Duration of β-adrenoceptor blockade associated with once-daily oral administration of atenolol in healthy dogs

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OBJECTIVE
To test the hypothesis that once-daily oral administration of atenolol would attenuate the heart rate response to isoproterenol for 24 hours.

ANIMALS
20 healthy dogs.

PROCEDURES
A double-blind randomized placebo-controlled crossover study was conducted. Dogs were assigned to receive atenolol (1 mg/kg, PO, q 24 h) or a placebo for 5 to 7 days. After a washout period of 7 days, dogs then received the other treatment. Heart rate at rest (HR_r) and heart rate induced by administration of isoproterenol (HR_i) as a constant rate infusion (0.2 μg/kg/min for 5 to 7 minutes) were obtained by use of ECG 0, 0.25, 3, 6, 12, 18, and 24 hours after administration of the final dose of atenolol or the placebo. A mixed-model ANOVA was used to evaluate effects of treatment, time after drug or placebo administration, treatment-by-time interaction, period, and sequence on HR_r and HR_i.

RESULTS
Effects of sequence or period were not detected. There was a significant effect of treatment and the treatment-by-time interaction on HR_i. Atenolol significantly attenuated HR_i for 24 hours but did so maximally at 3 hours (least squares mean ± SE, 146 ± 5 beats/min and 208 ± 5 beats/min for atenolol and placebo, respectively). The effect at 24 hours was small (193 ± 5 beats/min and 206 ± 5 beats/min for atenolol and placebo, respectively). Atenolol had a small but significant effect on HR_r.

CONCLUSIONS AND CLINICAL RELEVANCE
This study of healthy dogs receiving atenolol supported a recommendation for a dosing interval < 24 hours. (*Am J Vet Res* 2019;80:270–274)

Atenolol is a selective antagonist of β_1-adrenergic receptors that limits myocardial oxygen consumption and might preserve myocardial function by decreasing heart rate and contractility.[1–3] In humans, atenolol does not have an appreciable effect on HR_r at doses used clinically, but it attenuates tachycardia that develops in response to stimulation of the sympathetic nervous system.[1] Atenolol has been used in the treatment of cardiac diseases in veterinary patients, including aortic stenosis, pulmonic stenosis, tachyarrhythmia, hypertrophic obstructive cardiomyopathy, systemic hypertension, hyperthyroidism, and systolic dysfunction.[4–9]

Doses of 0.2 to 1 mg/kg, PO, and dosing intervals of both 12 and 24 hours have been recommended for use in dogs.[10] The elimination half-life of atenolol in healthy dogs is 3 to 7 hours, and peak plasma concentration is achieved 1 to 2 hours after oral administration.[11] Atenolol is excreted unchanged in the urine.[12] However, pharmacokinetic variables do not accurately predict pharmacodynamic effects of β-adrenoceptor blockers, and serial evaluation of serum concentrations is not a recommended monitoring strategy.[13,14] Optimal dosing intervals are determined by the duration of β-adrenoceptor blockade after oral administration of atenolol. The magnitude of the increase in HR_i is a surrogate measure of β-adrenoceptor blockade that has been used in investigations of β-adrenergic receptor antagonists.[15–17]

Determination of an appropriate dosing interval is important because abrupt withdrawal of β-adrenoceptor blockade is associated with adverse effects, including the development of tachycardia, and has a negative impact on prognosis in patients with cardiac disease. More specifically, deleterious cardiovascular effects of abrupt β-adrenoceptor blockade withdrawal have been detected in humans with hypertrophic cardiomyopathy and systemic hypertension.[18–20] The pathogenesis of these effects is believed to involve hypersensitivity to catecholamines that reflects upregulation of adrenergic receptors associated with chronic β-adrenoceptor blockade.[21] Results for studies[22,23] of the use of β-adrenoceptor blockers to control heart rate and blood pressure in human patients emphasize the importance of consistent pharmacological activity rather than the use of a specific

ABBREVIATIONS
HR_i Heart rate induced by administration of isoproterenol
HR_r Heart rate at rest

dose. Although the pharmacokinetics of atenolol is known, pharmacodynamics of β-adrenoceptor blockade for dogs orally administered atenolol at commonly prescribed doses has been incompletely described.

The purpose of the study reported here was to evaluate the influence of once-daily oral administration of atenolol on isoproterenol-induced effects on heart rate in dogs. We hypothesized that once-daily oral administration of atenolol to healthy dogs would attenuate the HR_i for 24 hours. A secondary hypothesis of the study was that atenolol administration would not affect HR_r.

Materials and Methods

Animals

Twenty purpose-bred young adult mixed-breed dogs were included in the study. There were 10 males and 10 females; age ranged from 22 to 30 months, and median body weight was 10.85 kg (range, 8.8 to 12.4 kg). All dogs were considered healthy on the basis of results of a physical examination, CBC, plasma biochemical analysis, and urinalysis. The study was approved by the Institutional Animal Care and Use Committee of the Virginia-Maryland College of Veterinary Medicine.

Study design

A double-blind randomized placebo-controlled crossover study was conducted. Dogs were randomly assigned (by computer-simulated coin toss) to initially receive atenolol (1 mg/kg, PO, q 24 h) or a placebo (methylcellulose) for 5 to 7 days. After a washout period of 7 days, dogs then received the other treatment. Atenolol and the placebo were administered in identical gelatin capsules with a small volume of commercial dog food in the morning at approximately the same time each day.

All dogs were acclimated to ECG examination by practice sessions in which the subjects were gently restrained in lateral recumbency before collection of heart rate data. Heart rate data were acquired on the last day of oral administration of atenolol or the placebo (5 to 7 days after the first dose of atenolol or the placebo). On the day of data acquisition, a resting ECG was recorded (time 0). Then, a 20-gauge, 1.25-inch catheter was placed in a cephalic vein of each dog, and atenolol or the placebo was orally administered. For each subsequent ECG recording, data were collected for 30 seconds, isoproterenol was infused IV (constant rate infusion; 0.2 µg/kg/min for 5 to 7 minutes), and data were recorded for an additional 90 seconds during infusion of isoproterenol after ≥ 5 minutes had elapsed since the start of the infusion. Heart rate was assessed at time 0 and 0.25, 3, 6, 12, 18, and 24 hours.

ECG analysis

The ECG recordings were digitally stored for subsequent analysis.[a] Individual recordings were selected from the data set in a systematic manner. More specifically, the computer files were renamed, and the order in which they were analyzed was determined by use of a sequence from a random number generator. The investigator (MIW) who evaluated the ECGs was unaware of dog identity, time of recording, and treatment assignment. A software program[b] was used to evaluate the data; it detected successive peaks or nadirs of the ECG signal to determine the heart rate, but each interval was confirmed by visual inspection. The ECG variables evaluated consisted of HR_r and maximum HR_i. The HR_r was the mean heart rate per minute calculated for a 20- to 30-second interval. Maximum HR_i was the highest mean heart rate for any successive 6-second interval during isoproterenol infusion.

Statistical analysis

Data were analyzed by use of a linear mixed model that included the fixed effects of time, treatment, period, and sequence as well as the random effects of dog on the response variables (ie, HR_r and HR_i). The time-by-treatment interaction was further analyzed to compare treatments at each time point and to compare time points within each treatment. The *P* values for 2-way comparisons were adjusted for multiple comparisons by use of the Tukey procedure. All data were reported as least squares mean ± SE, except as noted. Values of $P \leq 0.05$ were considered significant. Evaluation of residuals confirmed that the assumptions of the analyses were met. Analyses of data were performed with commercially available computer software.[c]

Results

None of the dogs developed clinical signs of illness during the study. Isoproterenol administration was associated with a few premature ventricular contractions and supraventricular complexes, but sustained pathological tachyarrhythmia was not detected.

Neither sequence nor period caused significant (all $P \geq 0.331$) effects. There was a significant ($P < 0.001$) effect of treatment, time, and the treatment-by-time interaction on HR_i. Atenolol significantly attenuated HR_i at all time points (0.25, 3, 6, 12, 18, and 24 hours after atenolol administration). Magnitude of attenuation of heart rate was greatest at 3 hours. When dogs received atenolol, HR_i at 3, 6, and 12 hours differed significantly ($P < 0.001$) from HR_i at all other times, except that HR_i at 3 and 6 hours did not differ significantly ($P = 0.340$). The HR_i did not differ significantly with time when dogs received the placebo **(Table 1)**.

Time and treatment had significant ($P < 0.001$) effects on HR_r, but the treatment-by-time interaction did not have a significant ($P = 0.132$) effect. Atenolol treatment resulted in a lower HR_r, but it differed significantly ($P < 0.001$) at 3 and 6 hours, compared with HR_r for the placebo treatment **(Table 2)**.

Table 1—Least squares mean ± SE values for HR_i over time in 20 dogs receiving atenolol (1 mg/kg, PO, q 24 h) and a placebo (methylcellulose) for 5 to 7 days in a crossover experiment (7-day washout period between treatments).

Time (h)	Atenolol (beats/min)	Placebo (beats/min)	*P* value*
0.25	191 ± 4.7	207 ± 4.7	0.002
3	146 ± 5.2	208 ± 5.2	< 0.001
6	155 ± 4.8	208 ± 4.8	< 0.001
12	170 ± 4.3	204 ± 4.3	< 0.001
18	188 ± 4.4	205 ± 4.4	< 0.001
24	193 ± 4.6	206 ± 4.6	0.006

The HR_i was determined electrocardiographically during a constant rate infusion of isoproterenol (0.2 µg/kg/min for 5 to 7 minutes) for up to 24 hours after administration of the final dose of atenolol or the placebo.

*Values between treatments were considered to differ significantly at $P \leq 0.05$ (mixed-model ANOVA).

Table 2—Least squares mean ± SE values for HR_r over time in 20 dogs receiving atenolol and a placebo for 5 to 7 days in a crossover experiment.

Time (h)	Atenolol (beats/min)	Placebo (beats/min)	*P* value*
0	110 ± 3.7	115 ± 3.7	0.170
0.25	97 ± 4.0	105 ± 4.0	0.100
3	92 ± 3.8	111 ± 3.8	< 0.001
6	90 ± 4.2	108 ± 4.2	< 0.001
12	99 ± 3.4	105 ± 3.4	0.076
18	104 ± 4.2	108 ± 4.2	0.347
24	93 ± 4.1	100 ± 4.1	0.146

The HR_r was determined electrocardiographically immediately before (time 0) and for up to 24 hours after administration of the final dose of atenolol or the placebo.

See Table 1 for remainder of key.

Discussion

Relative to results for the placebo, repeated once-daily administration of atenolol significantly attenuated HR_i for 24 hours. The magnitude of heart rate attenuation differed significantly over the 24-hour period, with a peak effect at 3 hours. Although magnitude of the heart rate attenuation associated with beneficial effects of β-adrenoceptor blockade has not been determined, the effect of atenolol 24 hours after dosing was small. Relative to results for the placebo, atenolol had a small but significant effect on HR_r at 3 and 6 hours after oral administration.

Atenolol is a competitive antagonist of β-adrenergic receptors.[1] The antagonist effect is selective in that β_1-adrenergic receptors, which are expressed to the greatest extent in cardiac tissue, are affected to a greater degree than are β_2-adrenergic receptors.[1] Predictably, pharmacodynamic effects of β-adrenoceptor blockade include bradycardia, slowing of atrioventricular conduction, and negative inotropism, although these effects are indirect. In fact, β-adrenoceptor blockade prevents the positive chronotropic, dromotropic, and inotropic effects that would otherwise result from β-adrenergic receptor agonists.[1]

Atenolol has high bioavailability in dogs and is excreted largely unchanged in the urine.[11,12] The elimination half-life after oral administration in dogs is approximately 6 hours and is independent of dose.[11,12] Atenolol is used as an antihypertensive agent in humans, although anecdotal evidence suggests that it has limited efficacy in the management of hypertension in dogs. Atenolol has been used in the medical management of subvalvular aortic stenosis, pulmonic stenosis, and ventricular tachyarrhythmias associated with arrhythmogenic right ventricular cardiomyopathy in dogs. Most clinical studies involved a dosing interval of 12 hours, but once-daily dosing has also been described.[5,6,24] In 1 veterinary formulary,[10] 12- and 24-hour dosing intervals are indicated.

Although HR_r was determined in the study reported here, the effect of atenolol primarily was inferred from HR_i. The present study was designed as a placebo-controlled, blinded crossover experiment. Relative to parallel studies, crossover designs increase statistical power but are subject to period and sequence effects. The former relates to within-subject changes that occur over time but independent of an intervention. Sequence effects occur when the order of treatment has an effect on response variables; in pharmacodynamic studies, sequence effects generally reflect inadequate withdrawal times that result in carryover effects into the subsequent treatment period. Period and sequence were included in the statistical model, and significant effects for period and sequence were not detected.

The HR_i is a practical way to quantify β_1-adrenoceptor blockade. Alternate methods include administration of other sympathomimetic drugs and exercise testing. The response to isoproterenol was evaluated during a constant rate infusion to ensure a consistent heart rate response, and the infusion rate was selected on the basis of previously published data; more specifically, it has been determined that infusion of isoproterenol to healthy dogs at rates between 0.13 and 0.28 µg/kg/min causes heart rates to exceed 180 beats/min.[16] Other investigators have used considerably higher infusion rates to evaluate β-adrenoceptor blockade, but in those studies,[17,25] isoproterenol was administered only to dogs that had received β-adrenoceptor antagonists. In addition, we were mindful of the possible adverse effects of high doses of sympathomimetic drugs for dogs when they received the placebo. At all of the time points, mean HR_i of dogs when receiving the placebo exceeded 200 beats/min, whereas mean HR_r was close to 110 beats/min; these observations provided indirect evidence of substantive stimulation of β-adrenergic receptors.

Results of few published reports can be directly compared with results of the study reported here. Dogs with experimentally induced mitral valve regurgitation have been used to determine the beneficial effects of chronic oral administration of atenolol.[25] Although the body weights of the dogs were not provided for that study,[25] it is likely that the atenolol

dose of 50 mg/dog/d was higher than the one used in the present study because 100% attenuation of the increase in heart rate in response to an infusion of isoproterenol (4 µg/kg/min) was detected. On the basis of the reported left ventricular volumes at baseline, it is likely that the dogs included in that study[25] weighed approximately 27 kg and that the daily atenolol dose was close to 2 mg/kg, whereas the present study involved use of a dose of 1 mg/kg.

Relative to results for the placebo, once-daily administration of atenolol significantly decreased HR_r at only 3 and 6 hours after administration. Relative activities of the sympathetic and parasympathetic nervous systems modulate heart rate. Because of this, the effect of β-adrenoceptor blockade on HR_r is dependent on pharmacodynamic effects as well as on the prevailing autonomic tone. Vagal effects predominate in resting dogs; the intrinsic sinus rate (ie, sinus rate during complete autonomic blockade) is higher than the HR_r, and magnitude of the effect of parasympatholysis on heart rate exceeds that of sympatholysis.[26] Therefore, it was not surprising that only a small decrease in heart rate was detected after administration of atenolol. This finding is consistent with results of other studies[15,16] of β-adrenoceptor blockade. Data from a recent study[27] provide evidence that heterogeneity of the canine genome may explain the variable responses to stimulation of adrenergic receptors and therefore β-adrenoceptor blockade. Genetic screening was not performed on the dogs of the present study, but it is possible that this factor contributed to variation in the data.

The present study had some limitations. The sample consisted of healthy mixed-breed dogs; therefore, results may not be applicable to a genetically diverse population of older patients that might concurrently receive medications or have comorbidities that interfere with the absorption, action, or elimination of atenolol. Patients with some forms of cardiac disease have activation of adrenergic receptors that is unopposed by vagal restraint, whereas in contrast, parasympathetic activity dominates in healthy dogs.[22,28,29] Agonism of β-adrenergic receptors that results from an infusion of isoproterenol might not replicate the endogenous stimulation of adrenergic receptors seen in clinical cases. Furthermore, although attenuation of the effect of an exogenous receptor agonist is a practical method of quantifying β-adrenoceptor blockade, there is no direct correlation between this attenuation and clinically relevant effects (eg, a decrease in HR_r) that might be observed in patients with heart disease. Ambulatory ECG monitoring might have better reflected the mean HR_r, but it was not practical for the study reported here. Furthermore, we investigated effects only on heart rate and not on other cardiovascular functional variables. However, despite these limitations and on the basis of the pharmacokinetics of atenolol, it appeared unlikely that the duration of drug effects would differ markedly in patients with heart disease, except perhaps when there is concurrent renal dysfunction.

In the present study, oral administration of atenolol to healthy Beagles significantly attenuated heart rate response to β-adrenergic receptor stimulation for 24 hours, but the effect after 12 hours was relatively small. Oral administration of atenolol caused a time-dependent decrease in HR_r such that the HR_r at 3 and 6 hours after administration was significantly different, compared with the HR_r after placebo administration. These findings suggested that the dosing interval for atenolol should be < 24 hours.

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Footnotes

a. MP150 analogue to digital converter, BioPac Systems Inc, Goleta, Calif.
b. AcqKnowledge software, BioPac Systems Inc, Goleta, Calif.
c. SAS, version 9.2, SAS Institute Inc, Cary, NC.

References

1. Westfall TC, Westfall DP. Adrenergic agonists and antagonists. In: Brunton L, Lazo JS, Parker KL, eds. *Goodman and Gilman's pharmacologic basis of therapuetics*. 11th ed. New York: McGraw-Hill, 2006;237–296.
2. Bristow MR. Beta-adrenergic receptor blockade in chronic heart failure. *Circulation* 2000;101:558–569.
3. Cleland JG. Beta-blockers for heart failure: why, which, when, and where. *Med Clin North Am* 2003;87:339–371.
4. Eason BD, Fine DM, Leeder D, et al. Influence of beta blockers on survival in dogs with severe subaortic stenosis. *J Vet Intern Med* 2014;28:857–862.
5. Monnet E, Orton EC, Gaynor JS, et al. Open resection for subvalvular aortic stenosis in dogs. *J Am Vet Med Assoc* 1996;209:1255–1261.
6. Meurs KM, Spier AW, Wright NA, et al. Comparison of the effects of four antiarrhythmic treatments for familial ventricular arrhythmias in Boxers. *J Am Vet Med Assoc* 2002;221:522–527.
7. Schober KE, Zientek J, Li X, et al. Effect of treatment with atenolol on 5-year survival in cats with preclinical (asymptomatic) hypertrophic cardiomyopathy. *J Vet Cardiol* 2013;15:93–104.
8. Brown S, Atkins C, Bagley R, et al. Guidelines for the identification, evaluation, and management of systemic hypertension in dogs and cats. *J Vet Intern Med* 2007;21:542–558.
9. Abbott JA. Beta-blockade in the management of systolic dysfunction. *Vet Clin North Am Small Anim Pract* 2004;34:1157–1170.
10. Papich MG. *Saunders handbook of veterinary drugs: small and large animal*. 3rd ed. St Louis: Elsevier Saunders, 2011;54–56.
11. McAinsh J, Holmes BF. Pharmacokinetic studies with atenolol in the dog. *Biopharm Drug Dispos* 1983;4:249–261.
12. Reeves PR, Barnfield DJ, Longshaw S, et al. Disposition and metabolism of atenolol in animals. *Xenobiotica* 1978;8:305–311.
13. Jürgens G, Graudal NA, Kampmann JP. Therapeutic drug monitoring of antiarrhythmic drugs. *Clin Pharmacokinet* 2003;42:647–663.
14. Talbert RL. Pharmacokinetics and pharmacodynamics of beta blockers in heart failure. *Heart Fail Rev* 2004;9:131–137.
15. Abbott JA, Broadstone RV, Ward DL, et al. Hemodynamic effects of orally administered carvedilol in healthy conscious dogs. *Am J Vet Res* 2005;66:637–641.

16. Gordon SG, Arsenault WG, Longnecker M, et al. Pharmacodynamics of carvedilol in conscious, healthy dogs. *J Vet Intern Med* 2006;20:297–304.
17. Uechi M, Sasaki T, Ueno K, et al. Cardiovascular and renal effects of carvedilol in dogs with heart failure. *J Vet Med Sci* 2002;64:469–475.
18. Houston MC, Hodge R. Beta-adrenergic blocker withdrawal syndromes in hypertension and other cardiovascular diseases. *Am Heart J* 1988;116:515–523.
19. Swedberg K, Hjalmarson A, Waagstein F, et al. Adverse effects of beta-blockade withdrawal in patients with congestive cardiomyopathy. *Br Heart J* 1980;44:134–142.
20. Gilligan DM, Chan WL, Stewart R, et al. Adrenergic hypersensitivity after beta-blocker withdrawal in hypertrophic cardiomyopathy. *Am J Cardiol* 1991;68:766–772.
21. Ross PJ, Lewis MJ, Sheridan DJ, et al. Adrenergic hypersensitivity after beta-blocker withdrawal. *Br Heart J* 1981;45:637–642.
22. Flannery G, Gehrig-Mills R, Billah B, et al. Analysis of randomized controlled trials on the effect of magnitude of heart rate reduction on clinical outcomes in patients with systolic chronic heart failure receiving beta-blockers. *Am J Cardiol* 2008;101:865–869.
23. McAlister FA, Wiebe N, Ezekowitz JA, et al. Meta-analysis: beta-blocker dose, heart rate reduction, and death in patients with heart failure. *Ann Intern Med* 2009;150:784–794.
24. Meurs KM, Lehmkuhl LB, Bonagura JD. Survival times in dogs with severe subvalvular aortic stenosis treated with balloon valvuloplasty or atenolol. *J Am Vet Med Assoc* 2005;227:420–424.
25. Tsutsui H, Spinale FG, Nagatsu M, et al. Effects of chronic beta-adrenergic blockade on the left ventricular and cardiocyte abnormalities of chronic canine mitral regurgitation. *J Clin Invest* 1994;93:2639–2648.
26. Cavero I, Riggenbach H, Wall M, et al. Analysis of cardiac chronotropic responses to some autonomic blocking agents in conscious trained dogs. *Eur J Pharmacol* 1976;39:193–202.
27. Meurs KM, Stern JA, Reina-Doreste Y, et al. Impact of the canine double-deletion beta1 adrenoreceptor polymorphisms on protein structure and heart rate response to atenolol, a beta1-selective beta-blocker. *Pharmacogenet Genomics* 2015;25:427–431.
28. Ware WA, Lund DD, Subieta AR, et al. Sympathetic activation in dogs with congestive heart failure caused by chronic mitral valve disease and dilated cardiomyopathy. *J Am Vet Med Assoc* 1990;197:1475–1481.
29. Zucker IH, Wang W. Modulation of baroreflex and baroreceptor function in experimental heart failure. *Basic Res Cardiol* 1991;86(suppl 3):133–148.