Improvement of Cardiac Function by a Cardiac Myosin Activator in Conscious Dogs With Systolic Heart Failure

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Background—Therapy for chronic systolic heart failure (sHF) has improved over the past 2 decades, but the armamentarium of drugs is limited and consequently sHF remains a leading cause of death and disability. In this investigation, we examined the effects of a novel cardiac myosin activator, omecamtiv mecarbil (formerly CK-1827452) in 2 different models of heart failure.

Methods and Results—Two different models of sHF were used: (1) pacing-induced sHF after myocardial infarction (MI-sHF) and (2) pacing-induced sHF after 1 year of chronic pressure overload left ventricular hypertrophy (LVH-sHF). Omecamtiv mecarbil increased systolic function in sHF dogs, chronically instrumented to measure LV pressure, wall thickness, and cardiac output. Omecamtiv mecarbil, infused for 24 hours, induced a sustained increase without desensitization \((P<0.05)\) in wall thickening \((25\pm6.2\%)\), stroke volume \((44\pm6.5\%)\) and cardiac output \((22\pm2.8\%)\), and decreased heart rate \((15\pm3.0\%)\). The major differences between the effect of omecamtiv mecarbil on cardiac function and the effect induced by a catecholamine, for example, dobutamine, is that omecamtiv mecarbil did not increase LV dP/dt but rather increased LV systolic ejection time by \(26\pm2.9\%\) in sHF. Another key difference is that myocardial O2 consumption \((\text{MVO}_2)\), which increases with catecholamines, was not significantly affected by omecamtiv mecarbil.

Conclusions—These results demonstrate that chronic infusion of the cardiac myosin activator, omecamtiv mecarbil, improves LV function in sHF without the limitations of progressive desensitization and increased MVO2. This unique profile may provide a new therapeutic approach for patients with sHF. (Circ Heart Fail. 2010;3:522-527.)

Key Words: cardiac myosin activator ■ heart failure ■ inotropic agents ■ omecamtiv mecarbil ■ CK-1827452

Heart failure (HF), the common end stage of most forms of heart disease, afflicted 2.5% of the US population (2.7 million people) in 2006 and was the cause of death for almost 300,000 people in 2005. The National Heart, Lung, and Blood Institute Framingham study has shown that 80% of men and 70% of women with HF under age 65 will die within 8 years. The estimated total cost of HF in 2009 is more than $37 billion.

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By definition, the physiology of the most common form of HF, systolic HF (sHF), always includes an increase in both preload and afterload and a decrease in systolic left ventricular (LV) function. Current therapy for sHF includes diuretics, aldosterone antagonists, angiotensin-converting enzyme inhibitors, angiotensin receptor inhibitors, and \(\beta\)-blockers. Whereas the control of loading conditions, for example, with diuretics and angiotensin-converting enzyme inhibitors, shows clinical benefits,\(^3\) improving the inotropic defect of the failing myocardium has proved more challenging.

The search for compounds that increase LV systolic function of the failing heart has been primarily limited to therapeutic approaches that revolve around increasing the concentration of cAMP and consequently Ca\(^{2+}\) as exemplified by sympathomimetic amines (isoproterenol, dobutamine, dopamine, and norepinephrine) and phosphodiesterase inhibitors. Whereas acute, short-term application of these therapies can be salutary in selected patients, use of drugs that increase cytosolic Ca\(^{2+}\) in HF has been uniformly deleterious.\(^4\) In both acute and chronic settings, an increased incidence of ischemia and arrhythmias, both atrial and ventricular, has been noted in randomized clinical studies.\(^5\) In addition to the adverse effect that catecholamines have on myocardial \(\text{O}_2\) requirements, a problem in patients with limited coronary reserve or with chronic coronary artery disease, desensitization of adrenergic receptors to catecholamines is another serious obstacle to their use in patients with severe sHF. Although LV systolic function can be temporarily restored by escalating doses of catecholamines, this benefit is at the expense of increased myocardial oxygen \((\text{O}_2)\) consumption \((\text{MVO}_2)\). Existing Ca\(^{2+}\) sensitizers, such as levosimendan, have similar limitations.\(^8\)

A novel approach to improve cardiac LV systolic function that may address these limitations is through activation of the...
force-generating protein itself, cardiac myosin. This approach is made possible with the novel cardiac myosin-activating compound omecamtiv mecarbil (formerly called CK-1827452). Cardiac myosin activators accelerate the transition of the actin-myosin complex from weakly bound to strongly bound configuration, thus increasing the number of “independent force generators” (myosin heads) interacting with the actin filament while at the same time reducing the rate of nonproductive ATP hydrolysis. These effects are totally independent from Ca\(^{2+}\) homeostasis and therefore the improvement in LV systolic function should not come at the cost of increased energy demand or arrhythmogenesis.

Therefore, the goal of the present study was to test the effects of omecamtiv mecarbil on cardiac contractile parameters in conscious dogs before and after sHF to show how this drug compares with classic inotropic agents in terms of desensitization and increased MVO\(_2\). One model used was rapid ventricular pacing after myocardial infarction (MI) induced at initial operation. The addition of the MI results in rapid ventricular hypertrophy (LVH), which typically develops in a setting of chronic sHF. Thus, the expanded goal of this study was to determine if omecamtiv mecarbil affected LV systolic function and cardiac output favorably in two models of sHF: rapid pacing after MI and rapid pacing after prolonged supravalvular aortic stenosis induced LV hypertrophy.

### Methods

#### Instrumentation

Animals used in this study were obtained from the Guide for the Care and Use of Laboratory Animals of the Institute (NIH, 1996) and New Jersey Medical School Institutional Animal Care and Use Committee. Mongrel dogs (weight, 15 to 18 kg) of either sex were anesthetized with thiopental (15 mg/kg IV) followed by halothane (1.0 to 1.5 vol%) and were placed on a ventilator with supplemental O\(_2\) during surgery. A left thoracotomy was performed in the 4th intercostal space. Tygon catheters were placed in the descending aorta and the right atrial appendages to measure their respective pressures as well as for injection of the test agent. A Silastic catheter was implanted on the endocardial and epicardial surfaces to measure LV pressure. Piezoelectric ultrasonic dimension crystals were implanted on the endocardial and epicardial surfaces to measure LV wall thickness. A Transonic flow probe was placed around the left circumflex coronary artery to measure coronary blood flow, and another probe was placed around the root of the ascending aorta for measurements of stroke volume and cardiac output. Stainless steel pacing wires were placed on the left atrium and right ventricle for rapid ventricular pacing to induce sHF. Catherers and electric leads were externalized between the scapulae, and the chest was closed in layers. Postoperative analgesics were administered: a Fentanyl patch, morphine (0.5 to 1 mg/kg IM or IV given at 4-hour and 8-hour postplacement of the Fentanyl patch), and buprenorphine (0.005 to 0.05 mg/kg IM or IV, administered BID for 3 days). Additional analgesics were given as needed based on clinical evaluation performed by the veterinarian.

#### Measurements of Cardiac Function

Cardiac function and systemic hemodynamic recordings were made using a PowerLab+Notocord data acquisition system and a multiple-channel oscillograph. Aortic and atrial pressure measurements were measured using strain gauge manometers that had been calibrated with a mercury manometer connected to the fluid-filled catheter. The solid-state LV pressure gauge was cross-calibrated with aortic and left atrial pressure measurements. LV dp/dt was obtained by electronically differentiating the LV pressure signal. A triangular wave signal was substituted for the pressure signals to directly calibrate the differentiator. LV wall thickness was measured using an ultrasonic transit-time dimension gauge. LV end-systolic dimension was determined at minimum LV dp/dt and LV end-diastolic dimension was measured at the time that coincided with beginning of the upstream of the LV dp/dt. Systolic wall thickening was calculated as the difference between end-diastolic and end-systolic wall thickness. MVO\(_2\) was calculated using the Fick equation as the product of the coronary blood flow and measured arterio-venous O\(_2\) difference. Stroke volume was calculated as the quotient of cardiac output and heart rate. LV systolic ejection time was defined as the difference between the peak positive and negative LV dp/dt and also the duration of aortic blood flow.

#### Animal Model of sHF After MI

At the time of the initial operation, MI was induced by ligation of the left anterior descending coronary artery at one third of its length (sHF after MI will be referred to as MI-sHF). After 1 to 2 weeks of recovery from surgery, sHF was induced over the next 3 to 4 weeks with rapid right ventricular pacing at 240 beat/min using an external programmed pacemaker. The pacemaker was turned off briefly during hemodynamic measurements.

#### Animal Model of sHF Superimposed With Cardiac Hypertrophy

Puppies of either sex, 8 to 10 weeks of age, were anesthetized with thiopental (15 mg/kg IV) followed by halothane (1.0 to 1.5 vol%) and placed on a ventilator with supplemental O\(_2\), during surgery. A right thoracotomy was performed via the 3rd intercostal space. A Teflon cuff was placed around the ascending aorta to induce a 50% reduction in aortic flow.

### Table 1. Baseline Values in Conscious Dogs Before Pacing (Normal, MI, and LVH) and in Conscious Dogs After Pacing (MI-sHF and LVH-sHF)

<table>
<thead>
<tr>
<th></th>
<th>Before Pacing</th>
<th>After Pacing</th>
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<tbody>
<tr>
<td></td>
<td>Normal (n=4)</td>
<td>MI (n=5)</td>
</tr>
<tr>
<td>Mean arterial pressure, mm Hg</td>
<td>92±6.6</td>
<td>88±2</td>
</tr>
<tr>
<td>Heart rate, bpm</td>
<td>90±7.2</td>
<td>116±11†</td>
</tr>
<tr>
<td>Mean left atrial pressure, mm Hg</td>
<td>3.4±1.0</td>
<td>6.8±1.5</td>
</tr>
<tr>
<td>LV systolic pressure, mm Hg</td>
<td>115±4.7</td>
<td>110±1.5</td>
</tr>
<tr>
<td>LV end-diastolic pressure, mm Hg</td>
<td>7.3±1.7</td>
<td>7.6±0.7</td>
</tr>
<tr>
<td>LV dp/dt max, mm Hg/s</td>
<td>2861±180</td>
<td>3425±215</td>
</tr>
</tbody>
</table>

*P<0.05 versus normal.
†There is 1 less animal in this measurement than for all others in this group.
diameter, which also prevented the measurement of aortic blood flow in these animals. This model is explained in more detailed in some of our original publications.13,14 About 1 year after aortic banding, these dogs were instrumented for LV function (see above) and experiments were conducted after 3 to 4 weeks of rapid ventricular pacing (shF superimposed with cardiac hypertrophy will be referred to as LVH-shF).

**Experimental Protocol**

Before and during the postoperative recovery period, the dogs were trained to lie quietly in the right lateral position. The experiments were performed in sinus rhythm. After a preliminary study to determine the dose-response effects of omecamtiv mecarbil (from 0.1 to 1.0 mg/kg bolus), an optimal dose regimen, bolus injection (0.25 mg/kg IV) followed immediately by an infusion (0.25 mg/kg/h IV) for 24 hours was used. This dose was selected because it produced plasma concentrations at steady-state that resulted in substantial effects as determined from the short-term protocol just described. In 4 MI-shF dogs, the infusion was sustained for 72 hours. Bolus administration of omecamtiv mecarbil was also performed in 4 additional normal, conscious dogs, in which the same instrumentation was present, but without MI and rapid ventricular pacing. Sampling of omecamtiv mecarbil plasma concentrations indicated that steady-state levels were achieved within 24 hours.

**Statistical Analysis**

Data are expressed as means±SE. The data in Table 1 were analyzed using Student t test. Both the MI-shF and the LVH-shF groups were compared with the same set of normal dog baseline values. The data in Table 2 were analyzed using a repeated-measures ANOVA with all multiple comparisons being made relative to baseline using Dunnett multiple comparisons procedure. The data in Table 3 were analyzed using Student t test. A probability value of <0.05 was considered significant.

**Results**

**Hemodynamics in Conscious Dogs Before and After MI-shF**

MI alone did not affect LV function substantially compared with baseline values in normal, conscious dogs (Table 1), but MI-shF was accompanied by significant (P<0.05) changes in LV function (Table 1). Compared with the control condition, MI-shF was accompanied by significant (P<0.05) changes in LV function (Table 1): increased heart rate (from 90±7.2 to 143±7.1 bpm), mean left atrial pressure (from 5.7±1.6 to 25.0±4.7 mm Hg), and LV end-diastolic pressure (from 156±5.7 to 24.5±4.8 mm Hg). LV systolic ejection time (from 17.0±4.6 to 24.5±8.7 ms), and cardiac output (from 2.6±0.3 to 1.5±0.17 L/min). Effects of Omecamtiv Mecarbil (24-Hour Infusion) in Conscious Dogs With Post-MI shF

The effects of omecamtiv mecarbil on hemodynamic parameters in dogs with MI-shF are illustrated in Table 2. Omecamtiv mecarbil significantly increased cardiac output, LV systolic ejection time, and stroke volume. These effects occurred in concert with decreases in heart rate, mean left atrial pressure, and LV end-diastolic pressure (Table 2). The salutary effects of omecamtiv mecarbil on LV systolic function in MI-shF persisted for the entire 24-hour infusion, indicating that desensitization did not occur. In dogs with MI-shF, in which we measured stroke volume and cardiac output, the 24-hour infusion with omecamtiv mecarbil induced a sustained increase in stroke volume (44±6.5%) and cardiac output (22±2.8%) and LV systolic ejection time (26±2.9%). Total peripheral resistance declined significantly at 24 hours. In a subset of the MI-shF dogs the infusion was continued for 72 hours, and the increases in cardiac output (32±8.2%) and stroke volume (45±7.8%)

**Table 2. Effects of Omecamtiv Mecarbil on LV Function in Conscious Dogs With MI-shF**

<table>
<thead>
<tr>
<th>Omecamtiv Mecarbil Infusion (% Change From Baseline)</th>
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<tbody>
<tr>
<td>n</td>
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<tr>
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<tr>
<td>Mean arterial pressure, mm Hg</td>
</tr>
<tr>
<td>Heart rate, beats/min</td>
</tr>
<tr>
<td>Mean left atrial pressure, mm Hg</td>
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<tr>
<td>LV systolic ejection time, ms</td>
</tr>
<tr>
<td>LV systolic pressure, mm Hg</td>
</tr>
<tr>
<td>LV end-diastolic pressure, mm Hg</td>
</tr>
<tr>
<td>LV dp/dt max, mm Hg/s</td>
</tr>
<tr>
<td>Systolic wall thickening, mm</td>
</tr>
<tr>
<td>Cardiac output, L/min</td>
</tr>
<tr>
<td>Stroke volume, mL</td>
</tr>
<tr>
<td>Total peripheral resistance, mm Hg/L/min</td>
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</tbody>
</table>

*P<0.05 versus baseline.
†This time point has 1 less animal than baseline.
‡Statistical analyses were only conducted on 15-minute to 24-hour data because only a subset of dogs were studied for 72 hours.

**Table 3. Effects of Omecamtiv Mecarbil on MVO2 in Conscious Dogs With MI-shF**

<table>
<thead>
<tr>
<th>Omecamtiv Mecarbil Infusion</th>
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<tbody>
<tr>
<td>n</td>
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<tr>
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</tr>
<tr>
<td>Coronary blood flow, mL/min</td>
</tr>
<tr>
<td>Arterial O2 content, mL/O2</td>
</tr>
<tr>
<td>Myocardial O2 consumption, mL/O2/min</td>
</tr>
<tr>
<td>Coronary sinus O2 content, mL/O2</td>
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</table>
were maintained for the entire 3-day infusion (Table 2), further confirming the lack of desensitization. The effects of the drug were no longer evident 24 hours after cessation of the infusion, a time when plasma levels of the drug were nearly absent. Figure 1 demonstrates that instead of increasing the maximal LV systolic pressure and dP/dt, as observed with most commonly used positive inotropic agents, omecamtiv mecarbil increased the duration of the LV systolic ejection time, resulting in increased stroke volume and cardiac output. Omecamtiv mecarbil had less effect on hemodynamic parameters in normal, conscious dogs; it increased LV systolic ejection time by 5.2% at this dose and did not affect LV dP/dt or cardiac output.

A major consequence of increased LV systolic function by inotropic agents is an increase in MVO$_2$. Therefore, we determined the changes in MVO$_2$ as a response to administration of omecamtiv mecarbil. Table 3 shows that MVO$_2$ and its determinants, that is, coronary blood flow and arterial and venous (coronary sinus) O$_2$ content, did not show any significant difference after infusion with omecamtiv mecarbil.

**Effects of Omecamtiv Mecarbil (24-Hour Infusion) in Conscious Dogs With LVH in the Presence of sHF**

The effects of omecamtiv mecarbil on hemodynamic parameters in dogs with LVH-sHF and those in MI-sHF were similar and not statistically different (Figure 2). In dogs with LVH-sHF, LV/body weight (g/kg) was increased (6.9±0.1) versus those in sHF after chronic MI (4.3±0.1), the MI-sHF model. In LVH-sHF, omecamtiv mecarbil induced similar significant ($P<0.05$) increases in systolic wall thickening and LV systolic ejection time as occurred in MI-sHF (Figure 2 and Table 1). The improved cardiac performance was accompanied by a decrease in heart rate and LV end-diastolic pressure (Figure 2), whereas MVO$_2$ remained unchanged (not

![Figure 1. Contractile effects of omecamtiv mecarbil. This figure compares the average of 5 to 10 phasic waveforms, from a representative animal, of LV pressure (top), LV dP/dt (middle) and aortic blood flow (cardiac output) (bottom) before (solid line) and after omecamtiv mecarbil (dashed line) in the presence of sHF. Omecamtiv mecarbil did not increase LV dP/dt but prolonged LV systolic ejection time and increased stroke volume and cardiac output.](image-url)
shown). In Figure 2, baseline values were those measured before drug administration but after pacing.

Discussion

Effects of Cardiac Myosin Activation by Omecamtiv Mecarbil

The present investigation demonstrates that activating cardiac myosin by omecamtiv mecarbil significantly improves LV function in 2 conscious dog models of sHF (MI-sHF and LVH-sHF) without a change in MVO₂, thereby markedly improving cardiac efficiency. The salutary effects of omecamtiv mecarbil on LV systolic function in sHF persisted for the entire 24-hour infusion, with similar findings when infusions were extended to 72 hours (MI-sHF, Table 2), indicating that desensitization did not occur. This pattern contrasts markedly with the administration of more commonly used positive inotropic agents. In particular, the increase in LV systolic wall thickening and stroke volume were not due to an increase in LV dP/dt, an index of myocardial LV systolic function characterizing the isovolumic phase of contraction that has previously marked the presence of a positive inotropic effect. Rather, enhanced cardiac performance was achieved by prolongation of the LV systolic ejection time, which reflects a novel mechanism. In addition, the improvement in cardiac function in response to omecamtiv mecarbil was more pronounced in sHF, in which cardiac output was increased by 22.8%, versus normal hearts, in which cardiac output was not increased. This is opposite to observations with traditional sympathomimetic amines, in which desensitization reduces inotropic effects, resulting in less improvement in cardiac output in the presence of sHF.

Cardiac myosin activators accelerate the transition of the actin-myosin complex from weakly bound to strongly bound configuration, thus increasing the number of “independent force generators” (myosin heads) interacting with the actin filament while at the same time reducing the rate of nonproductive ATP hydrolysis. These unique characteristics of cardiac myosin activation clearly provide a new potential therapeutic approach for patients with sHF, as supported by our study. Although our study is limited by using experimental animal models of HF, rather than patients with HF, the data collected in the current manuscript have been recently confirmed in preliminary reports for a clinical Phase II trial showing that omecamtiv mecarbil increases LV systolic ejection time, stroke volume, and cardiac output while reducing heart rate in a concentration-dependent manner. Importantly, another preliminary report also demonstrated that increases in LV systolic ejection time was generally well tolerated in patients with ischemic cardiomyopathy undergoing exercise conditions under which an increase in LV systolic ejection time might provoke intolerance. These potential adverse effects could be a greater problem in the hypertrophied heart. Interestingly, omecamtiv mecarbil was at least equally effective in sHF induced after chronic (1 year) pressure overload LVH, as it was in sHF after chronic MI. In fact, the salutary effects tended to be even greater, although not significantly, in the LVH-sHF model (Figure 2).

Contrast With Catecholamines

As we showed before in the same canine model, the characteristics of catecholamines are in marked contrast with the effects of cardiac myosin activators. Infusion of dobutamine in the dog without sHF induces an increase in heart rate, LV dP/dt, and LV systolic pressure, reflecting increased inotropy. This functional improvement comes at the cost of a major increase in MVO₂. These increases in MVO₂ with sympathomimetic amines are probably due to increases in heart rate and increased energetic costs associated with Ca²⁺ cycling. After the onset of sHF, the response of the physiological parameters to dobutamine is rapidly attenuated by about 65%, illustrating the classical example of catecholamine desensitization, which critically limits the use of catecholamines in the treatment of sHF. The only way to avoid desensitization is by progressively increasing the doses of dobuta-
mine, which leads to unacceptable increases in MVO₂ for a relatively modest increase in LV systolic function (O₂ wasting).²⁰

Conclusions
In conclusion, unlike existing inotropic agents, which generally demonstrate desensitization and increased MVO₂, a prolonged infusion of the cardiac myosin activator, omecamtiv mecarbil, produced a substantial improvement in LV function in sHF after chronic MI without desensitization or a change in MVO₂. These distinguishing features underlie the potential clinical significance of this novel therapeutic approach. Even when sHF was developed in the presence of severe LVH, omecamtiv mecarbil still improved LV systolic function, along with a reduction of preload, and without alteration in MVO₂.

As suggested by current early-phase clinical trials, the unique profile of this compound provides a new therapeutic approach for patients with sHF.

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Disclosures
Drs Malik and Morgans are employees of Cytokinetincs, Inc, and Drs Abarzua and Shen are employees of CV Dynamics, Inc. Dr Vatner has stock in CV Dynamics, Inc.

References
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