Korean J Physiol Pharmacol
Vol 4: 417–425, October, 2000

Comparison of the Determinants in the Differences in Force-Frequency Relationships between Rat and Rabbit Left Atria

Chang Mann Ko and Soon-Jin Kim

Department of Pharmacology and Institute of Basic Medicine, Wonju-College of Medicine, Yonsei University, Wonju 220–701, Korea

The underlying mechanism commonly applicable for both the positive and negative force-frequency relationships (FFR) was pursued in left atria (LA) of rat and rabbit. The species differences in the roles of Na^+/Ca^{2+} exchanger and sarcoplasmic reticulum (SR), which are major intracellular Ca^{2+} regulatory mechanisms in the heart, were examined in the amplitude accommodation to the frequency that changed from 3 Hz to the variable test frequencies for 5 minutes in the electrically field stimulated left atria (LA) of rat and rabbit. Norepinephrine strongly increased the frequency-related amplitude accommodation in both of rat and rabbit LA, while monensin, oubain or the reduced Na^+ and 0 mM Ca^{2+} containing Tyrode solution increased the frequency-related amplitude accommodation only in the rabbit LA. Monenisn was also able to increase the frequency-related amplitude accommodation only in 1-day old rat LA but not in 4-week old rat LA that had 75% less Na^+/Ca^{2+} exchanger with 97% higher SR than 1-day old rat LA. Taken together, it is concluded that the differences in the prevalence between myocardial Na^+/Ca^{2+} exchanger and SR in the amplitude accommodation to the frequency-change determine the difference in the FFR between rat and rabbit heart.

Key Words: Na^+/Ca^{2+} exchange, Sarcoplasmic reticulum, Force-frequency relationship, Monensin, Norepinephrine, Ryanodine, Left atria, Rabbit, Rat

INTRODUCTION

A sudden increase in stimulus frequency on hearts shifts the Ca^{2+} balance and the developed force, and therefore changes "force-frequency relationship (FFR)". It has long been known that the myocardial FFR differs in rat and rabbit. In the rabbit heart, the developed force increases as the stimulation frequency-increases (positive FFR), while it decreases in the rat heart (negative FFR). The underlying mechanism for this discrepancy is still unknown.

After frequency-increase, two major changes are induced in the heart. First, the free myoplasmic Na^+ concentration ($[Na^+]_i$) is increased by the more frequent opening of voltage-dependent Na^+ channels (Cohen et al, 1982; Wier & Yue, 1986; Boyet et al, 1987; Harrison et al, 1992; Lostan et al, 1995; Maier et al, 1997). Second, the sarcoplasmic reticulum (SR) loads less Ca^{2+} in the short time available and therefore releases less Ca^{2+} (Orachard & Lakatta, 1985; Frampton et al, 1991; Bers et al, 1993; Satoh et al, 1997). The $[Na^+]_i$ increase has been claimed to be the mechanism for the positive FFR, as it slows or reverses the Ca^{2+} extrusion to evoke a secondary Ca^{2+} gain by changing Na^+/Ca^{2+} exchange after frequency-increase (Wang et al, 1988; Harrison & Boyet, 1995). And the frequency-induced changes in the Ca^{2+} handling of SR has been suggested as a responsible mechanism for the negative FFR (Orchard & Lakatta, 1985; Bers et al, 1993; Satoh et al, 1997). Recently in a trial to pursue a certain mechanism commonly applicable to these two different FFRs, the phospholamban, a protein that regulates SR Ca^{2+}-ATPase, has been suggested as a common determinant of both FFRs (Kadambi et al, 1999; Bluhm et al, 2000).

Corresponding to: Chang Mann Ko, Department of Pharmacology, Wonju-College of Medicine, Yonsei University, 162 Ilsan-Dong, Wonju, Kangwon-Do 220-701, Korea. (Tel) +82-371-741-0301, (Fax) +82-371-742-4966, (E-mail) changmko@wonju.yonsei.ac.kr

However, search for the exact mechanism or mechanisms of the FFRs has become more confusing after it became clear that $[Na^+]_i$ also increased according to the frequency-increase in the rat heart (Frampton et al, 1991; Hattori et al, 1991; Hussain et al, 1997).

As Na^+/Ca^{2+} exchange and SR Ca^{2+}-ATPase compete for the internal Ca^{2+} (Orachard & Lakatta, 1985; Terracciano et al, 1998), the ratio between Na^+/Ca^{2+} exchange and SR Ca^{2+}-ATPase is very important for myocardial Ca^{2+} balance. If there are simultaneous decreases in the activities of both Na^+/Ca^{2+} exchange and SR Ca^{2+}-ATPase as in the case of frequency-increase, the result will be drawn by the sum of changes attained in these two. Myocardial Ca^{2+} balance after frequency-change will shift to the direction governed by the prevalent one between these two. Therefore, the present study aimed to clarify the mechanism simultaneously applicable for both of the positive and the negative FFR by examining the differences in the prevalence between Na^+/Ca^{2+} exchanger and SR in the heart from rat and rabbit during frequency-changes. In order to achieve this goal, it was investigated if there were any species differences in the influences of Na^+/Ca^{2+} exchanger and SR on the accommodation of LA twitch amplitudes to the given frequency differences during the 5-minute-stimulation frequency changed from the 3 Hz to the variable test frequencies, in the electrically field stimulated left atria (LA) of rat and rabbit.

METHODS

Sprague-Dawley rats of either sex weighing 250 g were sacrificed by decapitation and New Zealand white rabbit of either sex weighing 2.5~3 kg were sacrificed after a blow at the occipital eminence. The hearts were quickly excised and suspended in physiological salt solution (PSS) bubbled with mixed gases of 95% O_2 and 5% CO_2. Krebs Hensleit buffer (KHB) was used for the rat, and the Tyrode solution was used for the rabbit. The composition of PSS's was as follows (mM); KHB solution: NaCl 118.8, KCl 4.70, $CaCl_2$ 2.52, $MgSO_4$ 1.16, $NaHCO_3$ 24.88, KH_2PO_4 1.18, Glucose 5.55, Na-Pyruvate 2.0, and Tyrode solution: NaCl 145, KCl 4.7, $CaCl_2$ 3.0, $MgSO_4$ 1.2, KH_2PO_4 0.6, Glucose 5.0, $NaHCO_3$ 7.0. LA were cautiously dissected and mounted in 8 ml organ baths. LA contraction was evoked by electrical field stimulation delivered through platinum electrodes in square wave pulses of 0.5 msec duration with supramaximal voltages by digital stimulator (STM-1000, Hansung, Korea). The PSS was changed at intervals of 10 minutes throughout the experiment, if not mentioned. LA contraction was recorded on Polygraph (Model 7, Grass, USA) via force displacement transducer (FT. 03, Grass, USA). The resting stimulation was applied at 3 Hz in both LA from rat and rabbit throughout the experiment in order to maintain the same level of SR Ca^{2+} store, as the stimulation frequency affects the SR Ca^{2+} repletion (Frampton et al, 1991). After equilibration at resting frequencies for 30 minutes in normal PSS, NE 10^{-6} M was applied and the resulting maximal amplitudes were measured. After re-equilibration at the resting 3 Hz for 20 minutes, the stimulation frequencies were abruptly changed to the test frequencies for 5 minutes and returned to the resting frequencies to equilibrate again. The test frequency was applied at 0.1, 0.3, 0.5, 0.7, 1, 2, 4, 6, and 8 Hz using the same protocol. During the test frequencies, the changes in LA twitch amplitudes were recorded. The maximal amplitudes appeared within 30 sec (the PEAK) and the last amplitudes obtained at the end of the 5 minutes (the LAST). They were calculated as % to the NE 10^{-6} M-induced maximal amplitudes attained during 3 Hz stimulation. The FFR's of the PEAK and the LAST were plotted.

In the case of drug application, the test frequency was applied 1 minute after the drug effect was stabilized (usually around 5 minutes) except ryanodine that was pretreated for 40 minutes. In the case of Na^+ reduction and Ca^{2+} depletion (RNa-0Ca PSS), NaCl was substituted with equimolar Choline Chloride, $CaCl_2$ was just omitted, and 2 mM EGTA was added (The actual Na^+ concentrations were 4.6% of the normal Tyrode solution for the rabbit and 18.4% of the normal KHB for the rat, since there still were 7 mM of $NaHCO_3$ in the Tyrode and 24.88 $NaHCO_3$ +2 mM Na-Pyruvate in the KHB.). The organ bath was changed into RNa-0Ca PSS and the frequency was changed to the test frequencies without any delay for minimal duration just required to recognize the PEAK (around 1 minute). Right after, the buffer and the stimulation frequency was returned to normal PSS and to 3 Hz. Prior to RNa-0Ca PSS application at each test frequencies, its own control experiments with normal PSS were held at the same test frequencies for about 2 minutes in order to recognize the

PEAK and to check up any critical Ca^{2+} re-application-induced damages in the LA strips. In this case, the PEAK attained after RNa-0Ca PSS application was calculated as the net % increases to its own control.

RESULTS

Traditional investigations on the myocardial FFR have been held either by increasing the stimulation frequency from the lowest to the highest or *vice versa*. In the present study, the stimulation frequency was changed from the fixed 3 Hz to the variable test frequencies for 5 minutes, and the amplitude accommodations to the given test frequencies were measured in the LA from rat and rabbit. This protocol seems to be beneficial as it enables to show the frequency-difference dependent changes in the amplitude accommodations on the basis of constant level of initial SR Ca^{2+} loads (Frampton et al, 1991). By using this protocol, the species differences in the influences of Na^{+}/Ca^{2+} exchanger and SR on the LA amplitude accommodations between rat and rabbit were compared.

As shown in Fig. 1A & B, the LA twitches elicited typical 3-phased changes right after the frequency reduction from the resting 3 Hz to 0.3 Hz: the initial rapid increase to the peak within 30 sec, the rapid decrease, and the following slow decrease to the plateau in both of the rat and rabbit. The first- and the second-phase were much steeper in the rabbit LA, showing higher increase in the PEAK, and larger difference between the PEAK and the LAST than in the rat LA. Fig. 1C & D show the changes of test frequency-increase related LA amplitude changes. In the rat LA (Fig. 1C), both FFR's of the Peak and the LAST decreased inversely proportional to the test frequency-increase (except the PEAK at 6~8 Hz), showing negative FFR's. The differences between the

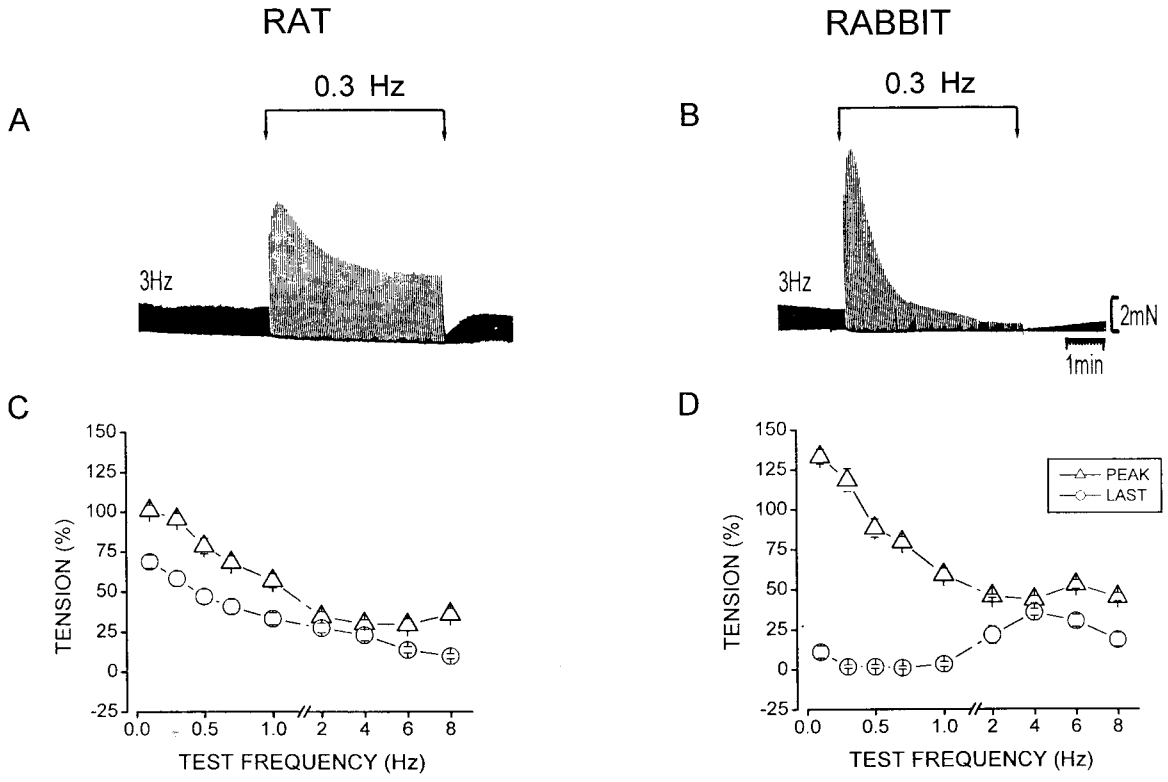

Fig. 1. Amplitude accommodations to the stimulation frequency change in the left atria from rat and rabbit. Left atrial contraction was evoked by electrical field stimulation (0.5 msec duration, supramaximal voltage) through platinum electrodes. Stimulation frequency was changed from 3 Hz to the test frequencies for 5 minutes. A & B. Actual tracings. C & D. Test frequency-related changes. PEAK: the maximal amplitude attained during test frequency, LAST: the last amplitude at the end of 5-minute-test frequency. Values are Means±S.E.M. Numbers of data are 7.

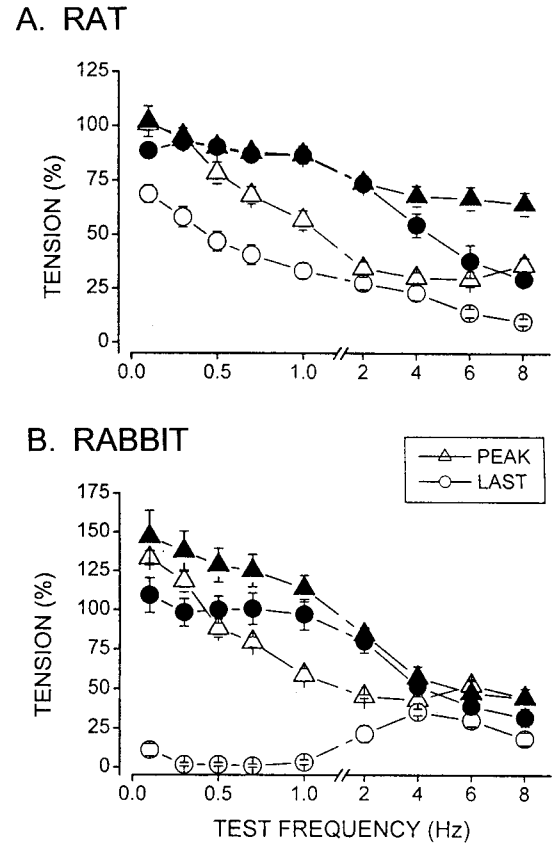

Fig. 2. Effects of norepinephrine (10^{-6} M) on the test frequency-related changes in the amplitude accommodation in the left atria from rat (A) and rabbit (B). (Open symbol: Control, Closed symbol: Norepinephrine). Other legends are same as Fig. 1.

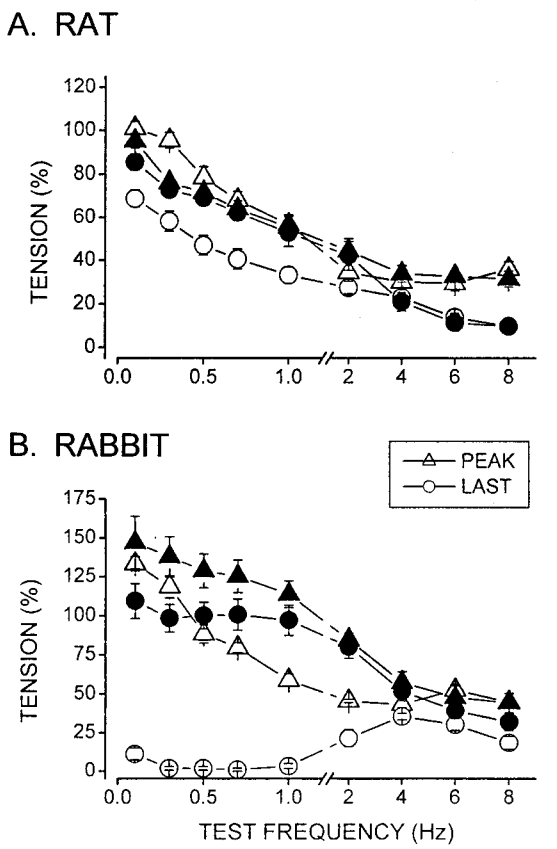

Fig. 3. Effects of monensin (3×10^{-6} M) on the test frequency-related changes in the amplitude accommodation in the left atria from rat (A) and rabbit (B). (Open symbol: Control, Closed symbol: Monensin). Other legends are same as Fig. 1.

PEAK and the LAST were less than 30%. However, in the rabbit LA (Fig. 1D), the LAST showed positive FFR at most of the frequencies and the PEAK showed positive FFR during 4~6 Hz where it elicited positive FFR. The differences between the PEAK and the LAST were much larger than those in the rat.

Interestingly, two positive inotropic agents with different action mechanisms, norepinephrine (NE, a beta-receptor agonist) and monensin (a Na^+ ionophore), had differential influences on the LA amplitude accommodations to the test frequencies in rat and rabbit. NE increased the LA amplitude accommodations in both species. The increments were more prominent in the LAST except in the rat during test frequencies higher than 4 HZ where the PEAK increased more prominently (Fig. 2A & B). In the case of monensin however, the LA amplitude accommodation enhancements were apparently different between rat and rabbit, as shown in Fig 3A & B. In the rabbit LA (Fig. 3B), monensin markedly enhanced both FFR's of the Peak and the Last as like those after NE. While in the rat LA (Fig. 3A), the effects of monensin were negligible. Similar results were obtained after ouabain application (Fig. 4A & B). Ouabain also enhanced the amplitude accommodations to the test frequencies in rabbit LA only. These results suggest that, in the rabbit LA, Na^+/Ca^{2+} exchange has enormous potential in enhancing the amplitude accommodation to the frequency-change, while the potential seems to be minor in the rat LA.

The reason for this species difference after monensin was pursued in the postnatal developing rat LA, in which the prevalent Ca^{2+} regulatory mechanism in the myocardial excitation-contraction (E-C) coupling changes from Na^+/Ca^{2+} exchange to SR as it matures. It has been reported that, in the rat heart, the

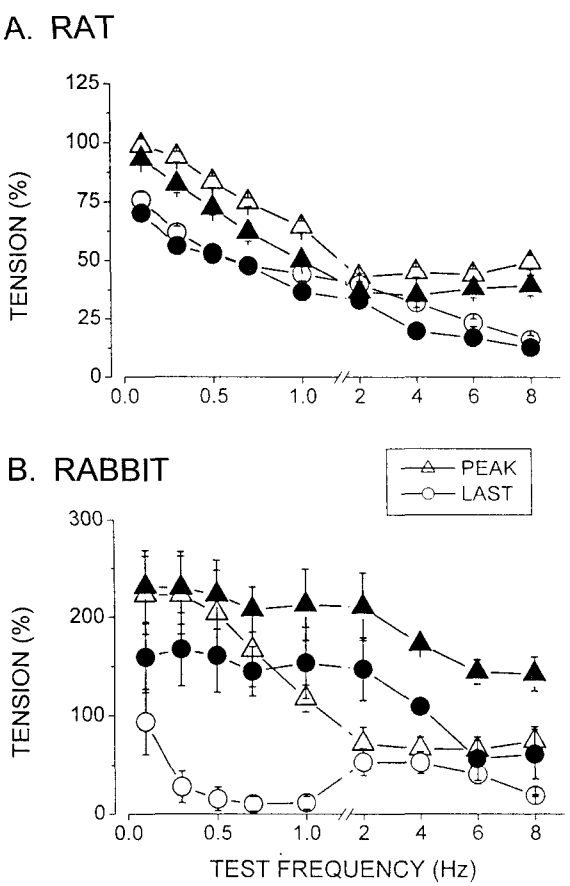

Fig. 4. Effects of ouabain (10^{-6} M) on the test frequency-related changes in the amplitude accommodation in the left atria from rat (A) and rabbit (B). (Open symbol: Control, Closed symbol: Ouabain). Other legends are same as Fig. 1.

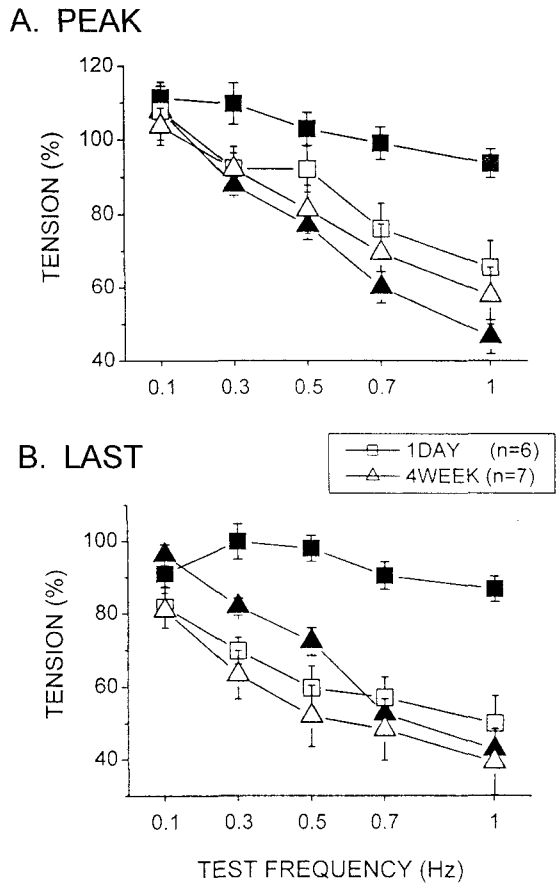

Fig. 5. Differential effects of monensin (3×10^{-6} M) on the test frequency-related changes in the amplitude accommodation in the left atria from postnatal developing rat. A. PEAK, B. LAST. Open symbol: Control, Closed symbol: Monensin. Parentheses are numbers of data. Other legends are same as Fig. 1.

SR attains full maturation at 4th week after birth as the SR Ca^{2+}-ATPase mRNA level increases by 97% between day 1 and 30 after birth, while the Na^{+}-Ca^{2+} exchanger reciprocally decreased by 75% during the same period from its maximal activity before birth (Olivetti et al, 1980; Chin et al, 1990; Ostadalova et al, 1993; Vetter et al, 1995; Vornanen 1996; Studer et al, 1997; Koban et al, 1998). As shown in Fig 5A & B, the postnatal developing rat LA (1-day and 4-week after birth) elicited negative FFR's in both of the Peak and the Last. In 1-day-old LA, monensin application increased both of the Peak (Fig. 5A) and the Last (Fig. 5B) in a frequency-dependent manner, totally flattening the FFR's. However, interestingly enough in 4-week-old LA, both of the Peak (Fig. 5A) and the Last (Fig. 5B) were hardly enhanced and the negative FFRs were maintained after monensin. These results imply that the LA amplitude accommodation to the test frequency is influenced by the prevalent Ca^{2+} regulatory mechanism in the myocardial E-C coupling. From these results therefore, it may be suggested that the differences in the prevalence of Na^{+}/Ca^{2+} exchanger and SR in the rat and rabbit LA determine the species difference in the LA amplitude accommodation to the frequency-change after monensin.

After suppression of Na^{+}/Ca^{2+} exchange, the changes in the LA amplitude accommodation to the frequency-change were tested in both of rat and rabbit. Na^{+}/Ca^{2+} exchange was suppressed by RNa-0Ca PSS application (26Na-0Ca KHB for rat LA, 7Na-0Ca Tyrode for rabbit LA: see method). In the rat LA as shown in Fig. 6A, the amplitude was rapidly increased to the peak and then slowly decreased, nevertheless the LA twitch contraction never

ceased during 5 minutes tested after 26Na-0Ca KHB. Differently from rat LA, the rabbit LA contraction lasts only 2~3 minutes and the amplitude showed 2-phased enhancement after 7Na-0Ca Tyrode application. Interestingly, the contractions ceased immediately after 0Ca PSS application (data not shown). These results imply that this much reduction in Na^+ in addition to Ca^{2+} omission in the PSS seemed to be enough to suppresses the Ca^{2+} extrusion via Na^+/Ca^{2+} exchange so that internal Ca^{2+} recycles in the cell and maintains the contraction. In this situation, the contraction is governed mainly by the SR, as the SR is the major source of internal Ca^{2+}. Other Ca^{2+} extrusion mechanism like sarcolemmal Ca^{2+}-ATPase affects the contraction minimally and may be responsible for the continuous amplitude decrease (Choi & Eisner, 1999). Although the PEAK's were enhanced after RNa-0Ca PSS application, FFR's of the attained amplitudes showed inverse proportion with the test frequency-increase in both of rat and rabbit LA (data not shown). In Fig. 6C, the net enhancements in the PEAK's attainted after RNa-0Ca PSS were calculated as % increase to their own controls (see method) in both rat and rabbit LA, and their FFR's were shown against the test frequency, which were interestingly different between rat and rabbit LA. In the rat LA, the % increase in the PEAK was maximum (70%) at 0.1 Hz and decreased in inverse

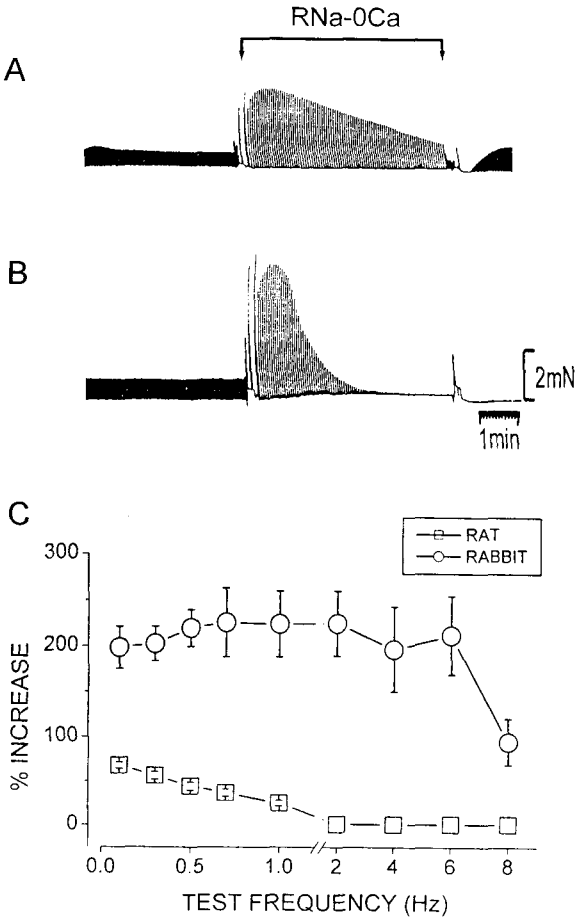

Fig. 6. Differential effects of Na^+-Ca^{2+} exchange inhibition between rat and rabbit on the test frequency-related changes in the left atrial amplitude accommodation. Na^+-Ca^{2+} Exchange was inhibited by Na^+ reduced, Ca^{2+} depleted (RNa-0Ca) Tyrode solution application. A: Actual tracings in rat LA. B: Actual tracings in rabbit LA. C: Test frequency-related changes of the PEAK amplitudes. RNa-0Ca: 26Na-0Ca for rat and 7Na-0Ca for rabbit (See method). Other legends are same as Fig. 1.

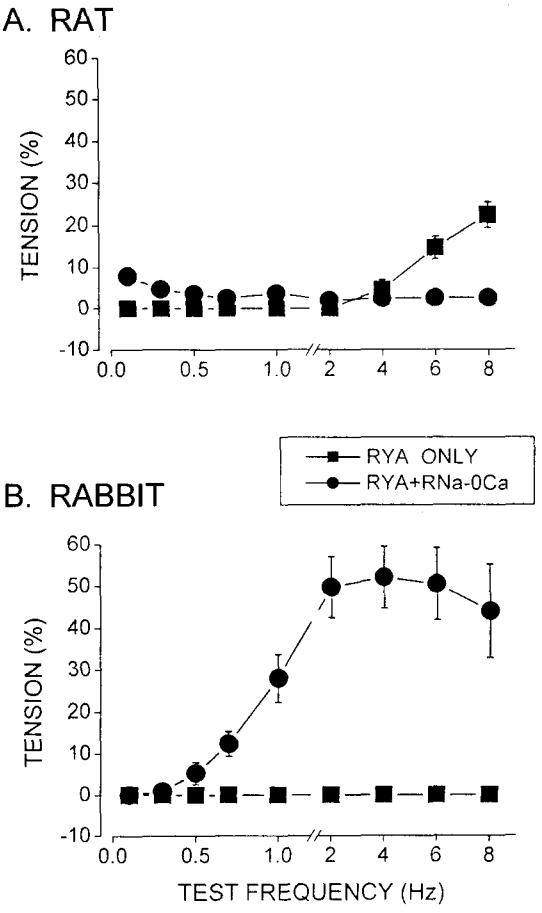

Fig. 7. Differential influences of RNa-0Ca PSS after ryanodine (3×10^{-9} M) between rat (A) and rabbit (B) on the test frequency-related changes in the left atrial amplitude accommodation. RYA: Ryanodine. RNa-0Ca: 26Na-0Ca for rat and 7Na-0Ca for rabbit (See method). Numbers of data are 5. Other legends are same as Fig. 1.

proportion to the test frequency-increase up to 2 Hz where there was no enhancement (0%) and became constant, while, in the rabbit LA, the PEAK enhancements were constant around 200% of their own controls regardless of the test frequency except 8Hz. The constancy of % increases in the PEAK's of rabbit LA does not necessarily mean that the FFR was flattened after 7Na-0Ca Tyrode since their own controls decreased as shown in Fig. 1D. Therefore, the FFR after 7Na-0Ca Tyrdoe was actually decreased according as the test frequency increased. These results suggest that the SR in the rat LA has stronger influence on the amplitude accommodation to the frequency-change than in the rabbit LA.

Conversely in Fig. 7, the influences of Na^+/Ca^{2+} exchanger on the amplitude accommodation were tested after SR Ca^{2+} release suppression with ryanodine in both of the rat and rabbit LA. 40 minutes after ryanodine (3×10^{-9} M) application, LA twitch during resting frequency (3 Hz) was totally suppressed in both of the rat and rabbit (data not shown). The suppression of LA twitch by ryanodine was continued after changing to the test frequencies, except 4～8 Hz in the rat LA (Fig. 7A & B). However, the twitch reappeared and elicited the test frequency-related enhancements in the amplitude accommodation after 7Na-0Ca Tyrode in the rabbit LA. The tensions attained after 7Na-0Ca Tyrode in the rabbit LA were negligible at low frequencies, but they increased up to 50% after 2 Hz, which was 1/4 of the tension attained by 7Na-0Ca Tyrode without ryanodine (Fig. 7B). The twitch also reappeared in the rat LA, however, the tensions attained after 26Na-0Ca KHB were almost negligible throughout the test frequencies in the rat LA (Fig. 7A). These results therefore show that the SR Ca^{2+} release plays a major role in the amplitude accommodation in both of the rat and rabbit LA. Nevertheless, the regulatory role of Na^+/Ca^{2+} exchanger in the amplitude accommodation is stronger enough to elicit frequency-related increase in amplitude in the rabbit LA than in the rat LA.

DISCUSSION

It has long been known that the myocardial force-frequency relationship (FFR) differs in rat and rabbit. In the rabbit heart, the developed force increases as the stimulation frequency-increases (positive FFR), while it decreases in the rat heart (negative FFR). After frequency-increase, two major changes are induced in the heart: first, increase in the free myoplasmic Na^+ concentration ($[Na^+]_i$) by the more frequent opening of voltage-dependent Na^+ channels (Cohen et al, 1982; Wier & Yue, 1986; Boyet et al, 1987; Harrison et al, 1992; Lostan et al, 1995; Maier et al, 1997), second, less sarcoplasmic reticulum (SR) Ca^{2+} loads in the short time available (Orachard & Lakatta, 1985; Frampton et al, 1991; Bers et al, 1993; Satoh et al, 1997). After frequency-increase, the $[Na^+]_i$ increase will enhance the myocardial force by slowing or reversing the Ca^{2+} extrusion to evoke a secondary Ca^{2+} gain (Wang et al, 1988; Harrison & Boyet, 1995), however, the decrease in SR Ca^{2+} loads will decrease the myocardial force by resulting in less SR Ca^{2+} release (Orachard & Lakatta, 1985; Frampton et al, 1991; Bers et al, 1993; Satoh et al, 1997). Therefore, the Ca^{2+} balance and the resulting myocardial force after frequency-increase will be determined by the sum of these two counteractive processes.

It has been known that the hearts in the different species have different compositions of SR and Na^+/Ca^{2+} exchanger. The extreme case would be the amphibian hearts which have no functional SR (Weiss & Morad, 1981; Klitzner & Morad, 1983). It has been reported that the compositions of SR and Na^+/Ca^{2+} exchanger in the rat heart is 92% and 7%, respectively, while in the rabbit heart, the balance is more in the range of 70% and 25～30%, respectively (Bers et al, 1996). The functional results of the present experiment in Fig. 7 are consistent with this report.

The difference in FFR between rat and rabbit heart could be derived from the different compositions of SR and Na^+/Ca^{2+} exchanger between rat and rabbit heart. The present results obtained from the postnatal developing rat LA in the Fig. 5 supports this idea. The increase in the Na^+/Ca^{2+} exchange activity after monensin, a Na^+ ionophore, could strongly enhance the FFR in the LA from 1-day old rat, which has high Na^+/Ca^{2+} exchanger with scant and immature SR. However, in the 4 week-old LA, which has 75% less Na^+/Ca^{2+} exchanger, and 97% more in quantity and fully matured in quality SR than 1-day old LA (Olivetti et al, 1980; Chin et al, 1990; Ostadalova et al, 1993; Vetter et al, 1995; Vornanen 1996; Studer et al, 1997; Koban et al, 1998), the increase in the Na^+/Ca^{2+} exchange activity after monensin could not enhance the FFR at all. These results clearly show that the magnitude of the influence of Na^+/Ca^{2+}

exchanger on the frequency-related amplitude accommodation is proportional to the composition of Na^+/Ca^{2+} exchanger (with reciprocally changing SR composition) in the LA. This idea can be extended to the difference in FFR between rat and rabbit heart. In rat heart, the change in the Na^+/Ca^{2+} exchanger activity attained after frequency-increase would be too weak as shown in Fig. 7A to overcome the decrease in the SR Ca^{2+} loads and the resulting decrease in SR Ca^{2+} release after frequency-increase. On the other hand in the rabbit heart, the change in the Na^+/Ca^{2+} exchange activity after frequency-increase shown in Fig. 7B would be strong enough to overcome the decrease in SR Ca^{2+} loads and releases. Therefore, it can be suggested that the differences in the prevalence of their Na^+/Ca^{2+} exchanger or SR in the amplitude accommodation to the frequency-change determine the difference in the FFR between rat and rabbit heart.

Interestingly in the rat LA, ryanodine application converted the FFR into positive during the test frequencies higher than 2 Hz in Fig. 7A. Similar changes have been reported in single rat ventricular myocytes (Borzak et al, 1991). The exact reason for this conversion after ryanodine is still unclear, however it may suggest the significance of SR in the FFR. In the line of this idea, phospholamban, a protein that regulates SR Ca^{2+}-ATPase, has been raised recently as a common determinant in both of positive and negative FFR's (Kadambi et al, 1999; Bluhm et al, 2000) in the line of this idea.

The FFR's presented in the present experiment appeared to be biphasic especially in the PEAK's in both of the rat and rabbit LA. However, similar biphasic results have been reported in perfused whole heart (Henry, 1975) and in thin myocardial preparation (Gulch & Ebrecht, 1987). Proper diffusion of oxygen and nutrients has been raised as a responsible cause for this discrepancy.

In this report, the mechanism for the difference in the frequency-force relationship between rat and rabbit heart were investigated in the electrically field stimulated left atria from rat and rabbit by using amplitude accommodation to the stimulation frequency-change from resting 3 Hz to the variable test frequencies for 5 minutes. It is concluded that the differences in the prevalence between myocardial Na^+/Ca^{2+} exchanger and SR in the amplitude accommodation to the frequency-change determine the difference in the FFR between rat and rabbit heart.

ACKNOWLEDGEMENTS

This work was supported by a faculty research grant of Yonsei Cardiovascular Research Institute and Woonkyung foundation (1996).

REFERENCES

Bers DM, Bassani JWM, Bassani RA. Na-Ca exchange and Ca fluxes during contraction and relaxation in mammalian ventricular muscle. *Ann NY Acad Sci* 779: 430−442, 1996

Bers DM, Bassani RA, Bassani JWM, Baudet S, Hryshko LV. Paradoxical twitch potentiation after rest in cardiac muscle: increased fractional release of SR calcium. *J Mol Cell Cardiol* 25: 1047−1057, 1993

Bluhm WF, Kranias EG, Dillmann WH, Meyer M. Phospholamban: a major determinant of the cardiac force-frequency relationship. *Am J Physiol (Heart Circ Physiol)* 278: H249−H255, 2000

Borzak S, Murphy S, Marsh JD. Mechanisms of rate staircase in rat ventricular cells. *Am J Physiol* 260: H884−H892, 1991

Boyet MR, Hart G, Levi AJ. Factors affecting intracellular sodium during repetitive activity in isolated sheep Purkinje fibres. *J Physiol (Lond)* 384: 405−429, 1987

Chin TK, Friedman WF, Klitzner TS. Developmental changes in cardiac myocyte calcium regulation. *Circ Res* 67: 574−579, 1990

Choi HS, Eisner DA. The role of sarcolemmal Ca^{2+}-ATPase in the regulation of resting calcium concentration in rat ventricular myocytes. *J Physiol (Lond)* 515: 109−118, 1999

Cohen CJ, Fozzard HA, Sheu SS. Increase in intracellular sodium ion activity during stimulation in mammalian cardiac muscles. *Circ Res* 50: 651−652, 1982

Frampton JE, Harrison SM, Boyet MR, Orchard CH. Ca^{2+} and Na^+ in rat myocytes showing different force-frequency relationships. *Am J Physiol* 261 (*Cell Physiol* 30): C739−C750, 1991

Gulch RW, Ebrecht G. Mechanics of rat myocardium revisited: investigations of ultra-thin cardiac muscles under high energy demand. *Basic Res Cardiol* 81(Suppl II): 263−274, 1987

Harrison SM, McCall E, Boyet MR. The relationship between contraction and intracellular sodium in rat and guinea-pig ventricular myocytes. *J Physiol (Lond)* 449: 517−550, 1992

Harrison SM, Boyett MR. The role of the Na^+-Ca^{2+} exchanger in the rate-dependent increase in contraction in guinea-pig ventricular myocytes. *J Physiol (Lond)* 482: 555−566, 1995

Hattori Y, Toyama J, Kodama I. Cytosolic calcium staircase in ventricular myocytes isolated from guinea pigs and rats. *Cardiovasc Res* 25: 622−629, 1991

Henry PD. Positive staircase effect in the rat heart. *Am J Physiol* 228: 360−364, 1975

Hussain M, Drago GA, Colyer J, Orchard CH. Rate-dependent abbreviation of Ca^{2+} transient in rat heart is independent of phospholamban phosphoylation. *Am J Physiol* 273 (*Heart Circ Physiol* 42): H695−H706, 1997

Kadambi VJ, Ball N, Kranias EG, Walsh RA, Hott BD. Modulation of force-frequency relation by phospholamban in genetically engineered mice. *Am J Physiol* 276 (*Heart Circ Physiol* 45): H2245−H2250, 1999

Klitzner T, Morad M. Excitation-contraction coupling in frog ventricle: Possible Ca^{2+} transport mechanisms. *Pflugers Arch* 398: 274−283, 1983

Koban MU, Moorman AF, Holtz J, Yacoub MH, Boheler KR. Expressional analysis of the cardiac Na-Ca exchanger in rat development and senescence. *Cardiovasc Res* 37: 405−423, 1998

Lostan CS, Miller SK, Simor T, Elgavish GA. Cardiac staircase and NMR-determined intracellular sodium in beating rat hears. *Am J Physiol* 269 (*Heart Circ Physiol* 38): H332−H340, 1995

Maier LS, Pieske B, Allen DG. Influence of stimulation frequency on $[Na^+]_i$ and contractile function in Langendorff-perfused rat heart. *Am J Physiol* 273 (*Heart Circ Physiol* 42): H1246−H1254, 1997

Olivetti G, Anversa P, Loud AV. Morphometric study of early postnatal development in the left and right ventricular myocardium of the rat. II. Tissue composition, capillary growth, and sarcolplasmic alterations. *Circ Res* 46: 503−512, 1980

Orchard CH, Lakatta EG. Intracellular calcium transients and developed tension in rat heart muscle. A mechanism for the negative interval-strength relationship. *J Gen Physiol* 86: 637−651, 1985

Ostadalova I, Kolar F, Ostadal B, Rohlicek V, Rohlicek J, Prochazka J. Early postnatal development of contractile performance and responsiveness to Ca^{2+}, verapamil and ryanodine in the isolated rat heart. *J Mol Cell Cardiol* 25: 733−740, 1993

Satoh H, Blatter LA, Bers DM. Effects of $[Ca^{2+}]_i$, SR Ca^{2+} load, and rest on Ca^{2+} spark frequency in ventricular myocytes. *Am J Physiol* 272 (*Heart Circ Physiol* 41): H657−H668, 1997

Studer R, Reinecke H, Vetter R, Holtz J, Drexler H. Expression and function of the cardiac Na^+/Ca^{2+} exchanger in postnatal development of the rat, in experimental-induced cardiac hypertrophy and in the failing human heart. *Basic Res Cardiol* 92(*Suppl 1*): 53−58, 1997

Terracciano CMN, De Souza AI, Philipson KD, MacLeod KT. Na^+-Ca^{2+} exchange and sarcoplasmic reticular Ca^{2+} regulation in ventricular myocytes form transgenic mice overexpressing the Na^+-Ca^{2+} exchanger. *J Physiol (Lond)* 512: 651−657, 1998

Vetter R, Studer R, Reinecke H, Kolar F, Ostadalova I, Drexler H. Reciprocal changes in the postnatal expression of the sarcolemmal Na^+-Ca^{2+} exchanger and SERCA2 in rat heart. *J Mol Cell Cardiol* 27: 1689−1701, 1995

Vornanen M. Excitation-contraction coupling of the developing rat heart. *Mol Cell Biochem* 163−164: 5−11, 1996

Wang DY, Chae SW, Gong QY, Lee CO. Role of a^i_{Na} in positive force-frequency staircase in guinea-pig papillary muscle. *Am J Physiol* 255 (*Cell Physiol* 24): C798−C907, 1988

Weiss RE, Morad M. Intrinsic birefringence signal preceding the onset of contraction in heart muscle. *Science* 213: 663−666, 1981

Wier WG, Yue DT. Intracellular calcium transients underlying the short-term force-interval relationship in ferret ventricular myocardium. *J Physiol (Lond)* 376: 507−530, 1986