A Role for the Sarcolemmal Na\textsuperscript{+}/H\textsuperscript{+} Exchanger in the Slow Force Response to Myocardial Stretch

Jonathan C. Kentish

Although the contractile performance of the myocardium is under continuous nervous and hormonal regulation, the myocardium possesses a number of intrinsic, load-dependent mechanisms by which it can adjust cardiac output to meet the needs of the circulation over periods ranging from seconds to years. In isolated hearts, an increase in ventricular end-diastolic volume (EDV), produced by increased venous return or decreased aortic outflow, leads immediately to a more powerful contraction via the Frank-Starling mechanism ("heterometric autoregulation"), so that cardiac output increases over a few beats to match venous return. However, over the next few minutes, there is a further increase in myocardial performance, such that EDV returns toward its original value. This second autoregulatory mechanism, the "Anrep effect" or "homeometric autoregulation," allows a given change in cardiac output to be achieved with a smaller change in EDV than if the Frank-Starling effect were the only compensatory mechanism. Finally, if the increase in EDV or wall stress is maintained, genes are switched on that eventually lead to myocardial cell hypertrophy.

Much is known about the cellular and molecular basis of the Frank-Starling mechanism and of the initial stages of load-induced hypertrophy, but the processes responsible for the Anrep effect are poorly understood. In this issue of Circulation Research, Alvarez et al\textsuperscript{1} suggest a novel mechanism for the slow increase in myocardial contractility: a stretch-induced activation of the sarcolemmal Na\textsuperscript{+}/H\textsuperscript{+} exchanger (NHE) by local autocrine/paracrine systems involving angiotensin II (Ang II) and endothelin-1 (ET-1).

The Contractile Response to Stretch of the Myocardium

Our knowledge of the cellular mechanisms involved in the time-dependent increase in contractility after myocardial fiber stretch has come chiefly from studies in which fiber length is controlled (for reviews, see References 4–6). Parmley and Chuck\textsuperscript{7} were the first to show that stretch of isolated papillary muscle leads to a rapid increase in active force (corresponding to the Frank-Starling mechanism in the intact heart), followed by a further increase in force over several minutes (analogous to the Anrep effect). The slow increase in force (here termed the "slow force response") has subsequently been confirmed in a range of isometrically contracting preparations, from isolated myocardial cells\textsuperscript{8} to isovolumic hearts in vitro,\textsuperscript{9,10} and in volume-loaded hearts in vivo.\textsuperscript{11} The immediate increase in force appears to be due to length-dependent properties of the cardiac myofibers, chiefly an increase in their sensitivity to [Ca\textsuperscript{2+}].\textsuperscript{4} In contrast, the slow force response can be explained qualitatively\textsuperscript{8,10,12} and quantitatively\textsuperscript{13} by a slow increase in the magnitude of the intracellular Ca\textsuperscript{2+} transient. However, the mechanism responsible for this slow potentiation of the Ca\textsuperscript{2+} transient has remained obscure. It cannot be explained by a length dependence of sarcoplasmic reticulum (SR) Ca\textsuperscript{2+} handling itself, because the slow force response remains in the presence of specific SR inhibitors.\textsuperscript{13–15} Other mechanisms could involve the observed stretch-induced increases in second messengers such as cAMP\textsuperscript{10} or InsP\textsubscript{3},\textsuperscript{16} although how these could increase the Ca\textsuperscript{2+} transient, in a way that does not depend on the SR, remains to be examined. Activation of myocardial stretch-activated channels (see Reference 17 for review) could enhance the Ca\textsuperscript{2+} transient by increasing (directly or indirectly) diastolic [Ca\textsuperscript{2+}], which would allow the SR to take up and release more Ca\textsuperscript{2+}. However, recent studies\textsuperscript{8,14} have failed to find any increase in diastolic [Ca\textsuperscript{2+}], that could account for the potentiation of the Ca\textsuperscript{2+} transient. Ca\textsuperscript{2+} entry could also be increased if the Ca\textsuperscript{2+} current (I\textsubscript{ca}) of the action potential were stretch sensitive, but this appears not to be the case.\textsuperscript{8} One transport process that has received little attention has been the sarcolemmal NHE, which regulates pH\textsubscript{i} by extruding H\textsuperscript{+} from the cell in a 1:1 exchange with Na\textsuperscript{+} ions. Stimulation of the NHE could potentially increase force, by either (1) increasing cell pH\textsubscript{i}, which would increase myofibrillar Ca\textsuperscript{2+} sensitivity, SR Ca\textsuperscript{2+} loading, and I\textsubscript{ca} (for review, see Reference 18), or (2) increasing cell [Na\textsuperscript{+}], which would tend to reduce Ca\textsuperscript{2+} extrusion from the cell during diastole via forward-mode Na\textsuperscript{+}/Ca\textsuperscript{2+} exchange, and raise Ca\textsuperscript{2+} entry during the action potential via reverse-mode Na\textsuperscript{+}/Ca\textsuperscript{2+} exchange. Stimulation of the NHE has been implicated in the positive inotropic actions of Ang II and ET-1 (reviewed in Reference 19), but evidence that the NHE and these hormones may play a part in the responses of the myocardium to stretch has come from another area of cardiac research: the triggering of cellular hypertrophy by stretch.

The Hypertrophic Response to Stretch of the Myocardium

The past decade has seen major advances in our understanding of the events that couple myocardial stretch to cellular...
hypertrophy. Changes triggered by stretch of myocardial cells include the activation of “intermediate-early” genes, such as c-fos, followed by “late-responsive” genes such as fetal contractile protein genes (eg, those for β-myosin and skeletal α-actinin). Multiple signal transduction pathways are activated, including phospholipases C, D, and A₂, and many types of protein kinase, including protein kinase C, tyrosine kinase, and the Raf-1 and mitogen-activated protein kinase (MAPK) cascades.20–22 The initial response to stretch is rapid: intermediate-early genes and the signaling pathways are switched on within minutes, ie, on the same time scale as the slow force response.

These changes may be due, at least in part, to the stretch-induced activation of local (ie, intracardiac) autocrine systems. It is known that myocardial cells possess local renin-angiotensin23 and endothelin systems.24 Using neonatal rat ventricular myocytes cultured on an elastic silicone substrate, it has been shown that stretch stimulates secretion of Ang II20–22,25 and ET-122,25 and that released Ang II alone,20 or both hormones,22,25 induces activation of MAPK, c-fos, etc. The importance of these autocrine systems in the intact adult heart is less clear, because adult cells have a different expression of genes and a different complement of receptors compared with neonatal cells. In addition, the myocardium possesses endothelial cells and fibroblasts, both of which may release Ang II or ET-1 in response to stretch and so could influence myocardial gene expression via a paracrine mechanism. In vivo, the situation is even more complex, and the hypertrophic effects of other factors such as sympathetic stimulation may become dominant.26

The local stretch-induced release of Ang II or ET-1 by the autocrine/paracrine pathways might be expected to stimulate the NHE, given that exogenous Ang II or ET-1 will do this. Indeed, Yamazaki et al22 recently demonstrated a stretch-induced activation of the NHE that triggered a hypertrophic response via an increase in pHᵢ, although unexpectedly this effect appeared to be independent of autocrinely released Ang II or ET-1. Cingolani’s group has now linked the contractile and molecular effects of myocardial cell stretch, by demonstrating a stretch-induced activation of the NHE that may account for the slow force response.

The Role of the NHE in the Slow Force Response to Muscle Stretch

If stretch activates the NHE, this should tend to increase pHᵢ and [Na⁺]. Cingolani et al27 found previously that stretch of cat papillary muscle did indeed produce a rise of pHᵢ over about 10 minutes. This alkalosis was due to the activation of the NHE, as it was abolished by the NHE blocker EIPA. The essential role of Ang II was shown by the demonstration that the alkalosis was also blocked by the AT₁ antagonist losartan. Interestingly, this seemed not to be a direct effect of Ang II on the NHE, given that the rise in pHᵢ produced by stretch (or by exogenous Ang II) was also inhibited by ET-1 antagonists. Cingolani et al27 suggested that Ang II, released from the papillary muscle (or added exogenously), caused the local release of ET-1, which then activated the NHE, and thereby increased pHᵢ. This agreed with previous studies showing that Ang II could induce the synthesis and/or release of ET-1 from myocardial cells, fibroblasts, or endothelial cells (see References 27 and 28). The conclusions27 were that stretch of an intact cardiac muscle caused the sequential release of endogenous Ang II and ET-1, both of which thus acted in an autocrine/paracrine role, and that the resulting increase in pHᵢ was mediated by stimulation of the NHE by ET-1.

In the present study,3 Cingolani’s group has extended their work by investigating the contractile consequences of this stretch-induced stimulation of the NHE. Using isolated rat trabeculae, they confirmed that muscle stretch increased pHᵢ (measured from all the cells in the preparation) when the muscle was bathed in HEPES-Tyrode’s solution but found that pHᵢ did not change significantly in Tyrode’s solution containing CO₂/HCO₃⁻ buffer (perhaps because an acid-loading Cl⁻/HCO₃⁻ exchanger was activated by Ang II). Because the slow force response was the same under both conditions, it was unlikely to be due to the increase in pHᵢ. There was however a marked increase in [Na⁺], after the stretch, with a time course similar to, or perhaps slightly preceding, the rise of force. The role of the NHE in the rise of both [Na⁺], and force was confirmed by the finding that EIPA inhibited the rise in both measurements. The stretch-induced changes in [Na⁺], and force were also reduced by AT₁ or ET-1 antagonists, confirming the primary roles of both hormones in the ionic and contractile changes. The slow increase in the magnitude of the Ca²⁺ transient was also blocked by these antagonists. Provided that all the antagonists used were acting specifically, the data from both studies3,27 lead to the following suggested mechanism for the slow force response:

\[
\text{Stretch} \rightarrow \text{Ang II} \rightarrow \text{ET-1} \rightarrow \text{Stimulation} \rightarrow \text{release} \rightarrow \text{release} \rightarrow \text{of NHE} \rightarrow \uparrow [\text{Na}⁺] \rightarrow \uparrow [\text{Ca}²⁺] \rightarrow \uparrow \text{Force}
\]

One uncertainty in the above scheme is how exactly the rise in [Na⁺] promotes a rise in the Ca²⁺ transient. Alvarez et al13 detected no increase in diastolic [Ca²⁺], after stretch of the muscles, as found previously,8,14 which suggests that the rise in [Na⁺], does not load the cell with Ca²⁺ by decreasing diastolic Ca²⁺ extrusion via Na⁺/Ca²⁺ exchange; there could be, however, an enhancement of Ca²⁺ influx during the action potential via reverse-mode Na⁺/Ca²⁺ exchange. Interestingly, a recent ionic model of the myocyte29 showed that the latter effect could account for many characteristics of the slow force response (although in the model the rise in [Na⁺], was considered to be most likely due to Na⁺/K⁺ pump inhibition). On the other hand, it is not clear whether the rise in [Na⁺], is a vital step in the slow force response. Some studies have reported that the inotropic effect of Ang II is not due to a rise in the Ca²⁺ transient but to the rise in pHᵢ,30 and in isolated myocytes there was no increase in [Na⁺], during the slow increase in contractile state after stretch.8 From the latter result, it could be suggested that there are at least two mechanisms for the slow force response: an [Na⁺]-independent one seen in isolated myocardial cells and another, conferred by endothelial (endocardial) cells or fibroblasts in cardiac muscle preparations, that acts to increase [Na⁺]. Another source of variability may be that NHE
stimulation can potentially increase force by either of two ionic mechanisms (increase in $[\text{Na}^+]_i$, and thus $[\text{Ca}^{2+}]_i$, or increase in pH), so one or the other of these might predominate during the inotropic response to stretch or Ang II, depending on the experimental conditions. The precise roles of changes in $[\text{Na}^+]_i$, pH, and of other potential mechanisms in the slow force response to stretch will need to be established by further quantitative studies. In this regard, it may be noted that the muscles used in the study by Alvarez et al.\textsuperscript{3,27} were superfused rather than perfused, which might enhance the autocrine/paracrine effects compared with slightly better perfused preparations, such as isolated myocytes and perfused hearts.

A number of other questions remain, including: Are myocardial cells, endothelial cells, or fibroblasts the primary source of secreted Ang II or ET-1? How does stretch cause the release of Ang II, and what is the mechanosensor (stretch-activated channels, cytoskeleton, etc.)? How exactly the release of Ang II, and what is the mechanosensor source of secreted Ang II or ET-1? How does stretch cause myocardial cells, endothelial cells, or fibroblasts the primary contractile state of isometrically contracting preparations and thus, more work is needed before we can fully understand NHE? Are these mechanisms important in the intact heart? (stretch-activated channels, cytoskeleton, etc.)? How exactly the release of Ang II, and what is the mechanosensor source of secreted Ang II or ET-1? How does stretch cause myocardial cells, endothelial cells, or fibroblasts the primary contractile state of isometrically contracting preparations and thus, more work is needed before we can fully understand NHE? Are these mechanisms important in the intact heart? (stretch-activated channels, cytoskeleton, etc.)? How exactly

References


Key words: Na$^+/H^+$ exchanger ★ stretch ★ Anrep effect ★ angiotensin II ★ endothelin
A Role for the Sarcolemmal Na\textsuperscript{+}/H\textsuperscript{+} Exchanger in the Slow Force Response to Myocardial Stretch

Jonathan C. Kentish

doi: 10.1161/01.RES.85.8.658

_Circulation Research_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1999 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circres.ahajournals.org/content/85/8/658

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in _Circulation Research_ can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the _Permissions and Rights Question and Answer_ document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to _Circulation Research_ is online at:
http://circres.ahajournals.org//subscriptions/