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INVITED REVIEW

Role of autocrine/paracrine mechanisms in response to myocardial strain

Horacio E. Cingolani · Irene L. Ennis · Ernesto A. Aiello · Néstor G. Pérez

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Abstract Myocardial strain triggers an autocrine/paracrine mechanism known to participate in myocardial hypertrophy development. After the onset of stretch, there is a rapid augmentation in developed tension due to an increase in myofilament calcium sensitivity (the Frank Starling mechanism) followed by a gradual increase in tension over the next 10–15 min. This second phase is called the slow force response (SFR) to stretch and is known to be the result of an increase in calcium transient amplitude. In the present review, we will discuss what is known thus far about the SFR, which is the in vitro equivalent of the Anrep effect and the mechanical counterpart of the autocrine/paracrine mechanism elicited by myocardial stretch. The chain of events triggered by myocardial stretch comprises: (1) release of angiotensin II, (2) release/formation of endothelin, (3) NADPH oxidase activation and transactivation of the EGFR, (4) mitochondrial reactive oxygen species production, (5) activation of redox-sensitive kinases, (6) NHE-1 hyperactivity, (7) increase in intracellular Na^+ concentration, and (8) increase in Ca^{2+} transient amplitude through the Na^+/Ca^{2+} exchanger. The evidence for each step of the intracellular signaling pathway leading to the development of SFR and their relationship with the mechanisms proposed for cardiac hypertrophy development will be analyzed.

The authors are established investigators of CONICET, Argentina.

H. E. Cingolani (✉) · I. L. Ennis · E. A. Aiello · N. G. Pérez
Centro de Investigaciones Cardiovasculares,
Facultad de Ciencias Médicas, Universidad Nacional de La Plata,
Calle 60 y 120,
1900 La Plata, Argentina
e-mail: cicmes@infovia.com.ar

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Introduction

Although it was known that cardiac hypertrophy was triggered by the intracellular signals that occur following myocardial stretch, investigators working in classic muscle mechanics did not extrapolate their findings regarding stretch-induced cardiac mechanics to the development of myocardial hypertrophy. This could have been because the time frames in which these two phenomena occur are quite different. However, the long journey toward myocardial hypertrophy begins with one step, and that first step may be the autocrine/paracrine (A/P) intracellular signaling pathway triggered by myocardial stretch, which leads to mechanical changes.

After stretch, cardiac muscle increases developed force in two phases. The first phase, which occurs rapidly, is generally attributed to enhanced myofilament responsiveness to Ca^{2+} and is probably not affected by the A/P mechanism. The second phase, known as the slow force response (SFR), occurs gradually and is due to an increase in the Ca^{2+} transient amplitude as a result of the A/P mechanism. The SFR is proposed to be the in vitro form of the Anrep phenomenon. This mechanical response to stretch has been known since 1912—at that time, von Anrep [72] observed that, when the aortic pressure was elevated, the ventricular volume initially increased and then declined toward the starting volume, indicating an increase in myocardial contractility (for a review, see

[14]). Later on, Sarnoff [62] coined the term homeometric autoregulation for this same phenomenon. Soon after, the effect was reported in myocardial strips by Parmley and Chuck in 1973 [53], and it is due to an increase in the Ca^{2+} transient [3, 37]. However, its genesis was unknown until we proposed that NHE-1 activation was the main step in the A/P mechanism, leading to the increase in contractility by increasing intracellular Na^+ [2].

In the following text, we present the evidence that led us to propose the A/P mechanism underlying the SFR, as well as its similarity to the signals that have been described in cardiac hypertrophy development.

Angiotensin II released by myocardial stretch

In 1995, in the hypertrophied myocardium of spontaneously hypertensive rats (SHR), we found evidence of enhanced NHE-1 activity without significant changes in the intracellular pH (pH_i) due to a simultaneous increase in the activity of the acidifying Na^+-independent Cl^-/HCO_3^- exchanger [54]. Later on, in 1998, we demonstrated in feline papillary muscles that one of the earliest intracellular signals following myocardial stretch was the activation of the NHE-1 as the result of an A/P mechanism in which angiotensin II (A2) and protein kinase C were involved [12]. Previously, Sadoshima et al. [61] had reported that the release of A2 stored in cytosolic secretory vesicles was rapidly induced after stretch of neonatal rat ventricular myocytes as they became hypertrophic. The A2 concentration in the surrounding medium was on the order of 0.5–1 nM and the A2 content of the stretched myocytes was approximately half that of the control nonstretched myocytes. In 1999, we reported that the mechanical consequence of the A/P loop was the SFR, Anrep effect [2], or homeometric autoregulation [62].

Figure 1 shows the mechanical response elicited by the A/P mechanism triggered by stretch, as well as its inhibition by the AT1 receptor inhibitor losartan, but not by the AT2 receptor blocker PD123319. The inotropic response to stretch can be mimicked by 1 nM exogenous A2 [15, 56], a concentration of the same order of magnitude as that detected by Sadoshima et al., as we mentioned above. Leri et al. [42], working with adult rat cardiac myocytes, also reported the stretch-induced release of A2.

A very interesting hypothesis about strain and AT1 stimulation, in which AT1 activation was the result of a conformational change induced by stretch, independent of A2 release was proposed by Zou et al. [85]. Interestingly, this conformational modification was also prevented by losartan.

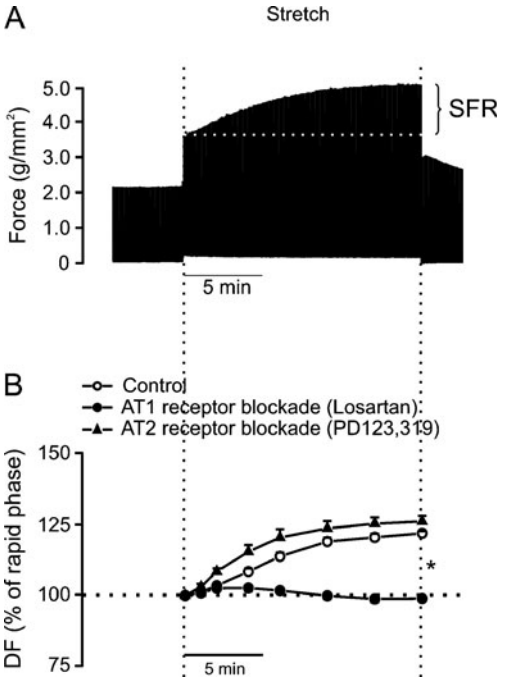

Fig. 1 **a** Original force record from a papillary muscle stretched from 92% to 98% of the length at which maximal twitch force was developed, where it can be appreciated as the biphasic response to stretch. The slow force response to stretch (*SFR*) stabilizes after 10–15 min and is due to an increase in the Ca^{2+} transient. **b** Suppression of the SFR after AT1 but not AT2 receptors blockade (adapted from Caldiz et al. [10])

A2-induced release/formation of endothelin following myocardial stretch

Many cardiovascular effects initially thought to be mediated by A2 have been found to be due to the A/P action of endogenous endothelin (ET)-1 released by A2 instead [32, 44, 52, 59]. In fact, the effects of stretch are mediated by the action of endogenous ET released by A2 since the SFR is blunted not only by AT1 blockade but also by the nonselective ET receptor blocker TAK044 and the ET_A receptor antagonist BQ123, as well as by the inhibition of the ET-converting enzyme with phosphoramidon [55]. It is important to note that the crosstalk between the peptides A2 and ET is unidirectional from A2 to ET and not the other way around since we demonstrated that the effect of exogenous A2 can be blocked by ET blockers, while losartan did not cancel the effects of exogenous ET [12, 56]. We identified by real-time RT-PCR experiments performed in cat papillary muscles the ET isoform(s) that participate in the response to stretch; these experiments showed an increase in ET-3 mRNA after stretch [26]. Provided that the translation efficiency is not altered, the stretch-induced increase in ET-3 mRNA may reflect an increase in ET-3 synthesis secondary to its release, most likely to restore intracellular pools. In connection with this, we should bear in mind that Tamamori et al. [70] reported

that ET-3 was able to trigger the synthesis and release of ET-1, which in turn mediates a hypertrophic response in cultured neonatal cardiomyocytes. Therefore, it is possible, although speculative, that in multicellular preparations stretch triggers ET-3 release, which could be responsible for the inotropic effect and for the sequential release/formation of ET-1, which would induce cell growth. In any case, ET isoforms (ET-1, ET-2, and ET-3) are equipotent with respect to the inotropic response in papillary muscles [26]. We have found further evidence in favor of ET-3 as mediator of the SFR: both the SFR and the ET-3-induced equipotent inotropic effect were sensitive to a low dose of BQ123 (0.3 μM), while a similar inotropic effect induced by ET-1 was not [26, 60].

In isolated cat ventricular myocytes, the increase in contractility induced by a low concentration (1 nM) of A2 (mimicking the SFR) is completely prevented by ET receptor blockade, supporting an autocrine interaction between these two hormones (Fig. 2) [15]. When myocytes are exposed to a higher concentration of A2 (100 nM), the increase in contractility is greater and it is only partially (by approximately 30%) inhibited by ET receptor blockade [15]. Therefore, if we assume that there are no other cell types in the suspension that could potentially participate in this mechanism, these findings suggest that the increase in contractility induced by 1 nM A2 is entirely due to an autocrine pathway involving ET release.

In connection with this, experiments performed by Anderson et al. [5], designed to explore BNP synthesis in cardiac hypertrophy, demonstrated that myocardial stretch induces ET-1 release/formation in an NADPH oxidase-dependent fashion in isolated neonatal rat ventricular myocytes. Unfortunately, the authors did not explore the upstream role of A2 at that time.

Transactivation of epidermal growth factor receptor

Over the past few years, a role for receptor tyrosine kinases such as the epidermal growth factor receptor (EGFR) due to transactivation by G protein-coupled receptor agonists (i.e., A2 and ET-1) has been reported in the myocardium [5, 20, 47, 66, 71, 80]. Furthermore, it was reported that, in transgenic mice with cardiac-specific overexpression of a mutant AT1 that is incapable of transactivating the EGFR, the development of pathologic cardiac hypertrophy is inhibited [80]. Furthermore, recent experiments performed in cultured neonatal rat cardiomyocytes showed that the EGFR is transactivated during stretch [5, 20].

Because many of the intracellular signals found to play a role in the SFR are also associated with cardiac hypertrophy development after stretch, we decided to test whether EGFR transactivation played a role in the SFR. Figure 3 shows that the SFR was completely abolished either by inhibiting Src tyrosine kinase, one of the proposed mediators of the EGFR transactivation, with PP1 or by preventing the release of the heparin-binding epidermal growth factor (HBEGF) by inhibiting matrix metalloproteinase activity, another proposed mediator of EGFR transactivation. Interestingly, Anderson et al. [5] demonstrated that the ET generated in response to myocardial strain induces metalloproteinase-mediated cleavage of pro-HBEGF and ectoshedding of HBEGF, a ligand of EGFR. Moreover, the antioxidant *N*-acetylcysteine and the NAD(P)H oxidase inhibitor apocynin inhibits the strain-dependent activation of the ET-1 promoter and HBEGF shedding. As expected, the SFR is also abolished after specific EGFR inhibition with AG1478, which prevents receptor phosphorylation (Fig. 3).

Reactive oxygen species production induced by the A/P loop

The main cardiac sources of reactive oxygen species (ROS) in the myocardium are NADPH oxidase, xanthine oxidase, uncoupled nitric oxide synthase, and the mitochondrial respiratory chain. Both A2 and ET-1 are well-known activators of NADPH oxidase and therefore inducers of anion superoxide formation. Because myocardial stretch releases A2, which in turn induces ET release/formation, both leading to the activation of NADPH oxidase, increased ROS production is to be expected after stretch. Indeed we have recently shown that myocardial stretch increases ROS formation, while pre-incubation with two well-known ROS scavengers like EUK8 and *N*-2-mercaptopropionyl-glycine (MPG) blunted ROS increase and inhibited SFR development [10]. In the same experiments, the SFR was also abolished by two different NADPH oxidase inhibitors, diphenyleneiodonium and apocynin, as well as by the mitochondrial K_{ATP} channel inhibitors 5-HD and glibenclamide, implicating both the NADPH oxidase and the mitochondria as the source of ROS [10]. These data are in line with the ROS-induced ROS release phenomenon proposed by Zorov et al. [84] and Kimura et al. [38], and they provide support for a crucial role for ROS as intracellular signals in determining the Anrep effect. However, elucidation of the precise function played by ROS in each of the several steps of the A/P mechanism requires further research. Superoxide anion is very unstable, being rapidly converted to H_2O_2 by superoxide dismutase; therefore, it is possible that H_2O_2 is the molecule involved in intracellular signaling.

In summary, the A/P loop triggered by myocardial stretch induces ROS formation that originates in the mitochondria and prevention of ROS activity is sufficient to blunt the SFR. Although ROS were classically

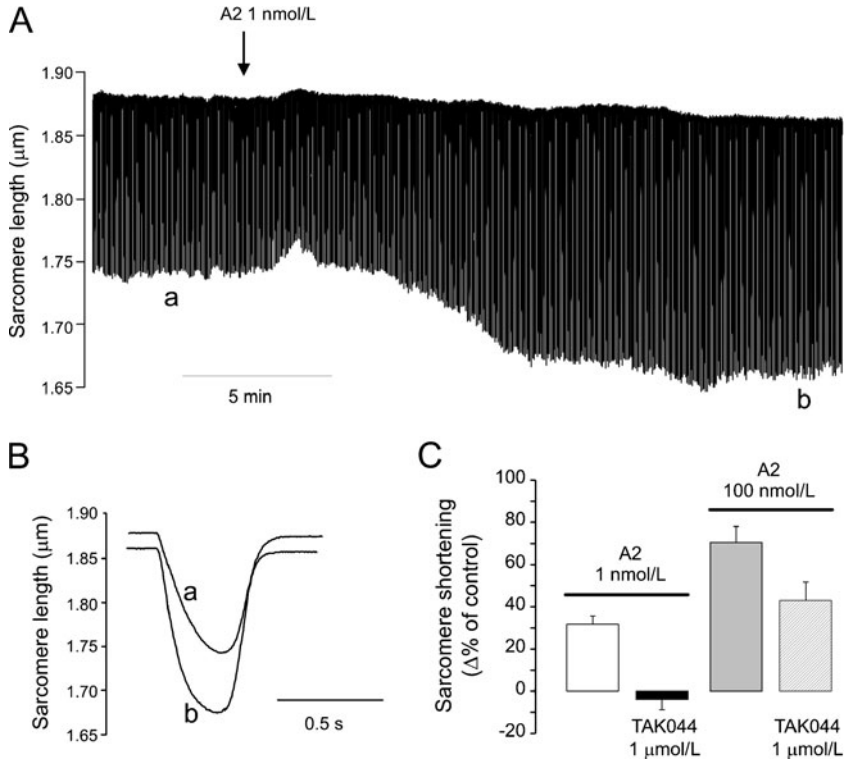

Fig. 2 A low dose of angiotensin II (*A2*, 1 nmol/L) elicited a significant increase in sarcomere shorting in isolated cat cardiomyocytes as shown in the typical records of **a** and **b**, an effect that was prevented by blocking the ET receptors with TAK044 (**c**). This low dose of A2 is equipotent to the inotropic effect induced by stretch, the SFR. Note that cancelation of the A2 effect by ET rceptors blockade is only valid for the low dose of the peptide since increasing A2 dosage increases inotropism through mechanisms other than ET release (**c**). If we assume that under the present experimental conditions there is no participation of cells other than myocytes, we can conclude that we are in the presence of an autocrine mechanism where the myocyte is source and target of ET

considered to be deleterious molecules, there is a growing body of evidence to support the notion that they can also function as intracellular signaling molecules, depending on the magnitude, duration, and timing of the redox signal [19]. Interestingly, a role for ROS in myocardial hypertrophy development has been proposed repeatedly [4, 18, 40, 43, 48, 49, 58, 64, 65, 68, 69, 77].

NHE-1 as a critical step of the A/P loop that induces the SFR

NHE-1 is a ubiquitously expressed membrane protein that exchanges intracellular H^+ for extracellular Na^+ and is one of the main pathways through which Na^+ enters the cell. The protein has two major domains: an NH_2-terminal membrane transport domain and a carboxyl-terminal cytosolic regulatory domain. NHE-1 plays a critical role in a number of cardiovascular disorders including ischemia/reperfusion injury, cardiac remodeling after myocardial infarction, and the development of pathologic cardiac hypertrophy [13, 35, 36]. As discussed above, in 1995, we reported an enhanced activity of NHE-1 in hypertrophied myocardium of the SHR [54]. Moreover, Nakamura et al. [51] demonstrated recently in elegant experiments that increased NHE-1 activity is sufficient to generate the Ca^{2+} signals necessary to induce cardiac hypertrophy. Interestingly, NHE-1 inhibition exerts beneficial effects in these circumstances [6, 11, 17, 23, 24, 78].

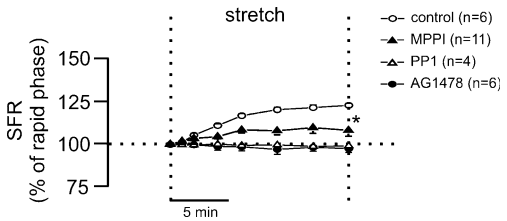

Fig. 3 Transactivation of the EGFR underlies the SFR. Averaged results for the SFR from control-stretched papillary muscles and muscles pretreated with a matrix metalloproteinase inhibitor (*MMPI*), a Src kinase inhibitor (*PP1*), or an EGFR blocker (*AG1478*) to cancel EGFR transactivation. All pharmacological interventions prevented the development of the SFR to stretch (adapted from Villa-Abrille et al. [71] with permission)

One of the mechanisms by which NHE-1 activity is increased is by phosphorylation of its cytosolic tail, especially at serine 703. The upstream kinases activating the exchanger appear to be ERK1/2 and $p90^{RSK}$. These are redox-sensitive kinases and are activated by stretch. As shown in Fig. 4, the A/P loop triggered by myocardial stretch increases ERK1/2 phosphorylation. The increased ERK1/2 phosphorylation is abolished by blocking the AT1 receptor with losartan, the first step in the stretch chain of events [10]. In line with this effect, inhibition of MEK, a kinase upstream ERK1/2, cancels the SFR, as shown in Fig. 4b. The stretch-induced increase in ERK1/2 phosphorylation is also prevented by impeding EGFR transactivation with two different pharmacological interventions: blocking the Src kinase with PP1 and inhibiting the EGFR with AG1478

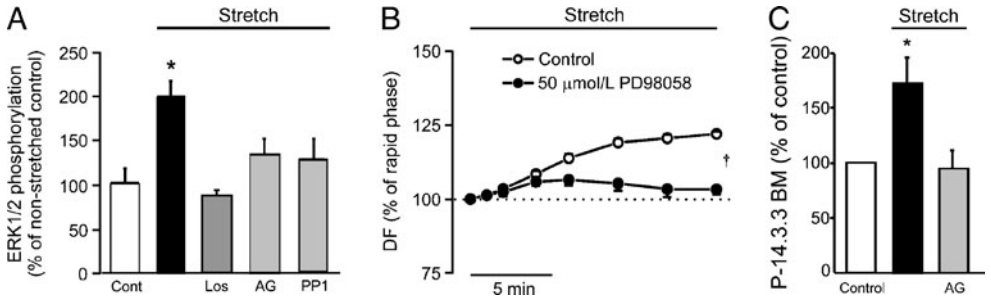

Fig. 4 **a** Myocardial stretch significantly increased ERK1/2 phosphorylation, an effect that was canceled by blocking the AT1 receptor with losartan (*Los*) or the EGFR with AG1478 (*AG*) or by Src kinase inhibition with PP1. **b** Inhibition of MEK (kinase upstream ERK1/2) by the specific inhibitor PD98059 canceled the SFR. **c** Stretch increased NHE-1 phosphorylation, an effect that was canceled by preventing EGFR activation with AG (reproduced from Caldiz et al. [10] and Villa-Abrille et al. [71] with permission)

[71]. Furthermore, exogenous EGF increases ERK1/2 phosphorylation, an effect that is canceled by preventing ROS formation [71]. These experiments, performed in adult multicellular preparations, are in line with previous reports in isolated neonatal cardiomyocytes focusing on cardiac hypertrophy development [5, 20].

Berk et al. [41] showed that NHE-1 phosphorylation at Ser 703 by $p90^{RSK}$ creates a binding motif for the chaperone protein 14:3:3 and offers a simple way of assessing phosphorylation of the exchanger by immunoblot. Using this method with an antibody against the phospho-Ser 14-3-3 binding motif in immunoprecipitated NHE-1, we detected that myocardial stretch increases NHE-1 phosphorylation (Fig. 4c). Interestingly, this does not occur in the presence of AG1478, a selective inhibitor of EGFR [71]. Moreover, when we incubated rat myocardium with A2 at a concentration that evokes an increase in developed force similar to that of the SFR, we induced ROS production, which was accompanied by the activation of ERK1/2 and $p90^{RSK}$ and phosphorylation of Ser703 of the carboxylic tail of NHE-1. These effects were prevented by losartan and the ROS scavenger MPG [28].

Why does an increase in NHE-1 activity induce the SFR development? Role of the Na^+/Ca^{2+} exchanger

Hyperactive NHE-1 extrudes H^+ and allows Na^+ to enter the cell. Thus, we would expect an increase in pH_i and in intracellular Na^+ concentration, respectively, as consequences of enhanced NHE-1 function. However, this happens only under non-physiological conditions, in which bicarbonate-dependent pH_i regulatory mechanism are not active [12, 54]. In contrast, under physiological conditions, pH_i changes are prevented by a simultaneous activation of the acidifying anion exchanger increasing intracellular H^+ [2, 55]. However, because of the Na^+ independence of the acidifying anion exchanger, the increase in Na^+ is not corrected [2, 55]. Unfortunately, these concepts are not often considered or they are misinterpreted. It has been erroneously pointed out that increased NHE-1 activity under physiological conditions induces an increase in both intracellular Na^+ and pH_i [74]. Interestingly, the increase in intracellular Na^+ that occurs in response to stretch, as well as the SFR, is prevented by scavenging ROS with MPG or by blocking NHE-1 with cariporide [10]. The role of the NHE-1-dependent increase in intracellular Na^+ concentration favoring the reverse mode of the Na^+/Ca^{2+} exchanger (NCX) has been confirmed by several authors [9, 46, 73]; however, NHE-1 activation after stretch and the mechanism that leads to the increase in intracellular Na^+ have been challenged [39, 75].

Experiments performed by Allen and Kurihara in 1982 [3] and later confirmed by other authors, including us [2, 37], demonstrated that the SFR was due to a progressive increase in the calcium transient. The phenomenon received considerable attention, but the source and mechanism responsible for the calcium transient increase are still under debate. Neither L-type Ca^{2+} currents [29] nor the sarcoplasmic reticulum [8, 30, 37] seem to contribute to the increase in calcium transient during the SFR. In 2001, Vila Petroff et al. [57] proposed that stretch activates the PI-3 kinase pathway to phosphorylate endothelial nitric oxide synthase; then, nitric oxide stimulates Ca^{2+} release from the sarcoplasmic reticulum, promoting the SFR. Unfortunately, this finding could not be reproduced by other authors in papillary muscles or isolated myocytes [9, 81]. Indeed several authors reported that the SFR occurred even after SR inhibition [8, 29, 37].

We proposed that the stretch-triggered A/P loop leading to NHE-1 activation was responsible for the increase in intracellular Na^+ concentration after finding that an approximately 3–6-mM increase in intracellular Na^+ concentration was induced by stretch [2, 55]. This increase in intracellular Na^+ was enough to shift the estimated reversal potential of the NCX to more negative

values, allowing the operation of the NCX reverse mode during a longer fraction of the action potential. In addition, Lu et al. [45] showed that the redox-sensitive $p90^{RSK}$ prolongs cardiac repolarization by inhibiting outward K^+ channel activity, further increasing the time during which the NCX reverse mode is active. On the other hand, the rise in intracellular Na^+ would reduce Ca^{2+} extrusion through the action of forward-mode NCX during diastole. Moreover, direct phosphorylation of NCX by redox-sensitive kinases may also be contributing to Ca^{2+} entry into the cell after stretch [1, 22, 33]. In fact, inhibition of NCX by equimolar replacement of extracellular Na^+ by either *N*-methyl D-glucamine (NMG) or LiCl cancels the SFR.[55] Lithium is transported by NHE-1, but not by NCX, while NMG is not carried by either transporter [27]. In agreement with these results, inhibition of reverse-mode NCX by 5 μM KB-R7943 canceled the SFR [55].

In summary, it is possible that the reverse mode of cardiac NCX is modulated by myocardial stretch or, equivalently, by the A2/ET network through the three different ROS-dependent pathways: (a) a Na^+-independent and protein kinase C-dependent pathway by direct stimulation of NCX; (b) a Na^+-dependent pathway, consistent with a negative shift in the NCX reversal potential after a rise in intracellular Na^+ due to NHE-1 activation; and (c) a prolongation of the action potential duration. All of these intracellular mechanisms appear to be contributing in concert to the increase in Ca^{2+} after stretch. Note that all of these effects are redox-sensitive and inhibited by blocking the effects of A2/ET.

From the Anrep phenomenon to myocardial hypertrophy

Figure 5 shows a cartoon with the A/P mechanism triggered by myocardial stretch, beginning with the release of A2 and ending with the SFR. A high similarity between the chain of events involved in the genesis of SFR and some of the intracellular pathways proposed to underlie the development of cardiac hypertrophy is evident. In addition, some of the agents used to block the SFR were used in the past, or are currently used, to treat cardiac hypertrophy. However, it is possible that certain intracellular signals triggered by stretch persist for only a limited period of time and, as suggested by Vatner's group [50], perhaps the ideal therapeutic strategy for hypertensive heart disease should be to identify and target the detrimental components while preserving the beneficial ones. The identification of which signals are detrimental and which ones are beneficial is out of the scope of the present review.

Contradictory results from other laboratories

Although a role for NHE-1 in the development of the SFR has been detected in cat [10, 55], human [73], rabbit [46], and rat [2, 9] myocardium, we are not aware of experiments demonstrating its contribution in mice. Involvement of stretch-operated channels in the stretch response was recently proposed in mouse ventricular muscle by Ward et al. [75]. These investigators, in addition to suppressing the SFR by inhibiting stretch-operated channels, were unable to

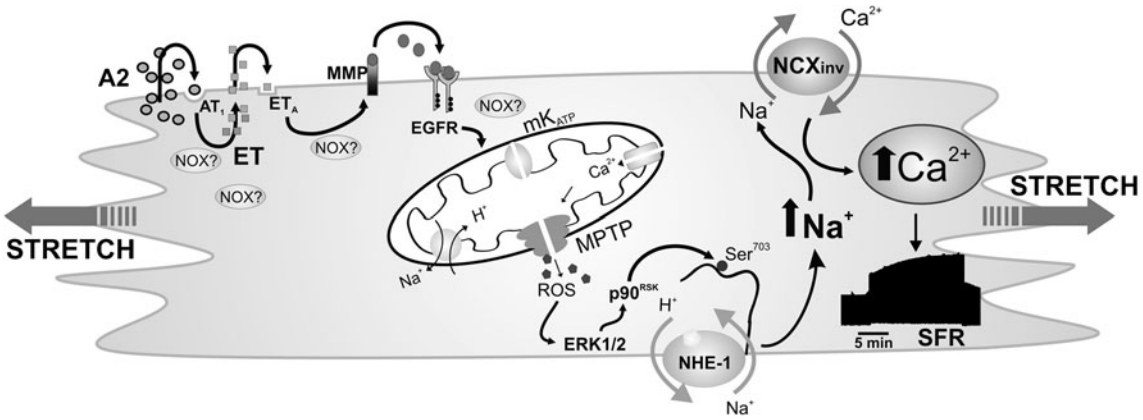

Fig. 5 Schematic representation of the chain of events triggered after myocardial stretch. Stretch and/or A2 through AT1 receptor stimulation trigger the release/formation of ET. NADPH oxidase-generated reactive oxygen species (*ROS*) probably play a significant role in this step of the signaling cascade. ET activates the ET_A receptor and induces epidermal growth factor receptor (*EGFR*) transactivation. The communication between the ET_A receptor and the EGFR involves matrix metalloproteinase (*MMP*) activation and possibly the ROS-sensitive Src kinase. EGFR activation triggers an intracellular signaling pathway that leads to mK_{ATP} channel opening, increasing ROS production and release through the mitochondrial permeability transition pore (*MPTP*). This causes ERK1/2 activation and NHE-1 phosphorylation at Ser703, increasing NHE-1 activity and intracellular Na^+ concentration. The latter favors intracellular Ca^{2+} accumulation through the Na^+/Ca^{2+} exchanger (*NCX*). The increase in ROS production can also contribute to the increase in intracellular Ca^{2+} through a direct stimulatory effect on the NCX [22] and by increasing action potential duration [83] (reproduced from Villa-Abrille et al. [71] with permission)

inhibit the SFR using NHE-1 inhibitors. However, the same authors recently proposed a model of stretch-triggered signals in cardiac myocytes in which release of A2, ROS formation, and NHE-1 activation were all involved, together with stretch-operated channels [74]. Similarly, Kondratev et al. [39], working in mouse ventricular myocytes, reported that cariporide cannot prevent the rise in Na^+ after stretch but cancels the increase in Ca^{2+}; they proposed that stretch-operated channels, and not NHE-1, were responsible for the increase in Na^+ after stretch. Stretch-operated channels allow either Ca^{2+} or Na^+ entry, which would permit a direct increase in Ca^{2+} transient or a Na^+-dependent NCX-mediated Ca^{2+} increase. This could conceivably explain the SFR. A role for stretch-operated channels was previously suggested by Calaghan and White in 2004 [9], but their role was in addition to NHE-1 activation. However, the pharmacological strategy used to inhibit these channels was challenged based on the possible actions of the inhibitors used on the NCX [82]. Moreover, although the existence of these stretch-operated channels in the myocardium has been confirmed by studies recording whole-cell currents in isolated cells [7, 21, 31, 34, 63], they have never been patch-clamped in adult ventricular myocytes [79], presumably because of their restriction to T-tubules.

Recently, canonical transient receptor-operated channels (TRPC) were shown to be sensitive to stretch in mice myocardium [75] and to A2 in human coronary artery smooth muscle cells [67]. Interestingly, it was proposed that TRPC channels were necessary mediators of pathological cardiac hypertrophy in mice, in part through calcineurin-NFAT signaling [76], a pathway that we have shown to be sensitive to NHE-1 inhibition in the rat [25]. The discrepancy in these results could be explained by two alternative hypotheses: (1) the TRPC channels were involved in one or some of the steps in the chain of events described previously, i.e., to induce A2 release after stretch or (2) by species differences.

In connection with the persistence or not along time of some early intracellular signals triggered by the A/P mechanism, such as NHE-1 activation, we recently induced cardiac hypertrophy by transverse aortic constriction in mice. After 7 weeks of aortic constriction, cardiac hypertrophy and decreased myocardial performance were detected along with increased activity of redox-sensitive $p90^{RSK}$ and NHE-1 phosphorylation. AT1 blockade with losartan prevented $p90^{RSK}$ and NHE-1 activation and decreased hypertrophy development, preserving contractility in spite of a higher workload [16].

Concluding remarks

Most intracellular pathways leading to cardiac hypertrophy are triggered by increases in intracellular calcium levels. Actually, this rise in calcium occurs after cardiac muscle is stretched for 10–15 min, causing the SFR or Anrep effect. Yet it seems surprising that most investigators in the field of excitation contraction coupling and cardiac mechanics have not established a link between the Anrep effect and cardiac hypertrophy. Years ago, we proposed the crucial role of the NHE-1 in SFR development; more recently, an elegant paper by Wakabayashi's group showed that NHE-1 activation is sufficient to generate calcium signals that induce cardiac hypertrophy and heart failure [51].

An attractive hypothesis, albeit speculative, will be that the fate of the myocardium could be determined during the first few minutes after it is stretched (i.e., whether or not a pharmacological intervention that prevents the Anrep effect from occurring will subsequently blunt the subsequent hypertrophy and failure).

Approximately 23 million people are afflicted with heart failure, and 2 million new cases of heart failure are diagnosed each year worldwide. All these basic studies mentioned in this review need to be considered when designing new therapeutic strategies in the treatment of cardiac hypertrophy and failure. Understanding the early triggering mechanisms that stretch imposes to the myocardium will allow us to design novel weapons to win the battle against this major disease.

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