INTRODUCTION

Motilin, a 22-amino-acid polypeptide, is related to the regulation of gastrointestinal motility (Itoh, 1997) and new chemical entities which act on motilin receptors have recently been expected for the treatment of functional gastrointestinal tract disorders (McCallum and Cynshi, 2007; Kamerling et al., 2004). Irritable bowel syndrome (IBS) is a functional bowel disorder in which abdominal discomfort or pain is associated with altered bowel habits (Thompson et al., 1999). Since some patients with IBS showed elevated blood motilin levels (Ohe et al., 1980; Simrén et al., 2001, 2005), motilin antagonism is considered effective for the treatment of IBS. We have developed an orally active motilin receptor antagonist, MA-2029, the chemical name of which is (S)-N-[(S)-2-(3-tert-Butyl-4-hydroxy-phenyl)-1-ethylcarbamoyl-ethyl]-3-methyl-2-[methyl-((S)-2-methylamino-3-phenyl-propionyl)-amino]-butyramide hydrochloride (Fig. 1), and which is a novel drug anticipated to be effective in the treatment of IBS (Sudo et al., 2008). Serotonin (5-hydroxytryptamine; 5-HT) is also highly related to gastrointestinal tract functions (Read and Gwee, 1994) and cisapride, a 5-HT4 receptor agonist, was considered to work well for IBS patients (Evans et al., 1997; Noor et al., 1998). However, cisapride was withdrawn from the market due to its proarrhythmic potential through a QT interval prolongation (Hill et al., 1998).

Several drugs such as cisapride have shown cardiovascular side effects associated with QT interval prolongation often accompanied by the fatal ventricular tachycardia known as Torsades de Pointes (TdP) (Redfern et al., 2003). Block of the rapid component of the repolarizing delayed rectifier potassium current (I\textsubscript{K}) in the heart is the main mechanism of drug-induced QT interval prolongation (Webster et al., 2002) and the human ether-a-go-go-

Original Article

Cardiovascular safety profile of MA-2029, a novel motilin receptor antagonist

Mitsuyasu Tabo, Ryuichi Komatsu, Masaki Honda, Misae Itoh and Kazuya Kimura

Fuji Gotemba Research Laboratories, Chugai Pharmaceutical Co., Ltd., 1-135 Komakado, Gotemba, Shizuoka 412-8513, Japan

(Received September 19, 2008; Accepted October 14, 2008)

ABSTRACT — The aim of this study was to assess the cardiovascular effect of MA-2029, a selective motilin receptor antagonist highly expected for the treatment of irritable bowel syndrome (IBS). MA-2029 inhibited the human ether-a-go-go-related gene (hERG) current at 100 μg/ml, but shortened action potential duration (APD) in isolated guinea pig papillary muscles at 10 and 100 μg/ml and the corrected QT (QTc) interval after oral administration of 30 and 300 mg/kg in conscious telemetered dogs. The discrepancy was probably caused by blockade of the Ca\textsuperscript{2+} channel because MA-2029 inhibited the Ca\textsuperscript{2+} current in isolated guinea pig myocytes. MA-2029 at 100 μg/ml also decreased the maximum rising velocity and action potential amplitude in the action potential study, indicating that MA-2029 has Na\textsuperscript{+} channel blocking potential. In the cardiovascular study, MA-2029 at 30 mg/kg induced slight cardiovascular changes such as hypotension, QTc shortening, and PR prolongation possibly caused by Ca\textsuperscript{2+} channel blockade. The plasma concentration at 4 hr after 30 mg/kg administration was 2.10 μg/ml, 200-fold higher than the effective concentration of MA-2029 as a motilin receptor antagonist. These results suggest that MA-2029 has sufficient cardiovascular safety although it inhibits multiple ion channels at supra-effective concentrations. On the other hand, cisapride, an effective IBS drug, showed clear hERG inhibition and APD prolongation at 100 ng/ml. Cisapride exhibited a narrow safety margin because it caused QT prolongation potential even at the therapeutic concentration. In conclusion, MA-2029 is a novel drug highly expected for the treatment of IBS with lower cardiovascular risk than cisapride.

Key words: MA-2029, Cisapride, Safety assessment, Cardiovascular system, hERG, Action potential

INTRODUCTION

Motilin, a 22-amino-acid polypeptide, is related to the regulation of gastrointestinal motility (Itoh, 1997) and new chemical entities which act on motilin receptors have recently been expected for the treatment of functional gastrointestinal tract disorders (McCallum and Cynshi, 2007; Kamerling et al., 2004). Irritable bowel syndrome (IBS) is a functional bowel disorder in which abdominal discomfort or pain is associated with altered bowel habits (Thompson et al., 1999). Since some patients with IBS showed elevated blood motilin levels (Ohe et al., 1980; Simrén et al., 2001, 2005), motilin antagonism is considered effective for the treatment of IBS. We have developed an orally active motilin receptor antagonist, MA-2029, the chemical name of which is (S)-N-[(S)-2-(3-tert-Butyl-4-hydroxy-phenyl)-1-ethylcarbamoyl-ethyl]-3-methyl-2-[methyl-((S)-2-methylamino-3-phenyl-propionyl)-amino]-butyramide hydrochloride (Fig. 1), and which is a novel drug anticipated to be effective in the treatment of IBS (Sudo et al., 2008). Serotonin (5-hydroxytryptamine; 5-HT) is also highly related to gastrointestinal tract functions (Read and Gwee, 1994) and cisapride, a 5-HT4 receptor agonist, was considered to work well for IBS patients (Evans et al., 1997; Noor et al., 1998). However, cisapride was withdrawn from the market due to its proarrhythmic potential through a QT interval prolongation (Hill et al., 1998).

Several drugs such as cisapride have shown cardiovascular side effects associated with QT interval prolongation often accompanied by the fatal ventricular tachycardia known as Torsades de Pointes (TdP) (Redfern et al., 2003). Block of the rapid component of the repolarizing delayed rectifier potassium current (I\textsubscript{K}) in the heart is the main mechanism of drug-induced QT interval prolongation (Webster et al., 2002) and the human ether-a-go-go-
related gene (hERG) channel is the molecular counterpart of the \( I_{\text{Kr}} \) (Sanguinetti et al., 1995). It is known that hERG channel blocking concentration of drugs is well linked to the plasma drug concentration when QT interval prolongation is observed in clinic (Webster et al., 2002). However, some multi-channel blockers have shown discrepancies between their hERG inhibitory potency and the actual potential of delayed ventricular repolarization (Tabo et al., 2007, 2008) because other ion channels are also involved in the configuration of cardiac action potentials (Kii et al., 2005). Thus, it is necessary to evaluate the effects of new chemical entities not only on hERG currents but also on cardiac repolarization periods like action potential duration (APD) and/or corrected QT (QTc) interval to clarify the risk of QT prolongation. In addition to the adverse effects related to the QT interval prolongation, studies on other vital parameters such as blood pressure, heart rate and other electrocardiogram (ECG) parameters are also indispensable for assessing cardiovascular safety.

The purpose of this study was to profile the cardiovascular safety of MA-2029 including QT prolongation risk. We examined its effects on blood pressure, heart rate, and ECG in conscious telemetered dogs. In addition, we performed in vitro electrophysiological studies on hERG tail currents recorded from stably transfected human embryonic kidney 293 (HEK293) cells and on action potential parameters in isolated guinea pig papillary muscles.

**MATERIALS AND METHODS**

**Drugs**

MA-2029 was synthesized in the organic chemistry laboratory of Chugai Pharmaceutical Co., Ltd. Cisapride was extracted from Risamol® fine granules (Mitsubishi Pharma Corp., Osaka, Japan).

For the in vitro electrophysiological studies, MA-2029 was dissolved in a perfusion solution (bath or Tyrode’s, described below in detail) to make 100 μg/ml and then serially diluted to 10 and 1 μg/ml. Cisapride was dissolved in dimethyl sulfoxide (DMSO: Wako Pure Chemical Industries, Ltd., Osaka, Japan or Sigma Chemical Co., St. Louis, MO, USA) to make 100 mg/ml and the DMSO-diluted stock solution was then diluted 1000-fold with the perfusion solution to achieve 100 ng/ml, for a final concentration of 0.1% (v/v) DMSO in the perfusate.

**hERG study**

HEK293 cells stably expressing the hERG potassium channel (University of Wisconsin) were used. The cells were maintained and continuously passaged using minimum essential medium supplemented with 10% fetal bovine serum, 1% non-essential amino acids, 1% sodium pyruvate, and 0.4 mg/ml geneticin. For the electrophysiological experiments, the cells were plated onto sterile glass coverslips in 35-mm² dishes containing 3 ml medium, excluding geneticin, at a density of 0.7-1.3 × 10⁵ cells per dish.

The cells were transferred to a recording chamber and continuously perfused at room temperature with a bath solution (mM; NaCl: 137, KCl: 4, CaCl₂: 1.8, MgCl₂: 1.0, D-glucose: 10, HEPES: 10, and adjusted to pH 7.4 with NaOH) at a rate of 1-2 ml/min. Ionic currents were measured under a whole-cell voltage clamp using the patch-clamp technique with Axopatch 1-B amplifier (Molecular Devices Corp., Sunnyvale, CA, USA). Recording electrodes (actual resistance range: 1.7-5.0 MΩ) were filled with a pipette solution (mM; KCl: 130, MgCl₂: 1.0, EGTA: 5, MgATP: 5, HEPES: 10, and adjusted to pH 7.2 with KOH). Voltage clamp pulses and data acquisition were controlled by computer software (pClamp 8, Molecular Devices Corp.). Once a stable patch had been achieved, cells were initially clamped at a holding potential of ~80 mV. hERG potassium currents were elicited by voltage pulses to +20 mV for 4.8 sec, −50 mV to form a hERG tail current for 5 sec and returned to the holding potential, giving a total pulse length of 15 sec.

After the voltage protocol was run a minimum of 10 times, either MA-2029 (1, 10, or 100 μg/ml), cisapride (100 ng/ml), or vehicle control (bath solution with or without 0.1% DMSO) was applied for approximately 10 min. Each test substance or vehicle was applied to six different cells. The effects of the test compounds or vehicles on hERG currents were examined at the peak of tail currents using the average current of 4 sequential voltage pulses before application and 10 min after application.
Cardiovascular safety profile of MA-2029.

**Action potential study**

Seventeen male Hartley strain guinea pigs (3-4 weeks old, 255-313 g) obtained from Japan SLC, Inc. (Shizuoka, Japan) were used in this experiment. All animal procedures were conducted in accordance with the ethical guidance for animal care. The experimental protocol was approved by the Animal Care Committee of the institute. Animal rooms were maintained under the following conditions: temperature, 22 ± 2°C; relative humidity, 55 ± 15%; and a 12-hr light period (07:00-19:00). The animals were anesthetized by ether inhalation and exsanguinated before removal of the heart. Immediately thereafter, the papillary muscle was isolated from the right ventricle and perfused at approximately 5 ml/min with Tyrode’s solution (mM; NaCl: 125, KCl: 4, NaHCO3: 25, NaH2PO4: 1.8, MgCl2: 0.5, CaCl2: 2.7, D-glucose: 5.5) that was aerated with a gas mixture (95% O2 + 5% CO2). The preparations were electrically stimulated by square waves (voltage: 1.5 times the threshold, 1.5-6 V, duration: 1 msec, frequency: 1 Hz) generated by an electrical stimulation apparatus (SEN-3301, Nihon Kohden, Tokyo, Japan) via an isolator (SS-403J, Nihon Kohden).

A glass microelectrode filled with 3 M KCl (resistance: 20-30 MΩ) was inserted into the papillary muscle cell and the transmembrane action potential was passed through amplifiers (MEZ-7200 and AVB-11A, Nihon Kohden) and recorded on a personal computer. In addition, the action potential was input to a differential apparatus (SS-1987, Nihon Kohden) and an amplifier (AVM-11, Nihon Kohden) to obtain the differential wave of action potential to measure the maximal upstroke velocity (V_{max}). The action potential parameters measured were resting membrane potential (RMP), action potential amplitude (APA), V_{max}, and action potential durations at 50% (APD_{50}) and 90% (APD_{90}) of repolarization, and were analyzed using computer software (pClamp 8, Molecular Devices Corp.).

After equilibrating the action potential parameters, either MA-2029 (1, 10, or 100 μg/ml), or vehicle control (Tyrode’s solution with or without 0.1% DMSO) was applied for 30 min. Each test substance or vehicle control was applied to six different preparations. The electrophysiological effects of test compounds or vehicles on the action potentials were examined using the average of 10 waveforms before application and 30 min after application.

**Cardiovascular study**

Four male beagle dogs (7-9-month old, 9.4-11.3 kg) were used in this experiment. All animal procedures were conducted in accordance with the ethical guidance for animal care for the facility and the experimental protocol was approved by the Animal Care Committee of the institute. The experimental animals were housed in individual stainless steel cages (W: 70 cm, D: 110 cm, H: 84 cm) in an air-conditioned room under the following conditions: temperature, 22 ± 3°C; relative humidity, 50 ± 20%; and a 12-hr light period (06:00-18:00). Approximately 300 g of solid diet was given daily and drinking water was provided ad libitum. Animals were surgically instrumented with telemetry transmitters (TL11M2-D70-PCT, Data Sciences International, St. Paul, MN, USA) in the right abdominal subcutaneous area under pentobarbital anesthesia. A catheter for measuring blood pressure was passed subcutaneously and inserted into the abdominal aorta via the femoral artery. The telemetered animals were given penicillin intramuscularly post surgery and were allowed to recover for at least 3 weeks before experimentation. The ECG was measured with M-X and R-L leads using a Holter electrocardiograph (QR1300, Fukuda M.E., Tokyo, Japan).

MA-2029 (0, 3, 30 or 300 mg/kg) was administered orally by capsule between 13:00 and 14:00. Each dosing was conducted with one-week intervals according to a dose-escalation program. Measurements were taken under quiet conditions in an animal room remote from the data acquisition, analysis, and storage room. Entry to the animal room was restricted except for physical checks, dosing, feeding, and blood collection from the animals, and cleanup of the room and cages. Diet was supplied after completion of data recordings and blood collection at the time point of 4 hr after dosing. Blood pressure and heart rate (HR) were recorded and analyzed by a data acquisition system (ART: Data Sciences International). Mean blood pressure (MBP), HR, and ECG were analyzed before and 1, 2, 3, 4, 6, 8, 12, and 24 hr after dosing. The values of MBP and HR at each time point were the average of values obtained over 20 sec every 5 min during the hour prior to the time point. ECG waveforms over 5 serial beats at each time point were used to analyze PR, QT and RR intervals and QRS duration using an analysis system (QS-2200, Fukuda M.E.). The QT interval was corrected for RR interval (QTc interval) using Fridericia’s formula (Fridericia et al., 1920), one of the most common correction formulas for conscious dogs (Toyoshima et al., 2005).

**Statistical analysis**

All data are expressed as mean ± S.D. The baseline values for each parameter in hERG, action potential and cardiovascular studies were compared among test groups using one-way analysis of variance (ANOVA). For all parameters obtained in the three studies, percent change
from the baseline was calculated. The MA-2029 groups were compared with vehicle control by parametric or non-parametric Dunnett’s test after confirming homogeneity using Bartlett’s test. The cisapride group was compared with vehicle control by Student’s or Welch’s t-test after confirming homogeneity using an F-test.

All statistics were calculated using the SAS system (Version 8.2, SAS Institute, Cary, NC, USA) and values of \( p < 0.05 \) were considered significant.

**RESULTS**

There were no significant differences among test groups of baseline values in any of the parameters obtained from hERG (amplitude of tail current), action potential (RMP, APA, \( V_{max} \), APD\(_{50}\), APD\(_{90}\)), or cardiovascular studies (MBP, HR and ECG parameters).

**Effects of MA-2029 and cisapride on hERG currents**

Table 1 shows the effects of MA-2029 (1, 10, or 100 \( \mu \)g/ml) and cisapride (100 ng/ml) on hERG tail currents and Fig. 2 shows a typical tracing of hERG current before and after application of MA-2029 at 100 \( \mu \)g/ml. Solvent controls for MA-2029 (bath solution) and for cisapride (0.1% DMSO) produced run-downs of tail currents of \(-21.0\%\) and \(-15.1\%\), respectively, from the baseline. Compared with the solvent control, MA-2029 had no effect on hERG tail current at 1 and 10 \( \mu \)g/ml (\(-15.6\%\) and \(-26.4\%,\) respectively) but apparently inhibited it at 100 \( \mu \)g/ml (\(-84.5\%\)). Cisapride (100 ng/ml), the reference substance, showed almost complete inhibition of tail current.

**Table 1.** Effects of MA-2029 and cisapride on hERG tail current in hERG-transfected HEK293 cells

<table>
<thead>
<tr>
<th>Drug</th>
<th>Concentration (( \mu )g/ml)</th>
<th>Tail current (% change from baseline)</th>
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<tbody>
<tr>
<td>Solvent*</td>
<td>0</td>
<td>(-21.0 \pm 2.5)</td>
</tr>
<tr>
<td>MA-2029</td>
<td>1</td>
<td>(-15.6 \pm 5.8)</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>(-26.4 \pm 7.9)</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>(-84.5 \pm 6.6**)</td>
</tr>
<tr>
<td>Solvent*</td>
<td>0</td>
<td>(-15.1 \pm 8.1)</td>
</tr>
<tr>
<td>Cisapride</td>
<td>0.1</td>
<td>(-95.6 \pm 2.3**)</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± S.D. of 6 different cells. hERG: human ether-a-go-go related gene, HEK293: human embryonic kidney 293. *Solvent for MA-2029 or cisapride was bath solution without or with 0.1% DMSO. The pre-values of MA-2029 at 0, 1, 10, 100 \( \mu \)g/ml and cisapride at 0, 0.1 \( \mu \)g/ml were 1.530 ± 0.636, 1.353 ± 0.654, 1.920 ± 0.483, 1.582 ± 0.514, 1.530 ± 0.699, 1.180 ± 0.614 nA, respectively. **Significantly different from the solvent of MA-2029 (\( p < 0.01 \)) by the Dunnett’s test. ##Significantly different from the solvent of cisapride (\( p < 0.01 \)) by the Welch’s t-test.

**Fig. 2.** Representative recordings of hERG current before (Pre) and after application of MA-2029 at 100 \( \mu \)g/ml in hERG-transfected HEK293 cells. The pulse protocol is shown on the bottom. HEK293: human embryonic kidney 293.
The effects of MA-2029 (1, 10, or 100 μg/ml) and cisapride (100 ng/ml) on action potential parameters in guinea pig papillary muscle are summarized in Table 2 and representative examples before and after application of MA-2029 at 10 and 100 μg/ml are illustrated in Fig. 3. Solvent controls for both MA-2029 (Tyrode’s solution) and cisapride (0.1% DMSO) had no effect on any of the parameters. MA-2029 at 1 μg/ml also showed no effect on any parameters. However, compared with the solvent control, MA-2029 at 10 and 100 μg/ml shortened APD_{50} by −13.6% and −43.6%, respectively, from the baseline and APD_{90} by −10.4% and −32.6%, respectively, in a concentration-dependent manner. In addition to the APD shortening, at 100 μg/ml, V_{max}, APA, and RMP significantly decreased by −60.5%, −18.4% and −3.1%, respectively. On the other hand, cisapride at 100 ng/ml caused prolongations of APD_{50} and APD_{90} by 12.6% and 15.4%, respectively, without affecting V_{max}, APA, or RMP.

**Effect of MA-2029 on cardiovascular system**

Fig. 4 shows the time courses for percent changes from baseline of MBP, HR, and ECG parameters (PR, QT, QTc intervals, and QRS duration) after oral administration of MA-2029 (0, 3, 30, or 300 mg/kg) after oral administration of MA-2029 (0, 3, 30, or 300 mg/kg). Vehicle control showed transient increases in MBP and HR after administration. Compared with the vehicle control, MA-2029 had no effect on any parameters at 3 mg/kg. MA-2029 at 30 mg/kg caused slight decreases in MBP, QT, and QTc intervals and a slight increase in PR interval and at 300 mg/kg showed an apparent PR prolongation of 33.1% from the baseline at 4 hr post-dosing in addition to slight hypotension and slight shortening of QT and QTc intervals. MA-2029 at 300 mg/kg also had a tendency to increase HR, although not significantly.

Plasma concentrations at 4 hr after administration of MA-2029 at 3, 30 and 300 mg/kg were 0.066 ± 0.037, 2.10 ± 0.65, and 6.79 ± 1.37 μg/ml, respectively.

**DISCUSSION**

The aim of this study was to clarify the cardiovascular safety profile of MA-2029, a pure motilin receptor antagonist highly expected to be an effective treatment for IBS. Concerning proarrhythmic potential, MA-2029 has a low risk of QT interval prolongation, because it shortened APD in guinea pig papillary muscle and QTc interval in conscious dogs at supra-effective concentrations although apparent hERG inhibition was observed at the highest concentration (100 μg/ml) tested in this study. MA-2029 not only shortened the cardiac repolarization period, it also caused slight hypotension and slight PR interval prolongation after oral administration of 30 mg/kg in conscious dogs. The plasma concentration at 4 hr after administration of 30 mg/kg was 2.10 μg/ml, 200-

<table>
<thead>
<tr>
<th>Drug</th>
<th>Concentration (μg/ml)</th>
<th>% change from baseline</th>
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<tr>
<td></td>
<td></td>
<td>RMP</td>
</tr>
<tr>
<td>Solvent*</td>
<td>0</td>
<td>0.6 ± 1.3</td>
</tr>
<tr>
<td>MA-2029</td>
<td>1</td>
<td>−0.7 ± 0.9</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>0.0 ± 1.3</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>−3.1 ± 1.6**</td>
</tr>
<tr>
<td>Solvent*</td>
<td>0</td>
<td>−0.4 ± 0.9</td>
</tr>
<tr>
<td>Cisapride</td>
<td>0.1</td>
<td>0.0 ± 0.7</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± S.D. of six different preparations.

RMP: resting membrane potential, APA: action potential amplitude, V_{max}: maximum rising velocity, APD_{50} or APD_{90}: action potential duration at 50% or 90% repolarization level.

*Solvent for MA-2029 or cisapride was Tyrode’s solution without or with 0.1% DMSO.

The pre-values of MA-2029 at 0, 1, 10, 100 μg/ml and cisapride at 0, 0.1 μg/ml were −93 ± 1, −93 ± 1, −94 ± 1, −93 ± 1, −93 ± 2, −93 ± 1 mV for RMP, 132 ± 2, 133 ± 2, 134 ± 2, 133 ± 2, 133 ± 2 ± mV for APA, 217 ± 26, 222 ± 21, 218 ± 13, 214 ± 14, 221 ± 25, 227 ± 26 mV/sec for V_{max}, 127 ± 10, 127 ± 7, 130 ± 5, 130 ± 12, 129 ± 8, 129 ± 7 msec for APD_{50}, 158 ± 11, 158 ± 6, 159 ± 8, 161 ± 12, 158 ± 9, 158 ± 8 msec for APD_{90}, respectively.

*: p < 0.05, **: p < 0.01; Significantly different from the solvent of MA-2029 by the Dunnett’s test

Significantly different from the solvent of cisapride (p < 0.01) by the Welch’s t-test.

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The shortening of APD at 10 μg/ml in papillary muscle in vitro was quite consistent with QTc shortening in conscious dogs observed with 30 and 300 mg/kg dosings. The shortened APD/QTc was likely caused by a Ca²⁺ channel blockade because MA-2029 at 5.6 and 56 μg/ml inhibited Ca²⁺ currents in isolated guinea pig myocytes by −42.6 and −86.7%, respectively (n = 4, data not shown). In addition, the hypotension and the increase in PR interval observed after administration of 30 and 300 mg/kg are typical changes induced by inhibition of the Ca²⁺ channel. MA-2029 at 100 μg/ml drastically shortened the APD with decreases of $V_{\text{max}}$ and APA despite almost complete hERG channel blockade. The suppression of $V_{\text{max}}$ is generally attributed to inhibition of the fast Na⁺ current, thus contributing to reduction of the APA (Hayashi et al., 2005). These findings suggest that MA-2029 would inhibit multiple cardiac channels at concentrations (10-100 μg/ml) beyond its effective concentration (10.5 ng/ml) and inhibition of Ca²⁺ and Na⁺ channels would offset the expected prolonging effect on APD and QTc intervals caused by hERG/IKr inhibitory action.

Since inhibition of the hERG current and prolongation of QT interval are only markers of the proarrhythmic risk of drugs, tests that can predict the actual proarrhythmic potential of drugs are required. Therefore, we conducted two additional studies. One was an electrophysiological cardiovascular study using halothane-anesthetized dogs (Sugiyama and Hashimoto, 1998) to measure the terminal repolarization period (TRP) of the right endocardial heart. The other was a proarrhythmic study using anesthetized rabbits to evaluate the proarrhythmic potential of test drugs using a concomitant infusion of the $\alpha_1$-adrenoceptor agonist methoxamine that may promote triggered activity and increase TdP susceptibility (Carlsson et al., 1997; Akita et al., 2004). MA-2029 did not increase the TRP at doses up to 21.1 μg/ml in dogs (n = 6) and showed no TdP at up to 4.63 μg/ml in rabbits (n = 6, data not shown for either study). On the other hand, cisapride caused a distinct increase in the TRP and induced TdP under the same test conditions (Kimura et al., 2007a, 2007b).

In conclusion, the results of this cardiovascular or electrophysiological study suggest that MA-2029 has a wide safety margin. Concerning QT prolongation risk in particular, MA-2029 showed no potential for QT prolongation because it shortened APD and QTc interval, contrary to the reference drug cisapride. Therefore, MA-2029 is a novel motilin receptor antagonist highly expected to be an effective treatment for IBS with lower cardiovascular risk than cisapride.
Cardiovascular safety profile of MA-2029.

Fig. 4. Effect of MA-2029 on mean blood pressure (A), heart rate (B), PR interval (C), QRS duration (D), QT interval (E) and QTc interval (F) in conscious telemetered dogs. MA-2029 (●: 3 mg/kg; Δ: 30 mg/kg; ■: 300 mg/kg) or vehicle (○: capsule) was orally administered. Each point represents the mean ± S.D. of 4 animals. The pre-values of MA-2029 at 0, 3, 30, 300 mg/kg were 105 ± 8, 106 ± 9, 112 ± 14, 103 ± 10 mmHg for MBP, 88 ± 16, 91 ± 7, 103 ± 20, 77 ± 10 beats/min for HR, 87 ± 8, 92 ± 5, 91 ± 6, 89 ± 7 msec for PR, 58 ± 10, 57 ± 5, 58 ± 6, 61 ± 5 msec for QRS, 212 ± 13, 215 ± 21, 228 ± 21, 235 ± 23 msec for QT, 233 ± 12, 233 ± 12, 256 ± 21, 251 ± 17 msec for QTc, respectively. Statistical significance: *p < 0.05 and **p < 0.01 versus time-matched vehicle control.
ACKNOWLEDGMENTS

The authors would like to thank Dr. Clare Quinn from Quintiles Ltd., Mr. Hideto Amano from Mitsubishi Chemical Safety Institute Ltd., and Dr. Noritsugu Shimizu from Fuji Biomedix Co., Ltd. for their excellent technical assistance and technical support of data analysis.

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