Model-Dependent Effects of the Gap Junction Conduction–Enhancing Antiarrhythmic Peptide Rotigaptide (ZP123) on Experimental Atrial Fibrillation in Dogs

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Background—Abnormal intercellular communication caused by connexin dysfunction may be involved in atrial fibrillation (AF). The present study assessed the effect of the gap junctional conduction–enhancing peptide rotigaptide on AF maintenance in substrates that result from congestive heart failure induced by 2-week ventricular tachypacing (240 bpm), atrial tachypacing (ATP; 400 bpm for 3 to 6 weeks), and isolated atrial myocardial ischemia.

Methods and Results—Electrophysiological study and epicardial mapping were performed before and after rotigaptide administration in dogs with ATP and congestive heart failure, as well as in similarly instrumented sham dogs that were not tachypaced. For atrial myocardial ischemia, dogs administered rotigaptide before myocardial ischemia were compared with no-drug myocardial ischemia controls. ATP significantly shortened the atrial effective refractory period ($P=0.003$) and increased AF duration ($P=0.008$), with AF lasting $\geq 3$ hours in all 6-week ATP animals. Rotigaptide increased conduction velocity in ATP dogs slightly but significantly ($P=0.04$) and did not affect the effective refractory period, AF duration, or atrial vulnerability. In dogs with congestive heart failure, rotigaptide also slightly increased conduction velocity ($P=0.046$) but failed to prevent AF promotion. Rotigaptide had no statistically significant effects in sham dogs. Myocardial ischemia alone increased AF duration and impaired conduction (based on conduction velocity across the ischemic border and indices of conduction heterogeneity). Rotigaptide prevented myocardial ischemia–induced conduction slowing and AF duration increases.

Conclusions—Rotigaptide improves conduction in various AF models but suppresses AF only for the acute ischemia substrate. These results define the atrial antiarrhythmic profile of a mechanistically novel antiarrhythmic drug and suggest that gap junction dysfunction may be more important in ischemic AF than in ATP remodeling or congestive heart failure substrates. (Circulation. 2007;115:310-318.)

Key Words: antiarrhythmia agents ▪ electrophysiology ▪ ion channels ▪ conduction ▪ electrocardiography ▪ ischemia

Atrial fibrillation (AF) is the most common arrhythmia in clinical practice. Although AF does occur in patients without evident heart disease, cardiac pathologies like congestive heart failure (CHF)$^{1,2}$ and coronary artery disease$^{2,3}$ are significant contributors to the occurrence and maintenance of AF. Traditional antiarrhythmic drugs have failed to provide safe and effective therapy for AF, and there has been increasing interest in targeting underlying atrial substrates with novel mechanism-based therapeutic approaches.$^4$

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Alterations in gap junction and connexin expression occur in AF-related remodeling in human$^{5,6}$ as well as animal$^{7-9}$ models. Abnormalities in connexin expression and function have also been reported in CHF,$^{10-12}$ and gap junction uncoupling plays a key role in arrhythmogenesis within the acutely ischemic myocardium.$^{13}$ Gap junctions and their connexin proteins that form intercellular communications are crucial for myocardial conduction,$^{14}$ and conduction abnormalities have been implicated in various AF paradigms, which include atrial tachycardia remodeling,$^{15}$ CHF,$^{16}$ and isolated acute atrial ischemia.$^{17}$

Recently, a stable antiarrhythmic-peptide analog, rotigaptide (formerly called ZP123), has been developed$^{18}$ and reported to increase junctional conductance and prevent ventricular tachycardia in a canine ventricular ischemia model.$^{19,20}$ Rotigaptide also has been reported to attenuate gap junction closure during experimental acidosis$^{21}$ and prevent atrial conduction slowing during metabolic stress.$^{22}$ Given the occurrence of connexin abnormalities and their potential...
pathophysiological role in AF, gap junction coupling promoters like rotigaptide might be useful for AF therapy. The present study was designed to assess the efficacy and electrophysiological actions of rotigaptide in 3 clinically relevant experimental AF-promoting paradigms: atrial tachycardia remodeling, CHF, and acute atrial ischemia.

Methods

Groups and Animal Preparations
All animal-handling procedures were approved by the Animal Research Ethics Committee of the Montreal Heart Institute and followed the guidelines of the Canadian Council on Animal Care. Thirty-seven mongrel dogs (18 to 45 kg; Laka, Inc, St-Basil-le-Grand, Quebec, Canada) were studied. Dogs were divided into the following groups: (1) atrial tachycardia–induced remodeling for 3 or 6 weeks (3W-atrial tachypacing [ATP] group, n = 5; 6W-ATP, n = 4); (2) atrial structural remodeling caused by CHF (ventricular tachypacing [VTP] dogs, n = 5); (3) isolated atrial ischemia (ischemia group, n = 13 total [7 ischemic control dogs and 6 dogs administered rotigaptide before baseline measurements and subsequent ischemia]); and (4) sham-operated nonpaced control dogs (n = 5 each for ATP and VTP, designated A-sham and V-sham, respectively).

ATP Group
Dogs were anesthetized with ketamine (5.3 mg/kg IV), diazepam (0.25 mg/kg IV), and halothane (1.5%). Unipolar pacing leads were inserted through jugular veins into the right ventricular (RV) apex and right atrial (RA) appendage and connected to pacemakers (Vitatron, Mississauga, Ontario, Canada) in subcutaneous pockets in the neck. A bipolar electrode was inserted into the RA for stimulation and recording during serial close- and open-chest electrophysiological studies. Atrophic ventricular block was created by radiofrequency-catheter ablation to avoid excessively rapid ventricular responses during ATP. The RV-demand pacemaker was programmed to 80 beats per minute (bpm). After 24 hours, a baseline close-chest electrophysiological study was performed, and then ATP was initiated at 400 bpm. A-sham animals were handled the same way but without pacemaker activation. ATP dogs were studied weekly to assess the time course of atrial effective refractory period (ERP) and AF duration (DAF) changes. One set of dogs was studied after 3 weeks of ATP, at a time when ERP changes reached a maximum, and a second set of dogs was studied when DAF reached a maximum, i.e., spontaneously sustained for >3 hours in closed-chest conditions, at 6 weeks of ATP.

CHF Group
Dogs were anesthetized with ketamine/diazepam/halothane as above. A unipolar pacing lead was inserted through a jugular vein into the RV apex and connected to a pacemaker in the neck. After 24-hour recovery, the ventricular pacemaker was programmed to capture the RV at 240 bpm. We confirmed that atrial rate was not affected by VTP in any dog at any monitoring point in the study. Dogs were subjected to VTP at 240 bpm for 2 weeks to create CHF, and then open-chest electrophysiological study was performed under morphine/α-chloralose anesthesia. V-sham animals were handled the same way but without tachypacing.

Ischemia Group
Dogs were anesthetized with morphine and α-chloralose and ventilated mechanically. A median sternotomy was performed and a pericardial cradle created. Isolated atrial ischemia was produced by double ligation of a branch of the right coronary artery that perfused the RA free wall but no ventricular tissue, as previously reported. To delineate atrial hypoperfusion and exclude ventricular ischemia, 6% thioflavin-S in 0.9%-NaCl was injected into a femoral vein at the end of the experiment. Two minutes later, the RA, left atrium (LA), and RV were removed. Hypoperfused regions were revealed as nonfluorescing areas under fluorescent light and traced for subse-quent quantification. No LA or RV hypoperfusion was seen in any dog.

Electrophysiological Study
In open-chest electrophysiological studies, the atrial tachy-pacemaker (for the ATP group) and the ventricular tachy-pacemaker (CHF group) were deactivated. Dogs were anesthetized with morphine (2 mg/kg SC) and α-chloralose (120 mg/kg IV, followed by 29.25 mg/kg per hour) and ventilated mechanically. Body tempera-ture was maintained at 37°C, and a femoral artery and both femoral veins were cannulated to monitor pressure and administer drugs. A median sternotomy was performed, and bipolar electrodes were hooked into the RA and LA appendages for recording and stimulation. The ERP was measured at various basic cycle lengths (BCLs; 150, 200, 250, 300, 360 ms) with 10 basic stimuli (S1) followed by premature S2 extrastimuli with 5-ms decrements. All stimuli were 2×threshold 2-ms pulses. The longest S1–S2 interval failing to capture defined the ERP. AF was induced with 1- to 10-second burst pacing (10 Hz, 4×threshold current). Silicon sheets that contained 240 bipolar electrodes were attached to the atria as previously described. Electrophysiological mapping was conducted with the Cardiomap system (Research Center, Sacré-Coeur Hospital and Biomedical Engineering Institute, École Polytechnique and Université de Montréal, Montreal, Canada). Programmed stimulation was performed at a BCL of 300 ms at 6 additional sites (RA and LA, posterior wall; RA and LA, inferior wall; RA and LA, Bachmann’s bundle) to evaluate atrial vulnerability to AF induction. Atrial vulnerability was determined as the percentage of atrial sites at which AF (>1 second) was induced by single extrastimuli. Conduc-tion velocity was determined from the regression of electrode distance on activation time as previously described. To estimate DAF in each dog, AF was induced 10 times for DAF <20 minutes and 5 times for 20- to 30-minute AF. Prolonged AF (>30 minutes) was terminated by direct-current electrical cardioversion. A 20-minute rest period was then allowed before measurements were continued. If prolonged AF was induced twice, no further AF induction was performed.

For closed-chest electrophysiological studies, dogs were anesthetized with ketamine (3.5 mg/kg IV), diazepam (0.25 mg/kg IV), and isoflurane (1.5%) and ventilated mechanically. ERP and DAF were measured. If AF persisted, dogs were left in AF to monitor spontaneous sustained DAF.

In ischemia dogs, ERP(s) of the RA appendage, RA posterior wall, and RA inferior wall were measured as described above at BCLs 150, 200, 250, 300, and 360 ms. The conduction times between adjacent electrodes across the ischemic border zone and in the nonischemic zone were measured to assess conduction speed and RA conduction indices based on phase-delay histogram analyses were obtained. DAF was measured as described above.

Study Protocols
Rotigaptide was administered as a loading dose of 14.6 μg/kg IV over 5 minutes, followed by a maintenance infusion at 14.04 μg/kg per hour. In ATP and CHF dogs, baseline electrophysiological measurements were obtained, and then rotigaptide was administered and measurements were repeated 25 minutes after drug infusion started. In ischemia dogs, a baseline electrophysiological study was performed, and then the right intermediate atrial artery was doubly ligated in the presence or absence of rotigaptide. Measurements were obtained at baseline (before coronary artery occlusion) and then at 0.5, 3, and 5 hours after occlusion.

Phase-Delay Histogram Analysis
Phase-delay analysis was performed in VTP, V-sham, and ischemia dogs to evaluate local conduction abnormalities. For V-sham and CHF dogs, all electrode sites over both atria were used. For ischemia dogs, only the RA electrode array, which included the entire ischemic area, was used, to avoid distortion of ischemia-induced changes by the many sites outside the ischemic zone on the other electrode arrays. In this analysis, a histogram of local phase-delay
times at each electrode site is obtained, and the histogram distribution properties are characterized. The median phase-delay value (P5) reflects overall conduction, and the P5/95 (phase-delay range between 5% and 95% of the phase-delay values in the histogram) represents the range of conduction delays, largely determined by values in the slowest-conducting zones. The P5/95, which indicates the conduction-delay range normalized to the P50 value, is an index of conduction heterogeneity independent of overall conduction speed changes.16,17 The P5/95 was used as the primary index of conduction heterogeneity to assess regional conduction-slowing abnormalities in the CHF and ischemia models.

Plasma Rotigaptide Concentration Measurements

Blood samples were obtained from each dog 8 minutes after the loading infusion, at a time expected to demonstrate pseudo-steady-state rotigaptide plasma levels,18–22 as well as 1 and 3 hours after start of drug-infusion. Plasma was removed and stored at −80°C for subsequent liquid chromatography and double mass spectroscopy.18

Statistical Analysis

Data are presented as mean±SEM. One-way ANOVAs were performed for hemodynamic data and plasma concentration. Three-way repeated-measures ANOVAs were conducted for ERP comparisons, and 2-way repeated-measures ANOVAs were conducted for conduction velocity in ATP and VTP groups, for conduction heterogeneity in VTP groups, for time-series data in the ischemia model, and for DAF and AF vulnerability data in ATP and VTP groups. DAF data were analyzed after normalization by log-transformation. When ANOVA revealed significant group effects, t tests were used to evaluate individual mean differences. The Bonferroni correction was applied to the probability values to adjust for multiple comparisons. A 2-tailed P<0.05 was considered statistically significant. Wherever possible, precise probability values are provided for statistical inferences; however, all probability values <0.001 were designated P<0.001.

All authors had full access to and take full responsibility for the integrity of the data. All authors have read and agree to the manuscript as written.

Results

Hemodynamic Variables and Plasma Concentrations

There were no statistically significant differences among groups in body weight and most hemodynamic variables (Table 1). The one exception was left ventricular end-diastolic pressure, which was significantly increased in VTP dogs. Rotigaptide did not affect hemodynamic indices in ATP and VTP dogs (data not shown). Although the plasma concentrations of 6W-ATP dogs and rotigaptide-treated atrial ischemia dogs were somewhat higher than for the other groups, all concentrations were above the minimum effective concentration of 1 nmol/L and of the same order as maximally effective concentrations (10 to 300 nmol/L) defined in previous studies.18–20,22 Concentrations remained stable within the maximally effective range over time (Table 1).

Changes in Electrophysiological Variables at Open-Chest Electrophysiological Study

We studied ATP dogs at 2 time points: after 3 weeks of ATP, when we found maximal ERP abbreviation, and at 6 weeks, when AF was sustained spontaneously for >3 hours in each dog. This allowed us to assess rotigaptide effects at the time of maximal ERP shortening, as well as at a later time when “second factors” came into play.23

Electrophysiological variables during open-chest electrophysiological study in 3W-ATP dogs are illustrated in Figure 1. ERP was shortened significantly by ATP compared with A-sham dogs, and ERP rate-adaptation was eliminated in both RA (Figure 1A) and LA (Figure 1B). Rotigaptide had no significant effects on ERPs (P>0.20 in A-sham and P>0.09 in ATP). Rotigaptide did not affect conduction velocity in A-sham dogs (Figure 1C and Figure 1D); however, the drug slightly but significantly increased conduction velocity in 3W-ATP dogs in both RA and LA.

Electrophysiological variables during open-chest electrophysiological study in VTP dogs are shown in Figure 2. There were no statistically significant differences in ERP values among groups in either RA (Figure 2A; P>0.40) or LA (Figure 2B; P>0.14). Conduction velocity was not changed by rotigaptide in V-sham dogs but was significantly increased in both RA (Figure 2C) and LA (Figure 2D) in VTP dogs.

Figure 3 shows electrophysiological variables in atrial ischemia dogs. ERPs were not significantly altered by acute ischemia, whether measured in nonischemic tissue (RA appendage, Figure 3A, left panel, P=0.47), the periphery of the ischemic zone (RA posterior wall, middle panel, P=0.06), or the central ischemic zone (RA inferior wall, right
Rotigaptide did not affect ERPs in the absence or presence of ischemia. Figure 4 illustrates changes in RA activation induced by myocardial ischemia in one preparation in the absence (top row) and another in the presence (bottom row) of rotigaptide. Atrial ischemia caused clear conduction slowing as indicated by isochrone crowding across the ischemic border (the hypoperfused zone as assessed by thioflavin perfusion in each preparation is shown by the shaded area). In the presence of rotigaptide, ischemic conduction changes were greatly attenuated. Mean conduction velocities across the ischemic border and in nonischemic tissue are shown in Figure 3B and 3C. Ischemia progressively slowed conduction into the ischemic zone in the absence of rotigaptide (□ in Figure 3B). In the presence of the drug (□ in Figure 3B), conduction slowing was greatly attenuated. There were no significant conduction changes in nonischemic tissue (Figure 3C).

The assessment of ischemic conduction changes shown in Figure 3B depends on ischemic zone location. To evaluate ischemia-induced conduction changes in a more objective and independent fashion, we subjected activation time data from RA electrode sites to phase-delay histogram analysis as described above to provide the results shown in Figure 5A and 5B. Acute ischemia produced statistically significant increases in P5–95/P50 at both BCLs in the absence of rotigaptide (□). At a BCL of 150 ms, ischemia significantly increased the P5–95/P50 index even in the presence of rotigaptide (□), but the changes were significantly attenuated compared with values in the drug’s absence. Ischemia failed to signif-
significantly increase P5–95/P50 in the presence of rotigaptide at a BCL of 300 ms.

We also applied the phase-delay analysis to data from all RA and LA electrode sites in V-sham and VTP dogs. The P5–95/P50 increases substantially with VTP-induced CHF, which reflects localized conduction slowing believed to be important in the pathogenesis of CHF-related AF. The P5–95/P50 was significantly increased in VTP dogs compared with V-sham dogs (Figure 5C). The mean P5–95/P50 values in CHF dogs were slightly lower after rotigaptide administration, but there were no statistically significant differences between results before versus after rotigaptide (P = 0.29 at BCL 150 ms and P = 0.22 at BCL 300 ms).

Figure 6 shows AF-promotion indices during the open-chest electrophysiological study in both 3W-ATP (Figure 6A and 6B) and VTP (Figure 6C and 6D) dogs. In sham-operated dogs, AF was short-lived and always terminated spontaneously within 5 minutes. Reproducible prolonged AF (>30 minutes) was induced during the open-chest electrophysiological study in 3 of 5 dogs in the 3W-ATP group and in 4 of 5 dogs in the VTP group. In 3W-ATP dogs, mean burst-pacing–induced AF duration was increased to >700 seconds and rotigaptide administration did not affect AF duration (Figure 6A). Atrial vulnerability was also significantly increased in 3W-ATP dogs, with AF inducible by single extrastimuli at >80% of sites. Rotigaptide infusion did not significantly affect atrial vulnerability (Figure 6B), which remained markedly increased relative to control. In VTP dogs, mean AF duration was increased to >1300 seconds and was not significantly affected by rotigaptide (Figure 6C, P = 0.23). As previously observed, VTP did not significantly increase atrial vulnerability (Figure 6D, P = 0.37), which remained low and not different from nonpaced dogs in the presence of rotigaptide (P = 0.19).

The development of AF promotion after the induction of acute atrial ischemia is shown as a function of time after coronary artery ligation in Figure 7. DAF increased progressively with ischemic time in dogs not administered rotigaptide ( ). Dogs that received rotigaptide before coronary artery occlusion showed marked attenuation of AF promotion ( ).

The results in 6W-ATP dogs are illustrated in Figure 8. Rotigaptide administration had no significant effects on ERPs (Figure 8A and 8B) in either A-sham (P > 0.18) or 6W-ATP dogs (P > 0.43). All 6W-ATP dogs showed sustained AF that required cardioversion for termination, and sustained AF that required cardioversion was seen in all dogs after rotigaptide treatment (Figure 8C, left). Atrial vulnerability was very high in 6W-ATP dogs and was not changed by rotigaptide (Figure 8C, right). Rotigaptide did not significantly alter conduction velocity in A-sham dogs and slightly but significantly increased conduction velocity in 6W-ATP dogs in both RA (Figure 8D) and LA (Figure 8E).

Discussion

In the present study, we evaluated the effects of a novel gap junction conduction–enhancing antiarrhythmic peptide, roti-
Rotigaptide, in 3 clinically relevant AF substrates. Rotigaptide suppressed AF only for the acute ischemic substrate, although some evidence of improved atrial conduction was observed in each of the models. The AF suppression effect of rotigaptide during acute atrial ischemia was associated with significant attenuation of ischemia-induced conduction heterogeneity and conduction slowing in the ischemic zone.

Relation to Previous Observations on the Effects of Antiarrhythmic Peptides
Rotigaptide is a novel and highly stable analog of an antiarhythmic peptide first described in 1980. It prevents acidosis-induced ventricular conduction slowing and reentrant ventricular tachycardia in dogs with acute myocardial infarction, effects attributable to improved gap junction–mediated intercellular communication. In addition, rotigaptide prevents spontaneous ventricular tachyarrhythmias induced in dogs by ischemia-reperfusion and delays the onset of ouabain-induced conduction block in mice. However, rotigaptide is ineffective against focal ventricular tachyarrhythmias in dogs with myocardial infarction and does not suppress triggered arrhythmias in vitro.

Figure 5. Results of conduction heterogeneity analysis performed on the basis of phase-delay maps. The conduction heterogeneity index (P5–95/P50) is an index of localized conduction slowing that is independent of overall conduction velocity changes. A, Comparison of conduction heterogeneity index in ischemia dogs without versus with rotigaptide treatment, as measured at a BCL of 150 ms as a function of time. B, Same analysis as in A but at a BCL of 300 ms. C, Conduction heterogeneity index in VTP dogs as a function of BCL before VTP versus after RTG treatment. Abbreviations as in Figures 2 and 3.

Figure 6. Indices of AF promotion before and after rotigaptide treatment at the final open-chest study in 3W-ATP (A and B) and VTP (C and D) dogs. A, Mean ± SEM. AF duration (DAF), as determined with 5 to 10 AF inductions in each dog, for A-sham and 3W-ATP dogs before and after rotigaptide. B, Atrial vulnerability (% of sites at which single extrastimuli induced AF) for A-sham and 3W-ATP dogs before and after rotigaptide. C, Mean ± SEM. DAF for V-sham and VTP dogs before and after rotigaptide.

Conduction heterogeneity index

![Graph A](image)

**Figure 5.** Results of conduction heterogeneity analysis performed on the basis of phase-delay maps. The conduction heterogeneity index (P5–95/P50) is an index of localized conduction slowing that is independent of overall conduction velocity changes. A, Comparison of conduction heterogeneity index in ischemia dogs without versus with rotigaptide treatment, as measured at a BCL of 150 ms as a function of time. B, Same analysis as in A but at a BCL of 300 ms. C, Conduction heterogeneity index in VTP dogs as a function of BCL before VTP versus after RTG treatment. Abbreviations as in Figures 2 and 3.

![Graph B](image)

![Graph C](image)

Atrial vulnerability (%) before and after rotigaptide treatment in 3W-ATP and VTP dogs. A, Mean ± SEM. AF duration (DAF), as determined with 5 to 10 AF inductions in each dog, for A-sham and 3W-ATP dogs before and after rotigaptide. B, Atrial vulnerability (% of sites at which single extrastimuli induced AF) for A-sham and 3W-ATP dogs before and after rotigaptide. C, Mean ± SEM. DAF for V-sham and VTP dogs before and after rotigaptide.
of ion channels and receptors and does not affect heterologously expressed HERG current.22

The mechanisms by which rotigaptide alters connexin function are unclear. Rotigaptide may improve connexin function by protein kinase C–mediated phosphorylation.26 It also has been reported that rotigaptide suppresses dephosphorylation of serine residues in the C-terminal tail of connexin 43 in rat hearts subjected to ischemia.27 However, another study failed to observe changes in the phosphorylation status of connexin 43 in response to rotigaptide.28 Rotigaptide increases connexin 43 expression after 24 hours in neonatal rat cardiomyocytes,29 but such an effect is unlikely to explain changes after short-term intravenous administration.

Although rotigaptide’s antiarrhythmic effects have not been assessed in ischemic AF models, it has been found to prevent atrial conduction slowing induced by metabolic stress, a result compatible with our findings.22 A very recent publication reported that rotigaptide suppressed AF promotion in a model of mitral insufficiency but not in dogs with VTP-induced CHF, consistent with our observations in VTP dogs.30 In addition, rotigaptide has recently been found to accelerate conduction without altering AF inducibility in an arteriovenous shunt-induced, volume-overloaded rabbit model.31

We elected to analyze rotigaptide effects in a panel of 3 clinically relevant experimental AF models and noted modelspecific efficacy: prevention of AF promotion in the context of acute atrial ischemia, but inefficacy for substrates induced by CHF and atrial tachycardia remodeling. Furthermore, we examined 2 time points in ATP remodeling to assess rotigaptide effects both at maximum ERP shortening and at a time of longer-lasting sustained AF, which implies the presence of “second factors.”

Relation to AF Mechanisms and Potential Significance

Connexin abnormalities have been described in all of the experimental AF paradigms that we studied. In the atrial tachycardia remodeling context, a variety of findings (some discrepant) have been reported. Elvan et al showed upregulation of connexin 43 in dogs subjected to atrial tachypacing,7 whereas Van der Velden et al noted unchanged connexin 43 but spatially heterogeneous downregulation of connexin 40.8,9 In rats subjected to 24 hours of atrial tachypacing, connexins are redistributed toward lateral membranes.5 Clinical studies have shown that chronic AF with complex activation is associated with reduced connexin 40 immunofluorescence.12 On the basis of the findings that suggest reduced connexin expression with atrial remodeling, a beneficial effect of enhanced gap junctional coupling might have been expected in the atrial tachycardia–induced AF substrate. Our results with rotigaptide suggest that this is not the case.

Similar arguments pertain to CHF-related AF. Ventricular conduction slowing in CHF is caused in large part by
connexin dysfunction related to reduced expression and/or impaired phosphorylation.\textsuperscript{10–12} Atrial conduction slowing appears to play an important role in the AF substrate induced by CHF,\textsuperscript{16} and improved atrial conduction indices are likely significant in the beneficial effects of angiotensin-converting enzyme inhibition against CHF-related AF.\textsuperscript{33} If connexin dysfunction were important in CHF-related AF, reversal by rotigaptide might be expected to significantly improve conduction heterogeneity and reduce AF duration in the CHF substrate. We did not observe such an effect.

Atrial ischemia significantly promotes AF maintenance and is a candidate to contribute to AF occurrence in patients with chronic coronary artery disease, acute myocardial infarction, and post–cardiac surgery settings.\textsuperscript{17} Ischemic heart disease is one of the strongest predisposing factors to AF,\textsuperscript{34} and postoperative AF is a major clinical problem.\textsuperscript{35} Ischemia-induced gap junction uncoupling is thought to play a substantial role in ischemic conduction slowing and arrhythmogenesis.\textsuperscript{19–21} Rotigaptide significantly attenuated atrial conduction abnormalities and AF promotion induced by acute isolated atrial ischemia in the present studies. These observations support the potential utility of rotigaptide and compounds with similar mechanisms of action in ischemia-related AF and indicate the importance of gap junction uncoupling in ischemic AF, a subject that has received little attention to date.

\textbf{Potential Limitations}

The plasma concentrations of rotigaptide varied somewhat among groups. However, plasma levels in all groups and at all times were well within the maximally effective concentration range (10 to 300 nmol/L) defined in previous studies\textsuperscript{18–20,22} and remained relatively stable over time in each group. Furthermore, rotigaptide efficacy differences were not related to plasma concentrations: The highest concentrations were seen in 6W-ATP dogs, in which no efficacy was seen. Our findings are not consistent with the expectation that improvement of intercellular communication with rotigaptide would suppress AF in atrial tachycardia remodeling and CHF-related substrates, and they argue against an important role of gap junction uncoupling in these settings. However, our findings do not completely exclude a pathophysiological role for connexin abnormalities, because rotigaptide’s action may be insufficient to overcome the specific connexin dysfunction in these contexts, eg, changes caused by connexin redistribution, downregulation of expression, or dephosphorylation. Our observations ideally will lead to further studies of the specific nature of the beneficial actions of this class of peptides for different types of gap junction dysfunction, which may lead to an improved mechanistic understanding of therapeutic interventions on gap junction conduction as well as to a clearer comprehension of the role of gap junctions in pathological conditions like atrial tachycardia remodeling and CHF that promote AF.

\textbf{Conclusions}

The antiarrhythmic peptide rotigaptide improves atrial conduction in various experimental models of AF but suppresses AF only for the acute ischemia substrate. Our results define the atrial antiarrhythmic profile of a mechanistically novel antiarrhythmic drug and may contribute to new insights into mechanisms and disease-dependent therapeutic approaches for AF in patients with different underlying substrates.

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\textbf{Disclosures}

Dr Haugan was an employee of Zealand Pharma, which holds intellectual property interests in rotigaptide; Dr Hennan is an employee of Wyeth Pharmaceuticals, which holds intellectual property interests in rotigaptide. Dr Nattel received an honorarium from Wyeth for participating in a consultation meeting in 2004.

\textbf{References}


CLINICAL PERSPECTIVE

Atrial fibrillation (AF) is the most common sustained cardiac arrhythmia, and its prevalence is increasing with the aging of the population. Current drug therapy approaches have incomplete efficacy and/or disturbing adverse effect risks. A recently developed class of antiarrhythmic drugs with a very different mechanism of action from traditional antiarrhythmic interventions is the gap junction coupling enhancers, which are agents that increase cell-to-cell coupling via gap junctions. We studied the actions of one member of this class, rotigaptide, in 3 different experimental models of AF, induced in the presence of experimental congestive heart failure, atrial tachycardia remodeling (like that caused by AF itself), and isolated atrial ischemia. Previous data have suggested a role for gap junction dysfunction and conduction slowing in each of these models. Rotigaptide improved atrial conduction in all 3 models, although its effects were much more marked in the substrate caused by acute atrial ischemia. The compound had no effect on the AF maintenance caused by congestive heart failure or atrial tachycardia remodeling but markedly reduced the ability of acute atrial ischemia to create a substrate for persistent AF. Our results show that the effects of rotigaptide on AF depend very much on the underlying mechanism, with particularly good efficacy in ischemic AF. The results also suggest that gap junction dysfunction may be more important in AF associated with acute atrial ischemia than that caused by congestive heart failure and atrial tachycardia remodeling, and/or that the effects of gap junction coupling enhancers may depend on the specific mechanisms that underlie gap junction dysfunction.
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