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Electrocardiographic Effects of Zatosetron and Ondansetron, Two $5HT_3$ Receptor Antagonists, in Anesthetized Dogs

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ABSTRACT

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The pharmacology of 5-hydroxytryptamine$_3$ ($5HT_3$)-antagonists is an area under active investigation, and several agents of this class are currently under development for multiple therapeutic indications. Recently, two $5HT_3$ receptor antagonists of a tropane derived series, ICS 205 930 and zatosetron, have been shown to alter electrocardiographic properties of heart muscle. A prototypical, but structurally distinct (imidazole) $5HT_3$-antagonist, ondansetron, was examined for its comparative cardiovascular activity in anesthetized dogs at intravenous doses of 0.66–5.25 mg/kg. Similar to zatosetron, a significant, dose-dependent prolongation of the duration of the action potential of the electrocardiogram (Q-T_c interval) occurred following ondansetron exposure, with a maximum increase of 28%. Other cardiovascular parameters (heart rate, mean arterial pressure, pulmonary pressure, cardiac output, peripheral vascular resistance, stroke volume and work index) were essentially unchanged by ondansetron treatment. At equivalent $5HT_3$ blocking doses, both ondansetron and zatosetron prolonged the Q-T_c interval in anesthetized dogs similarly. However, for both compounds, the doses required to increase Q-T_c interval were higher than the doses required to demonstrate $5HT_3$ receptor blockade. The fact that ondansetron, an imidazole, exhibited electrophysiological effects on cardiac muscle like the $5HT_3$ receptor antagonists derived from tropane suggests that the electrocardiographic effects are related to some property shared by $5HT_3$ receptor antagonists rather than a property of the tropane structure.

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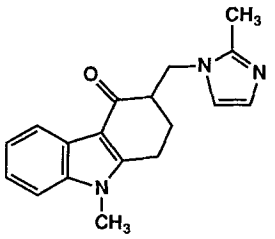
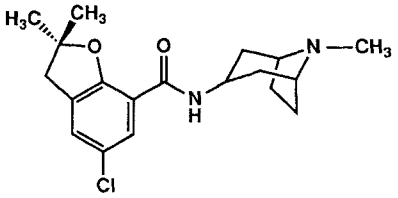


Ondansetron



Zatosetron

Fig. 1. Chemical structures of ondansetron and zatosetron. Ondansetron is chemically identified as (+/−)1,2,3,9-tetrahydro-9-methyl-3[(2-methyl-1H-imidazol-1-yl)methyl]-4H-carbozol-4-one. Zatosetron is chemically identified as endo-5-chloro-2,3-dihydro-2, 2-dimethyl-N-(8-methyl-8-azobicyclo[3.2.1] oct-3-yl)-7-benzofurancarboxamide (Z)-2-butenedioate (1:1).

Key words: Q-T_c interval prolongation, $5HT_3$-antagonism, electrophysiological effects

INTRODUCTION

5-hydroxytryptamine (5-HT_3) antagonists are currently under development for several therapeutic indications such as migraine, anxiety, psychosis, and emesis [King and Sanger, 1989; Upward et al., 1990]. Such multiple uses are derived from the fact that $5HT_3$ receptors have been identified in brain and on afferent nerves in cardiac and gastrointestinal tissue.

Studies using neuronal cell lines have strongly supported the concept that the $5HT_3$ receptor is associated with an ion channel, presumably a potassium ion channel [Bobker and Williams, 1990; Peters and Lambert, 1989; Steinberg and Robertson, 1990]. Based upon the presence of $5HT_3$ receptors in cardiac muscle, the association of potassium channels to the $5HT_3$ receptor, and the importance of ion channels with regard to the electrophysiology of cardiac muscle, it is not surprising the $5HT_3$ receptor antagonists can alter ionic currents in cardiac muscle. In fact, several reports have suggested that the tropane derivative, ICS 205 930 [Williams et al., 1985; Scholtysik et al., 1988], and a more recently identified tropanamine derivative $5HT_3$ receptor antagonist, zatosetron [Williams et al., 1991], can affect electrophysiological parameters of cardiac muscle.

Based on these initial observations, the present study was designed to determine if the cardiac electrophysiological effects of these compounds were restricted to compounds from the tropane chemical series or were due to a property common to other structurally unrelated $5HT_3$ receptor antagonists. This latter possibility would support the notion that modulation of the $5HT_3$ receptor or a closely related receptor can affect cardiac electrophysiology. In the present study, two potent and selective, but chemically distinct, $5HT_3$ receptor antagonists, zatosetron and ondansetron (Fig. 1), were compared for their ability to prolong the duration of the electrocardiographic action potential (Q-T_c interval) in anesthetized dogs.

MATERIALS AND METHODS

Male beagle dogs (Marshall Farms, North Rose, NY; 8–15 kg) were anesthetized with pentobarbital (30 mg/kg, IV) and ventilated with room air using a Harvard dual phase pump respirator. Lead II ECG was obtained using Grass E2 subdermal needle electrodes and a Beckman R711 recorder. The P-R, QRS, and Q-T intervals were measured from the ECG recording. To correct the Q-T interval for heart rate changes, the Q-T_c interval was calculated as the quotient of the Q-T interval divided by the square root of the R-R interval. Femoral flow was measured using a Carolina Medical flow meter and flow probe and a Beckman R711 recorder. A Swan-Ganz catheter was inserted through the femoral vein, inferior vena cava and right heart and into the pulmonary artery. Pulmonary arterial pressure and pulmonary capillary wedge pressure were measured through this catheter using a Spectramed P23XL pressure transducer. Cardiac output was measured by the thermal dilution method using the Swan-Ganz catheter and cardiac output computer [Fegler, 1954].

Ondansetron was administered at doses of 0.66, 1.31, 2.63, or 5.25 mg/kg by 15 min IV infusion. Zatosetron was administered at doses of 0.219, 0.438, 0.875, 1.75, or 3.50 mg/kg for comparative purposes. The control vehicle (Milli-Q water) was given at a dose volume of 1 ml/kg by IV infusion. Dogs, in groups of three, received drug or vehicle. Cardiovascular parameters were measured prior to dosing and at 2, 15, 30, and 45 min after dosing. In some experiments, an additional 7 min time point was monitored.

Calculations of body surface area, stroke work index, stroke volume, peripheral systemic vascular resistance, and pulmonary vascular resistance were made as follows: body surface area (BSA), in square meters, was determined using the equation

$$BSA = \frac{W^{2/3}}{10^3}$$

where W is the dog's weight in grams. Stroke work index (SWI) in g-m/beat/m^2 was calculated as follows:

$$SWI = \frac{CO}{HR \times BSA} \times (MP - PCWP) \times 0.0136$$

where CO is cardiac output in liters/min, HR is heart rate, MP is mean arterial pressure in mm Hg, PCWP is pulmonary capillary wedge pressure in mm Hg, and 0.0136 is the factor for converting to centimeter-gram-second (CGS) units. Peripheral systemic vascular resistance (VR) in dyne-sec-cm^{-5} was calculated as follows:

$$VR = \frac{MP \times 1{,}332}{Q}$$

where MP is mean pressure in mm Hg, 1332 is the factor for converting to CGS units, and Q is the blood flow in ml/sec. The pulmonary vascular resistance (PVR) in dyne-sec-cm^{-5} was calculated as follows:

$$PVR = \frac{(P - PCWP) \times 1{,}332}{Q}$$

where P is pulmonary pressure in mm Hg, PCWP is pulmonary capillary wedge pressure in mm Hg, and Q is the blood flow in ml/sec. Stroke volume was determined using the equation

$$SV = \frac{CO}{HR}$$

where CO is cardiac output in ml/min and HR is heart rate.

Data were analyzed using VMS SAS production release 5.16 at Eli Lilly and Company. At each time point, the mean percent changes from predrug (0 min) values for the treatment

groups were compared to the vehicle control group. Data were analyzed using an analysis of variance and a comparison of means for the treatment groups compared to the vehicle control group using the least significant difference method (multiple t-tests based on ANOVA) at the $P=0.05$ level [Cody and Smith, 1985].

Zatosetron and ondansetron were prepared by Joseph Krushinski at Eli Lilly and Company (Indianapolis, IN) and John Fairhurst at Eli Lilly and Company (Erl Wood, England), respectively. The chemical structures of ondansetron and zatosetron are shown in Figure 1.

RESULTS

Both zatosetron (0.22 to 3.5 mg/kg i.v.) and ondansetron (0.66 to 5.25 mg/kg i.v.) produced dose-dependent increases in Q-T_c interval in anesthetized dogs (Fig. 2). Neither compound produced any significant effects on other cardiovascular parameters including mean pressure, cardiac output, peripheral vascular resistance, femoral arterial flow, pulmonary wedge pressure, pulmonary arterial pressure, pulmonary vascular resistance, stroke volume, or stroke work index when administered in doses up to 5.25 mg/kg i.v. and 3.5 mg/kg i.v. for ondansetron and zatosetron, respectively. Furthermore, other electrocardiographic parameters were also unaltered by the same doses of either ondansetron or zatosetron, including QR interval and QRS duration.

Since zatosetron was approximately threefold more potent than ondansetron in blocking $5HT_3$ receptors after oral administration to rats [Cohen et al., 1989, 1990], a comparison of the elevation in Q-T_c interval produced by pharmacologically equivalent doses of ondansetron and zatosetron are indicated in Table 1. Actual changes in cardiovascular and electrocardiographic parameters at pharmacologically equivalent doses of zatosetron and ondansetron (0.88 and 2.63 mg/kg, respectively) are shown in Table 2. Furthermore, the time course for the alteration in Q-T_c interval was similar for pharmacologically equivalent doses of ondansetron and zatosetron (Fig. 3). It is apparent that both agents at doses that might be anticipated to produce equivalent blockade of $5HT_3$ receptors exerted similar prolongation of the Q-T_c interval.

DISCUSSION

The present studies documenting prolongation of Q-T_c interval with two $5HT_3$ receptor antagonists, ondansetron and zatosetron, extend the previous observation that other $5HT_3$ receptor antagonists such as ICS 205 930 can exert electrophysiological effects in cardiac muscle [Scholtysik, 1987; Scholtysik et al., 1988]; effects that have been suggested to result from alterations in potassium ion channel function [Steinberg and Robertson, 1990]. Furthermore, equivalent $5HT_3$ receptor antagonist doses of ondansetron and zatosetron produced equivalent increases in the magnitude and duration of Q-T_c prolongation in anesthetized dogs.

The doses required to observe these cardiac electrophysiological effects on Q-T_c interval were not associated with the development of cardiac arrhythmias in these dogs. In fact, others have suggested that the electrophysiological effects observed with another $5HT_3$ receptor antagonist, ICS 205 930, may account for its possible antifibrillatory activity [Williams et al., 1985]. It is also important to note, that the doses of these $5HT_3$ receptor antagonists required to prolong significantly Q-T_c interval are well in excess of the doses necessary to document effective $5HT_3$ receptor blockade in other studies [Cohen et al., 1989, 1990]. Thus, the relationship of these electrocardiographic effects to the clinical use of this class of compounds as antagonists of serotonin acting at $5HT_3$ receptors is unclear at this time. It is possible that in high doses these compounds may affect a $K+$ channel in cardiac muscle that is similar to but not identical with the $5HT_3$ receptor.

Nevertheless, the present study documents that the ability of $5HT_3$ receptor antagonists to increase Q-T_c interval is not restricted to tropane derivatives. Ondansetron, a structurally

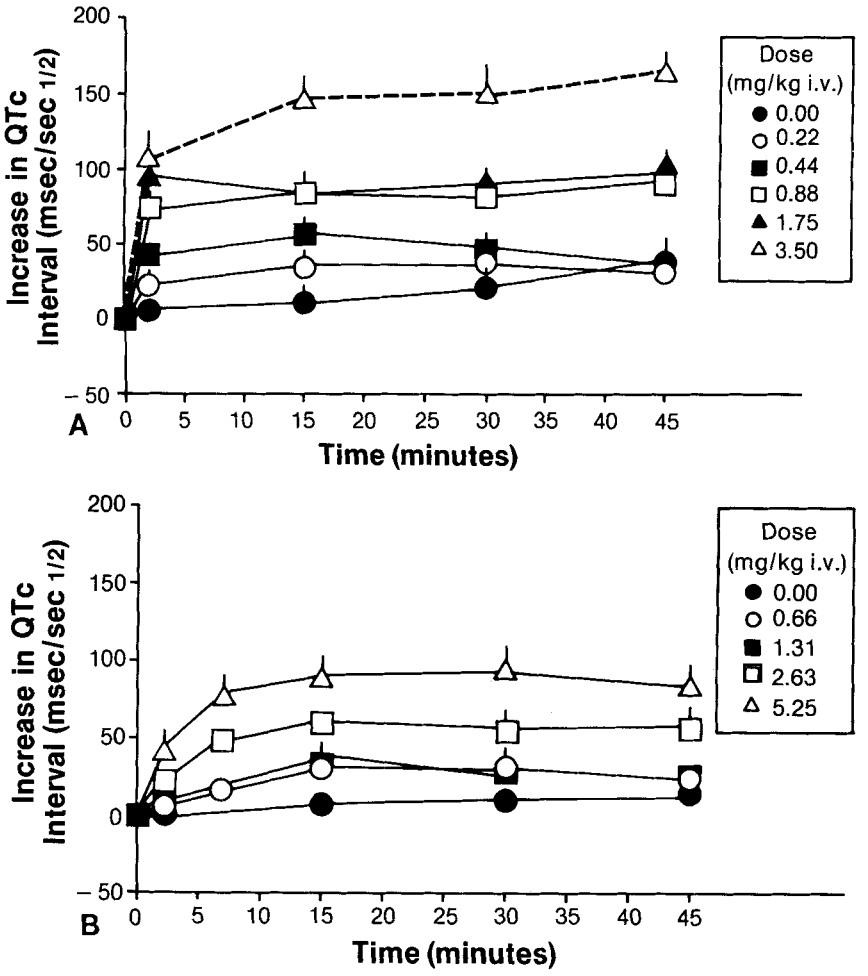


Fig. 2. **A:** Effect of zatosetron on QT_c interval in anesthetized beagle dogs. Shown are the actual increases from baseline QT_c interval values following intravenous infusion of zatosetron over 15 min at the doses shown. To correct the Q-T interval for heart rate changes, the Q-T_c interval was calculated as the quotient of the Q-T interval divided by the square root of the R-R interval. The mean ± S.E. of each group (n = 3) is displayed. **B:** Effect of ondansetron on QT_c interval in anesthetized beagle dogs. Shown are the actual increases from baseline QT_c interval values following intravenous infusion of ondansetron over 15 min at the doses shown. To correct the Q-T interval for heart rate changes, the Q-T_c interval was calculated as the quotient of the Q-T interval divided by the square root of the R-R interval. The mean ± S.E. of each group (n = 3) is displayed.

distinct imidazole $5HT_3$ receptor antagonist, clearly possesses a similar property, i.e., ondansetron dose-dependently prolonged the Q-T_c interval in anesthetized dogs. This observation provides some additional support of the possibility that blockade of $5HT_3$ receptors rather than some other effect of tropane derivatives is most related to the ability of these agents to increase Q-T_c interval. Furthermore, the widespread clinical development of ondansetron without reports of electrophysiological or cardiac effects reinforces the likelihood that these effects on Q-T_c interval require doses higher than are currently used clinically.

TABLE 1. Effects of Zatosetron and Ondansetron on Q-T_c Interval at Pharmacologically Equivalent Doses†

Group[a]	Compound	Dose	% change in Q-T_c interval[b]
Dose 0	Saline	1.0 ml/kg	3 ± 1
Dose 1	Zatosetron	0.22 mg/kg	10 ± 3
	Ondansetron	0.66 mg/kg	9 ± 0
Dose 2	Zatosetron	0.44 mg/kg	17 ± 3
	Ondansetron	1.31 mg/kg	12 ± 3
Dose 3	Zatosetron	0.88 mg/kg	20 ± 1*
	Ondansetron	2.63 mg/kg	19 ± 2*
Dose 4	Zatosetron	1.75 mg/kg	26 ± 2*
	Ondansetron	5.25 mg/kg	28 ± 5*

†Based upon an approximately three-fold difference in pharmacological ($5HT_3$)potency (zatosetron>ondansetron) [Cohen et al., 1990].
[a]Groups of 3–4 beagle dogs were administered saline, zatosetron, ondansetron by intravenous infusion (15 min).
[b]Percent changes are shown as mean values ± S.E. observed at the completion of dose administration (15 min).
*$P<0.5$, two-tailed Dunnett T on raw data relative to control values.

TABLE 2. Cardiovascular and Electrocardiographic Parameters in Anesthetized Dogs Following Administration of Zatosetron and Ondansetron at Pharmacologically Equivalent Doses†

	MP	HR	CO	PCWP	PULM	FLOW	PR
Vehicle	122.3 ± 3.7 (4.0)	116.7 ± 5.4 (−2.3)	1.6 ± 0.1 (0.1)	6.7 ± 0.3 (0.0)	14.7 ± 1.2 (0.0)	81.0 ± 26.1 (8.3)	89.7 ± 4.9 (−1.0)
Zatosetron	117.3 ± 7.1 (3.0)	114.7 ± 1.5 (−9.3)	1.5 ± 0.1 (0.0)	6.0 ± 1.0 (−0.3)	13.7 ± 0.9 (0.0)	72.0 ± 6.0 (1.7)	83.0 ± 9.3 (−1.7)
Ondansetron	117.3 ± 5.2 (−1.0)	112.0 ± 5.3 (−6.7)	1.6 ± 0.2 (0.1)	4.7 ± 0.7 (0.0)	11.3 ± 0.7 (0.3)	72.0 ± 24.0 (6.0)	82.3 ± 3.5 (−2.0)
	QRS	QTC	SV	SWI	PVR	VR	
Vehicle	47.7 ± 1.9 (−0.7)	315.7 ± 12.6 (8.0)	14.0 ± 0.8 (1.1)	36.9 ± 1.3 (4.1)	363 ± 97 (−29.1)	6,072 ± 475 (147)	
Zatosetron	45.7 ± 2.4 (1.3)	365.7 ± 15.4 (60.2)*	12.7 ± 0.8 (1.1)	35.7 ± 2.1 (3.9)	440 ± 28 (14.7)	6,531 ± 652 (61)	
Ondansetron	49.0 ± 3.6 (0.0)	369.5 ± 21.6 (59.9)*	14.6 ± 1.6 (2.1)	43.0 ± 6.8 (6.1)	332 ± 45 (−2.8)	5,867 ± 544 (146)	

†Shown are the mean ± S.E. values (3 dogs/group) for parameters following intravenous infusion of vehicle (Milli-Q water), zatosetron (0.875 mg/kg) or ondansetron (2.63 mg/kg) over 15 min. Values in parentheses = actual changes from baseline values for the respective groups.
MP, mean pressure, mm Hg; HR, heart rate, beats/min; CO, cardiac output, liters/min; PCWP, pulmonary capillary wedge pressure, mm Hg; PULM, pulmonary pressure, mm Hg; FLOW, femoral flow, ml/min; PR, P-R interval, msec; QRS, QRS interval, msec; QTC, QTC interval, msec/sec½; SV, stroke volume, ml/beat; SWI, stroke work index, g-m/beat/m^2; PVR, pulmonary vascular resistance, dyne-sec-cm-^{5}; VR, peripheral vascular resistance, dyne-sec-cm-^{5}.
*< 0.05 relative to vehicle group by Dunnett two-tailed test.

In summary, these studies document that ondansetron, like other $5HT_3$ receptor antagonists, can prolong the Q-T_c interval in anesthetized dogs. This observation strengthens the possible association of this class of compounds with alteration in electrocardiographic param-

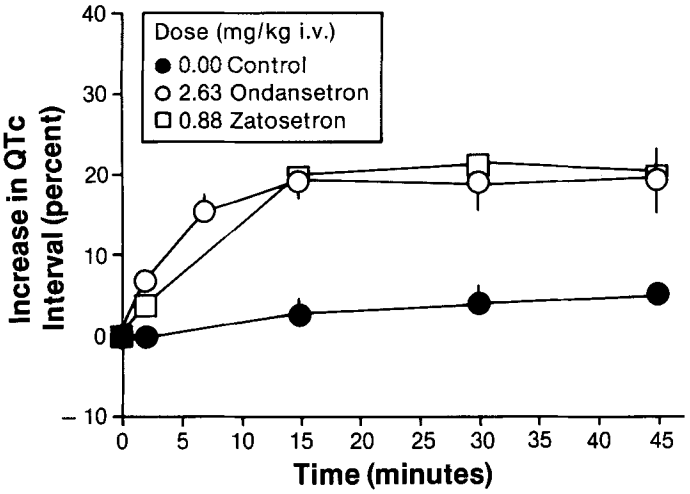


Fig. 3. Percent increase in QT_c interval at pharmacologically equivalent doses of zatosetron and ondansetron infused over 15 minutes in anesthetized beagle dogs. Shown are the mean ± S.E. of each group (n = 3).

eters, an effect that requires high doses and may be either directly or indirectly related to their ability to block $5HT_3$ receptors.

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