applied pressure locally restricts the interstitial space and changes the local interstitial resistance.

The factors that affect the local tissue length constant will impact the correspondence of the MAP and the TAP. One such factor is the total ionic membrane resistance ($G_m$). Because all ionic currents contribute to $G_m$, $g_{press}$ can alter the tissue length constant, and therefore $\gamma$, MAPA, and $V_{max}$. Increasing $g_{press}$ to 4.0 mS/cm² increases $G_m$, leading to a decreased $\lambda$ and $\gamma$ and an increase in MAPA to 24.2 mV and $V_{max}$ to 9.8 V/s. It is important to note that $G_m$, and therefore the tissue length constant and $\gamma$, change during the course of an action potential. In these studies, we have only reported $\lambda$ and $\gamma$ at diastole.

The inclusion of a shaft and compression of interstitial space both act to increase the magnitude of the MAP, however, a large $g_{press}$ may also account for the large MAPA. Simulations resulting in an MAPA on the order of 20 mV without a shaft and no compression required $g_{press}$ to be $\sim$35 mS/cm², an order of magnitude greater than that of stretch-activated channels. As a result, $V_{max}$ increased to 14.8 V/s and the large $I_{press}$ dominated all activity under the electrode such that the amplitude of $\phi_t$ was 1.1 mV.

Slow propagation or increased electrode size increases the time to establish and evolve the boundary current sources during activation and recovery and therefore influence MAPA and $V_{max}$. Knollmann et al. (21) showed that smaller tipped electrodes produce larger MAPA and $V_{max}$ and more accurate MAPD values than larger-tipped electrodes. The values of $\gamma$ do not change significantly with electrode size; however, the effective spatial extent of the electrode is closer to the radius of the electrode plus $\gamma$. Because the propagation front moves at a constant velocity, more time is required to traverse the depolarized region when the electrode is larger. This explanation accounts for the increase in MAPA and $V_{max}$ of smaller electrodes. Similarly, increasing the depth of $I_{press}$ causes the electrode region to be larger, and after a transient increase, MAPA decreases. Fast propagation along fibers produced larger MAPA and $V_{max}$ and a better MAPD/TAPD correlation, whereas slow propagation had the converse effect.

The physical size of experimental and clinical MAP electrodes generally increase as the heart size increases. The results from the simulations on electrode size suggest that the measured MAPAs should be smaller in large animals. This, however, is not the case. Larger amplitude signals are typically measured in larger hearts (7, 8, 21). The results presented here, as well as those from the models of Vigmond and Leon (35) and Hirsh et al. (13), agree that the quantity of tissue and the depolarization depth do not significantly affect MAPA. One explanation for the discrepancy between the experimental and simulation findings may be related to the fraction of interstitial space in the region below the electrode. Because thick-walled hearts require more pressure to achieve a maximal MAPA, the value of $\Psi_{elec}$ may be reduced to a greater extent. It is unlikely that the mechanism responsible for increased MAPA in thicker heart walls lies in a single parameter but rather is a result of several factors, such as intrinsic packing, nonlinearities in stretch-sensitive channels, or the size of the insulating electrode.

**Effects of bath loading.** The results of the simulations without a shaft showed that the MAPA for the two cases are both markedly reduced and closer in value (5.2 mV with compression versus 4.1 mV without compression). The findings suggest that the insulating shaft acts to force the current through the interstitial space under the electrode and increase the amplitude of the MAP.

In contrast to previous simulation studies (35), the results from a full bidomain model presented here show that the properties of the surrounding bath have a significant impact on MAPA. The adjoining bath provides an alternative path for extracellular current to flow away from the tissue surface. In general, the larger the bath conductivity, the smaller the MAPA. This result differs from the model study of Vigmond and Leon (35), who found that the absence of a bath increased

---

**References:**

MAPA by only \( \sim 5\% \). Our results show that removing the insulating shaft from the bath and replacing it with saline cause a 60% decrease in MAPA. Vigmond and Leon (35) also found that limiting values of MAPA are equal to TAPA. The model used by Vigmond and Leon did not explicitly account for an interstitial space at the tissue surface in their model, hence the limiting MAPA being equal to \( V_m \) is consistent with that obtained in a bidomain with no interstitial space. In the full bidomain model without a loading bath, the limiting values of MAPAs approach the core conductor predictions.

**MAPD to TAPD correlation.** It is generally assumed that the time course of the MAP corresponds with the normal TAP that would exist in the absence of the pressure. The parameter variation study showed that changes in the nature or volume of the depolarized region altered the shape of the TAP in the electrode region (TAP\textsuperscript{elec}) and MAP in the same manner. Our model suggests that the MAP time course corresponds most closely to the TAP\textsuperscript{elec}, which has been depolarized due to the pressure. Whereas the amplitudes of TAP\textsuperscript{elec} and full TAP differ significantly, the APD measurements at the two sites differ (in the nominal case) by \( < 1 \) ms (the sampling rate used for most MAP recordings), and therefore the MAPD is a good representation of the normal TAPD in the absence of pressure. Future studies are required to investigate the relationship in regions of heterogeneity in membrane properties.

\( \phi_t \) to \( V_m \) correlation. Using the bidomain model, we have distinguished between the depolarization of the intracellular potential (\( \phi_t - \phi_{\text{ref}} \), where \( \phi_{\text{ref}} \) is the extracellular MAP reference electrode located 4 mm above the tissue) and the depolarization of the transmembrane potential (\( \phi_t - \phi_x \), where \( \phi_x \) is the interstitial potential directly across the cell membrane). Because \( \phi_{\text{ref}} \) and \( \phi_x \) are not equal, \( \phi_t \) and \( V_m \) are not equivalent and depolarized to different degrees. Specifically, for the model discussed here, \( \phi_t \) was always found to be less depolarized than \( V_m \) near the MAP electrode.

**Limitations.** Whereas recent experimental information has allowed for the construction of a more accurate 3-D model, there are still several limitations to these simulation studies. The lack of experimentally measured intrinsic anisotropic conductivities in the mouse RV and the values of \( \Psi_i \), \( \Psi_e \), and \( \Psi_{\text{elec}} \) require that the model match more indirect global measurements such as conduction velocity and MAPA. Conduction velocity, however, also depends on the resting length constant, and cellular upstroke dynamics. Hence, the choice of cellular dynamics will impact the conductivity needed to reproduce a given conduction velocity. These studies also assume that changes in \( I_{\text{press}} \) and \( \Psi_{\text{elec}} \) are spatially abrupt, occur in unison, and are all or none. Whereas a constriction of interstitial space and the opening of stretch receptive channels are impacted by the same mechanical load, the size of the affected region is most likely different and nonuniform for the two phenomena. Additional inhomogeneities may include natural and pathological gradients in \( I_{\text{ion}} \), fiber organization, and conduction velocities and pathways. While we have attempted to more realistically represent the presence of the electrode by including an insulating shaft, the precise weighting of potentials across the tip was not taken into account. The model also assumes the point load pressure and the effect on the interstitial space is the same throughout the cardiac cycle. It is possible that the properties may change as a function of pressure load in the ventricle, but this awaits more detailed experimental study and analysis.

Another limitation is that the tissue was assumed to behave as a uniform bidomain. The impact of changes in the discrete structure, such as increased gap junction resistance or knockout of connexin (17), on MAP shape is not well approximated by the model. On the basis of our studies of propagation across fibers, we expect an increase in intracellular resistance to decrease MAPA and \( V_{\text{max}} \).

Franz has proposed that the offset of the MAP relative to 0 mV may indicate asymmetric local current wave forms (7) leading to a redistribution of charge in space and may account for the degradation of the MAP over time. Although the net charge over the entire domain is zero, the charge accumulation at a location in space can be found as the area under the transmembrane current versus time plot. It is clear that more area lies below zero in Fig. 6, suggesting a negative charge accumulation on the border of the electrode. Further studies with multiple paced beats will be necessary to explore this idea.

Whereas there is good agreement between the model and the experiment using a linear stretch-sensitive channel as the depolarization mechanism and a constriction of interstitial space, there are other physiological parameters that may contribute to the MAP. For example, it is quite possible that multiple stretch/voltage receptors respond in concert to the same mechanical stimulus. Despite these limitations, we believe the mechanism of the clinically recorded MAP is most likely related to the mechanism proposed here, in which the time course follows that of the transmembrane currents formed at the border of a region that is depolarized due to pressure below the contact electrode.

**APPENDIX**

The following parameters were used for the nominal case: \( g_{\text{press}} \) (2.0 mS/cm\(^2\)), \( E_{\text{press}} \) (\( -6 \) mV), \( \sigma_i^0 \) (7.14 mS/cm), \( \sigma_e^0 \) (0.714 mS/cm), \( \sigma_i^0 \) (7.14 mS/cm), \( \sigma_e^0 \) (13.33 mS/cm), \( \sigma_s^0 \) (4.33 mS/cm), \( \sigma_t^0 \) (4.33 mS/cm), \( \Psi_i(0.7) \), \( \Psi_e(0.3) \), \( \Psi_{\text{elec}}(0.1) \), \( \Delta x, \Delta y, \Delta z(50 \mu m) \), \( \Delta t(2 \mu s) \), and \( \beta(1.666 \text{ cm}^{-1}) \).

The values of \( g_{\text{press}} \) and \( E_{\text{press}} \) are based on the measurements of Zeng et al. (37). The bathing conductivity was chosen to simulate the electrical properties of saline. Intrinsic tissue conductivities compare well with those of Kleber and Riegger in Ref. 19 (\( \sigma_i^0 = 6.02 \text{ mS/cm} \), \( \sigma_e^0 = 15.9 \text{ mS/cm} \)) and Clerc in Ref. 2 (\( \sigma_i^0 = 0.28 \text{ mS/cm} \), \( \sigma_e^0 = 7.87 \text{ mS/cm} \)). Kleber and Riegger report \( \Psi_i = 0.25 \) although this value will vary due to structural inhomogeneities and mechanical restriction of the interstitial space. In regions of electrode pressure \( \Psi_{\text{elec}} = 0.1 \).

**GRANTS**

This work was supported by National Heart, Lung, and Blood Institute Grant RO1-HL-63346, National Science Foundation Grant DII-9974533, American Heart Association Grant SDG 0130285N (to B. C. Knollmann), a Pharmaceutical Research and Manufacturers of America Foundation Fellowship (to B. C. Knollmann), and by the North Carolina Supercomputing Center.

**REFERENCES**


