Comparison of pharmacodynamic variables following oral versus transdermal administration of atenolol to healthy cats

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Objective—To describe the disposition of and pharmacodynamic response to atenolol when administered as a novel transdermal gel formulation to healthy cats.

Animals—7 healthy neutered male client-owned cats.

Procedures—Atenolol was administered either orally as a quarter of a 25-mg tablet or as an equal dose by transdermal gel. Following 1 week of treatment, an ECG and blood pressure measurements were performed and blood samples were collected for determination of plasma atenolol concentration at 2 and 12 hours after administration.

Results—2 hours after oral administration, 6 of 7 cats reached therapeutic plasma atenolol concentrations with a mean peak concentration of 579 ± 212 ng/mL. Two hours following transdermal administration, only 2 of 7 cats reached therapeutic plasma atenolol concentrations with a mean peak concentration of 177 ± 123 ng/mL. The difference in concentration between treatments was significant. Trough plasma atenolol concentrations of 258 ± 142 ng/mL and 62.4 ± 17 ng/mL were achieved 12 hours after oral and transdermal administration, respectively. A negative correlation was found between heart rate and plasma atenolol concentration.

Conclusions and Clinical Relevance—Oral administration of atenolol at a median dose of 1.1 mg/kg every 12 hours (range, 0.8 to 1.5 mg/kg) in cats induced effective plasma concentrations at 2 hours after treatment in most cats. Transdermal administration provided lower and inconsistent plasma atenolol concentrations. Further studies are needed to find an effective formulation and dosing scheme for transdermal administration of atenolol. (Am J Vet Res 2008;69:39–44)

β-Adrenergic receptor antagonists are commonly prescribed for cats with hypertrophic cardiomyopathy.1–5 Results of human studies6–9 have consistently revealed the benefit of β-adrenergic receptor antagonists in the treatment of cardiac patients, and β-adrenergic receptor antagonists are a cornerstone of treatment for hypertrophic cardiomyopathy in people. Benefits of β-adrenergic receptor antagonist administration in people with hypertrophic cardiomyopathy include a reduction in exertional outflow obstruction and myocardial oxygen demand and improved left ventricular diastolic filling via heart rate reduction.10,11 Atenolol, a relatively β1 (cardiac)-specific, β-adrenergic receptor antagonist, is frequently the β-adrenergic receptor antagonist chosen for treatment of cats with cardiac disease.12–14 Atenolol is not approved for use in animals. However, the human preparation generally is administered at a dose ranging from 6.25 to 12.5 mg/cat once or twice daily.13,14 Orally administered atenolol (as a tablet) has been determined to be sufficiently bioavailable in cats and is able to blunt an isoproterenol-induced heart rate challenge.13 Plasma atenolol concentrations associated with this pharmacodynamic response were 260 to 897 ng/mL.13

Daily oral administration of atenolol to cats is sufficiently challenging, which may result in a lack of compliance and impact therapeutic success. Results of 1 study of cats with hypertrophic cardiomyopathy revealed decreased owner compliance with regard to medication recommendations in 56 of 260 cats. In response to difficulty administering oral medications to cats, different formulations have been proposed in recent years.15–17 Transdermal delivery systems have been investigated because they are an effective method of drug transport in humans,16,17 and clinical experience suggests that transdermal preparations appear to be better tolerated by cats than tablets. Efficacious administration of nitroglycerin, lidocaine, and fen-
tanyl via a transdermal route has been accomplished in many species, although via different formulations.10-21

Results of a recent study22 revealed a positive therapeutic effect following the administration of methimazole as a transdermal gel form in cats, although a previous study23 did not result in consistent transdermal absorption of methimazole in a single dose trial. Additionally, atenolol is absorbed transdermally in mice and guinea pigs.3,24-26 Currently, transdermal administration of atenolol is being substituted for oral administration in clinical practice, typically in doses equal to those given orally, although, to our knowledge, no published study exists reporting plasma atenolol concentrations or a clinical effect of this approach in cats. Compoundings pharmacies use, almost exclusively, a pluronic lecithin organogel formulation for transdermal administration of medications to animals.

The study reported here was designed to evaluate plasma concentrations of atenolol and certain pharmacodynamic end points that might be affected by β-adrenergic blockade following oral and transdermal administration of atenolol in cats. A blunting of the increase in heart rate in response to an isoproterenol challenge has been used as a measure of adequate β-adrenergic antagonism in a previous study.13 However, administration of isoproterenol is not without risk and is best avoided in client-owned animals. Currently, atenolol use in cats is recommended without any defined criteria for adequate β-adrenergic receptor blockade in clinical patients. A decrease in heart rate is used as a goal, but no definitive heart rate goal has been established.13-15

In a previous study, a significant increase in heart rate and blood pressure was found when cats were brought into an examination setting, the so-called white coat effect, with heart rates increasing from 181 to 215 beats/min.26 Clinically applicable methods to evaluate the degree of β-adrenergic receptor blockade in cats with heart disease are desirable, and we hypothesized that the degree of alteration of heart rate and blood pressure following stressors (hospitalization and phlebotomy) before and after attempt β-adrenergic receptor blockade might serve as a suitable surrogate measurement for β-adrenergic receptor blockade. Therefore, the aim of the study reported here was to determine the effect of transdermal administration of atenolol on heart rate and blood pressure following hospitalization and phlebotomy in comparison to oral administration of atenolol and baseline measures. Additionally, potential correlations between plasma atenolol concentrations and blood pressure, heart rate, or change in heart rate were evaluated.

Materials and Methods

Animals—Healthy male castrated client-owned adult cats were studied. Inclusion criteria consisted of normal findings on physical examination, cardiac auscultation, ECG, echocardiography, CBC, and serum biochemical profile. Cats were excluded if they had abnormal SUN and creatinine concentrations, cardiovascular physical examination and echocardiographic evidence indicative of heart disease, abnormal ECG, or evidence of a concurrent illness or ongoing health problem. Informed consent was obtained from all owners, and the study was approved by Tufts Cummings School of Veterinary Medicine’s Institutional Animal Care and Use Committee.

Protocol—Cats were initially screened on the basis of history and physical examination findings. If no abnormalities were found, echocardiography was performed. Cats were studied further if the results of 2-dimensional, M-mode, and Doppler echocardiography were within accepted reference ranges.27-29 For cats with a normal echocardiogram, a simultaneous limb lead ECG was used to record the heart rate and rhythm during restraint for phlebotomy. Blood pressure was measured immediately after phlebotomy by use of an ultrasonic Doppler flow detector. Five blood pressure readings were taken and averaged.

Cats were assigned to receive atenololb (6.25 mg, q 12 h) either orally or transdermally for 1 week. For transdermal administration, owners were instructed to apply gel to the inner pinnae of the ears. On the seventh day of administration, owners were instructed to withhold food from cats for 12 hours but to continue giving the standard dose of atenolol. Cats were hospitalized as soon as possible after atenolol administration. Blood samples were taken at 2 and 12 hours after atenolol administration for assay of plasma atenolol concentrations. An ECG was recorded for ≥ 1 minute prior to phlebotomy and for ≥ 30 seconds after phlebotomy. The maximum heart rate for any 3-second period was determined from the baseline ECG (before phlebotomy) and during or after phlebotomy. Additionally, in an attempt to obtain the highest instantaneous heart rate, the shortest R-R interval during the baseline ECG recording and the shortest R-R interval during or after phlebotomy were recorded. After a minimum 1-week washout without drug,13 cats crossed over to the other formulation of atenolol and all procedures were repeated.

Transdermal gel formulation—The gel was made in 100-g lots and consisted of 1% atenolol in a clear, transparent vehicle. The gel contained propylene glycol and glycerin as pharmaceutical excipients and also contained 2.5% surfactant (Tween 20) to facilitate cutaneous penetration. The foundation was a 1% carbomer gel that was affected by neutralization with 0.45% triethanolamine. The gel was preserved with a 0.1% antimicrobial-preservative solution.4 The gel was formulated by taking the propylene glycol, glycerin, and preservative and dispersing it in water. The atenolol was dissolved in this mixture, and then the surfactant was added. In a separate beaker, the carbomer gel was dispersed in water. Each of these was then mixed together along with the triethanolamine to form the gel. The gel was stored at room temperature (22° to 25°C) after preparation.

The density of the gel was determined by use of a pyknometer and was found to have a mean value of 1.013 g/mL. The stability of the gel at room temperature was determined over a 2-month period. The stability was determined by use of high-performance liquid chromatography and was found to be in the range of 90.1% to 105% of the labeled amount for samples held at room temperature. The gel appeared clear and transparent with no apparent color change during this period.
Sample analysis—Briefly, samples were prepared by use of solid-phase extraction. Samples were conditioned with 1 mL of methanol followed by 1 mL of water to which 200 µL of sample containing an unknown concentration of atenolol was added. Samples were washed with 750 µL of water-methanol (95:5, vol/vol) and eluted with 1 mL of methanol. The eluant was nitrogen evaporated at room temperature and reconstituted with 200 µL of mobile phase. The injection volume of the eluant was 40 µL. The mobile phase consisted of 83% phosphate buffer (0.01M containing 0.1% triethylenediamine) and 17% acetonitrile (pH, 2.9). Separation was achieved by use of reverse-phase chromatography with a C18 column¹ maintained at 30°C, with a mobile phase flow rate of 1.2 mL/min. Atenolol was detected by use of fluorescence spectrophotometry, with excitation occurring at a wavelength of 275 nm and emission at 300 nm. Unknown atenolol concentrations were predicted on the basis of an equation that correlated signal to concentration. The correlation was based on signals derived from samples to which known concentrations of pure atenolol had been added. The upper and lower limits of quantification were 1,000 and 25 ng/mL, respectively. Controls, which spanned the range of quantification, were predicted within 15% of the known concentrations.

Data analysis—Plasma \( t_{1/2} \) was calculated for each cat on the basis of the relationship between the \( k_{el} \) and \( t_{1/2} \) as follows:

\[
t_{1/2} = \frac{0.693}{k_{el}}
\]

where \( k_{el} \) is determined from the slope of the line defined by the 2 time points (peak and trough) as follows:

\[
k_{el} = \ln(C_1/C_2)/t_2 - t_1,
\]

where \( C \) is the concentration and \( t \) is the time at peak \( (C_1, t_1) \) and trough \( (C_2, t_2) \), respectively.

Data were examined graphically. Most variables were not normally distributed, so data are presented as median (range) values and nonparametric tests were used throughout. A nonparametric paired \( t \) test (Wilcoxon signed rank test) was used to compare baseline variables with the same variables at various times and for treatment conditions. Comparison of effect between oral and transdermal administration of atenolol at various time points was accomplished by use of a Mann-Whitney \( U \) test. Correlation between heart rate and plasma atenolol concentrations was calculated by use of a Spearman correlation coefficient. Values of \( P < 0.05 \) were considered significant. All analyses were performed by use of commercially available statistical software.

Results

Twenty-two cats were screened as presumed clinically normal for the study; however, 15 cats were excluded from the study on the basis of unsuitable behavior (\( n = 3 \)), cardiac arrhythmia (2), cardiac murmur grade > 3/6 (1), or echocardiographic abnormalities (left ventricular hypertrophy; 9). Seven cats with a mean ± SD weight of 5.8 ± 0.1 kg and an age of 6.6 ± 2.3 years were enrolled.

During initial examination, median maximum baseline heart rate from the initial ECG was 180 beats/min (range, 140 to 220 beats/min); median R-R interval was 0.32 seconds (range, 0.25 to 0.4 seconds). Median maximal baseline heart rate after phlebotomy was 190 beats/min (range, 150 to 220 beats/min); median R-R interval was 0.30 seconds (range, 0.28 to 0.38 seconds). Baseline values for heart rate and R-R interval were not significantly (\( P = 0.50 \) and 0.44, respectively) different for assessments before and after phlebotomy values (Table 1). Median baseline blood pressure after phlebotomy was 148 mm Hg (range, 140 to 190 mm Hg).

Atenolol was administered to cats at a median dose of 1.1 mg/kg (range, 0.8 to 1.5 mg/kg) every 12

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**Table 1**—Median (range) cardiovascular variables in cats (\( n = 7 \)) during initial (baseline) prescreening examination and after 7 days of oral or transdermal administration of atenolol for 7 days.

<table>
<thead>
<tr>
<th>Route</th>
<th>Variable</th>
<th>Before phlebotomy</th>
<th>After phlebotomy</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline</td>
<td>2 hours</td>
<td>12 hours</td>
</tr>
<tr>
<td>Oral</td>
<td>Max HR (beats/min)</td>
<td>180 (140–220)</td>
<td>150 (120–155)*</td>
</tr>
<tr>
<td></td>
<td>R-R interval (s)</td>
<td>0.32 (0.25–0.40)</td>
<td>0.38 (0.36–0.46)*</td>
</tr>
<tr>
<td></td>
<td>SAP (mm Hg)</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Transdermal</td>
<td>Max HR (beats/min)</td>
<td>180 (140–220)</td>
<td>160 (135–190)</td>
</tr>
<tr>
<td></td>
<td>R-R interval (s)</td>
<td>0.32 (0.25–0.40)</td>
<td>0.36 (0.32–0.42)</td>
</tr>
<tr>
<td></td>
<td>SAP (mm Hg)</td>
<td>ND</td>
<td>ND</td>
</tr>
</tbody>
</table>

*Significantly (\( P < 0.05 \)) different from baseline. Max = Maximum. HR = Heart rate. SAP = Systolic arterial blood pressure. ND = Not determined.
hours. Median plasma atenolol concentration 2 hours after oral administration was significantly (P = 0.004) higher (569 ng/mL [range, 242 to 834 ng/mL]) compared with 2 hours after transdermal administration (135 ng/mL [23 to 380 ng/mL]). Median plasma atenolol concentration at 12 hours after oral administration was also significantly (P = 0.004) higher compared with 12 hours after transdermal administration (175 ng/mL [118 to 465 ng/mL] vs 63 ng/mL [44 to 88 ng/mL], respectively). Median half-life for atenolol following oral administration was 11.4 hours (range, 3.6 to 21.9 hours), and median half-life for atenolol following transdermal administration was 5.7 hours (range, 3.9 to 13.3 hours; P = 0.29). Two of 7 cats reached a plasma atenolol concentration that has been proposed as a target for effective β-adrenergic receptor blockade (260 ng/mL) at 2 hours following transdermal administration, but plasma atenolol concentrations were below this therapeutic end point by 12 hours following transdermal administration in all cats. Six of 7 cats reached a plasma atenolol concentration of ≥ 260 ng/mL at 2 hours following oral administration. Three of 7 cats remained above this threshold plasma concentration at 12 hours after oral administration. No correlation was found between body weight and plasma atenolol concentrations.

Compared with baseline, heart rate at 2 hours after oral administration of atenolol was significantly (P = 0.02) lower and R-R intervals were significantly (P = 0.019) longer before and after phlebotomy (Table 1). The 2-hour R-R interval after phlebotomy was significantly longer than baseline values following oral (P = 0.02) or transdermal (P = 0.04) administration of atenolol. Heart rate after phlebotomy was also significantly lower than baseline at the 2 hours following oral (P < 0.001) or transdermal (P = 0.02) administration of atenolol. Heart rate after phlebotomy was significantly (P = 0.04) lower than at the initial visit at the 12-hour sample collection time for cats that received atenolol orally. No other significant differences were found in cardiovascular variables between sample collection times and baseline values.

When data from both methods of atenolol administration were combined, correlations were found between some measures of heart rate and the plasma atenolol concentration. At 2 hours, no measures of heart rate under normal (before phlebotomy) or stress (during or after) conditions were correlated with plasma atenolol concentrations. At 12 hours during or after phlebotomy, heart rate was negatively correlated (r = 0.72; P = 0.009) and R-R interval was positively correlated (r = 0.68; P = 0.02) with plasma atenolol concentrations. The R-R interval was also positively (r = 0.65; P = 0.02) correlated with plasma atenolol concentrations at 12 hours before phlebotomy. No correlation was found between blood pressure and plasma atenolol concentrations. When evaluating across all treatment groups and times, a correlation was found between plasma atenolol concentrations and R-R interval and heart rate (r = 0.61; P = 0.001 and r = 0.55; P = 0.003, respectively). All cats that had achieved therapeutic plasma atenolol concentrations had a heart rate ≤ 170 beats/min. However, in the 18 instances in which cats had plasma atenolol concentrations < 260 ng/mL, 10 had heart rates ≤ 170 beats/min and 8 had heart rates > 170 beats/min. The change in heart rate between baseline and treatment values was not correlated with plasma atenolol concentrations (r = 0.36; P = 0.07), but the change in R-R interval from baseline to treatment was correlated with plasma atenolol concentrations (r = 0.42; P = 0.03).

Discussion

Results of this study revealed that oral administration of atenolol achieved more consistent, longer-lasting, and higher plasma atenolol concentrations than the transdermal formulation of atenolol used in this study. Transdermal application of atenolol produced inconsistent and lower plasma atenolol concentrations than oral administration. This is consistent with the findings of at least 1 other study of transdermal medication. In addition, pharmacodynamic correlates of the efficacy of β-adrenergic receptor blockade confirmed that oral administration of atenolol produced heart rates that were consistently lower than baseline heart rates, while transdermal atenolol did not. Lower plasma atenolol concentrations and inconsistent absorption occurred despite what had been hypothesized to be an adequate length of administration prior to testing drug concentrations.

In theory, the transdermal route of administering medications has many potential advantages. It is noninvasive and not demanding technically, avoids first-pass hepatic metabolism and gastrointestinal breakdown, has potential for sustained release formulations, and can be administered over a large surface area. Transdermal administration of medication has been shown to achieve blood concentrations of drug that are considered to be therapeutic (eg, fentanyl) or efficaciously affect physiologic surrogates (eg, methimazole, nitroglycerine, and lidocaine). Feasibility of transdermal medication varies on a drug-by-drug basis. Characteristics for an ideal passive transdermal drug have been proposed. A suitable drug is neither too lipophilic nor too hydrophilic, and it should not be too large (molecular weight < 500 d). The ideal drug should also have a melting point of < 200°C, the pH of saturated aqueous solution should be in a relatively physiologic range (pH, 5 to 9), and the dose delivered should not be > 15 mg/d.

Based on the aforementioned criteria, atenolol has some of the characteristics of a suitable candidate for transdermal administration. Atenolol has a molecular weight of 266 d and has a daily dose in cats that is generally < 20 mg/d. However, it is a moderately hydrophilic drug, which means that it may not cross the stratum corneum easily, making it a less suitable drug for transdermal medication. Because some concern existed about the suitability of atenolol as a transdermal medication, the gel contained propylene glycol and glycerin as pharmaceutical excipients and also contained 2.5% surfactant (Tween 20) as an enhancer of cutaneous penetration. This combination has not been used previously in veterinary medicine, but is used frequently in human medication. Pluronic lecithin organogel is the most commonly used vehicle for veterinary transdermal medication. In this formulation, lecithin isopropyl


References

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palmitate is purported to act as an emulsifying agent by increasing the fluidity of stratum corneum, while pluronic F127 is purported to enhance micelle formation in gel and to modulate drug release and permeation of the stratum corneum. Because pluronic lecithin organogel is commonly used in veterinary medicine, this transdermal formulation, if proved effective or superior to the one studied here, would have more practical applicability.

A 1-week period of transdermal medication administration was chosen to ensure adequate aural stratum corneum softening and to achieve adequate drug concentrations. Results of previous research revealing that transdermal methimazole is efficacious involved testing a physiologic surrogate for methimazole after 3 to 4 weeks. Perhaps longer administration of transdermal atenolol would have led to higher plasma concentrations. The reason for a longer length of administration leading to better drug concentrations is that the vehicle disrupts the stratum corneum over time; thus, higher drug concentrations eventually will be achieved. The fact that atenolol actually had a lower plasma \( t_{1/2} \) after transdermal application is unexpected, as many transdermal formulations either accomplish a drug reservoir effect after repeated application or are designed for sustained release specifically.

Two of 7 cats with transdermal atenolol administration in the current study reached a threshold plasma atenolol concentration (260 ng/mL) as determined in previous research as the plasma target for effective \( \beta \)-adrenergic receptor blockade at 2 hours. This is in contrast to oral administration where 6 of 7 cats reached threshold concentrations at 2 hours. Only 1 of 7 cats receiving atenolol orally was above this threshold plasma atenolol concentration at 12 hours. A previous study in which oral administration of atenolol at a higher dose than the current study (3 mg/kg) resulted in adequate \( \beta \)-adrenergic receptor blockade at 12 hours in all cats.

The failure of most cats that received atenolol transdermally to reach therapeutic plasma atenolol concentrations raises questions about direct substitution of transdermal medications for oral medications at equivalent doses. This is a particularly important concern with atenolol because, unlike transdermal administration of methimazole, there are no easily quantifiable physical examination findings (weight gain), historical findings (decrease in appetite or activity level), or easily available laboratory tests (total thyroxine) for atenolol. Often, heart rate reduction is proposed as a surrogate end point to determine the efficacy of atenolol treatment. However, to date, no firm recommendations have been made on whether heart rate is indicative of adequate \( \beta \)-adrenergic receptor blockade. In our study, the absolute heart rate, rather than the change in heart rate, had a better correlation with plasma atenolol concentrations.

Neutered male cats were chosen as subjects in the current study because they comprised 193 of 260 (74%) cats in a recent study of cats with hypertrophic cardiomyopathy and 56 of 74 (76%) in a prior study. While there exists a possibility that female and male cats may have differences in drug absorption, this has yet to be proven.

Two cats did reach therapeutic plasma atenolol concentrations with the transdermal formulation. This suggests that the transdermal route may be an efficacious method of administering atenolol to some cats. Potentially, increasing either the amount of gel applied or the concentration of the gel might increase the number of cats that reach therapeutic plasma atenolol concentrations with this transdermal formulation. However, because not all cats reach therapeutic plasma atenolol concentrations, a dilemma exists about which cats are being effectively treated. Plasma atenolol concentrations or refinement of the above \( \beta \)-adrenergic receptor blockade assessment might provide the needed information for cats that are treated with transdermal atenolol.

Another possibility for so few cats reaching therapeutic plasma atenolol concentrations is that the cut-off for therapeutic plasma atenolol concentrations may have been too high. The value of 260 ng/mL was chosen from a previous study. All cats in that study had adequate \( \beta \)-adrenergic receptor blockade in response to an isoproterenol challenge, and the lowest plasma atenolol concentration was 260 ng/mL. However, 12 hours later, none of the cats had adequate \( \beta \)-adrenergic receptor blockade and the highest plasma atenolol concentration was 42 ng/mL. Therefore, the therapeutic plasma atenolol concentration capable of producing \( \beta \)-adrenergic receptor blockade lies between 260 and 42 ng/mL, and potentially more cats would have reached therapeutic concentrations if a lower concentration had been chosen.

In summary, oral administration of atenolol at a median dose of 1.1 mg/kg every 12 hours (range, 0.8 to 1.5 mg/kg) in cats achieved plasma atenolol concentrations > 260 ng/mL at 2 hours after treatment in most cats. Transdermal administration provided lower and inconsistent plasma atenolol concentration. Plasma atenolol concentration correlated with heart rate but not blood pressure. Further studies are needed to find a transdermal formulation and dosing regimen that will consistently result in plasma atenolol concentrations > 260 ng/mL.