

# Effects of theophylline on tracheal mucociliary clearance rates in healthy cats

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**Objective**—To determine tracheal mucociliary clearance rate (TMCCR) by use of a standard protocol in healthy anesthetized cats and to determine the effect of theophylline on TMCCR in healthy anesthetized cats.

**Animals**—6 healthy cats.

**Procedure**—Cats were anesthetized with propofol, and a droplet of the radiopharmaceutical technetium Tc 99m macroaggregated albumin was placed endoscopically at the carina. Dynamic acquisition scintigraphic imaging was performed, using the larynx as the end point. The TMCCR was determined by measuring the distance the droplet traveled by frame rate. Each cat was imaged 6 times as follows: 3 times following placebo administration and 3 times following the administration of sustained release theophylline (25 mg/kg, PO). Serum theophylline concentrations were assessed during imaging to ensure therapeutic concentrations.

**Results**—The TMCCR in healthy adult cats anesthetized with propofol was  $22.2 \pm 2.8$  mm/min. Tracheal mucociliary clearance rate in cats receiving theophylline was  $21.8 \pm 3.5$  mm/min. Theophylline administration did not significantly alter TMCCR.

**Conclusions and Clinical Relevance**—Theophylline has been shown to increase TMCCR in humans and dogs. In our study, we determined TMCCR in healthy anesthetized cats and found that it was not accelerated by the administration of theophylline. (*Am J Vet Res* 2002;63:1320–1322)

A thin layer of mucus coats the surface of all airways from the bronchioles to the larynx. This mucus is secreted by goblet cells and submucosal glands located within the respiratory epithelium, and it protects the

airways by absorbing inhaled particles, humidifying inspired air, maintaining mucosal hydration, and playing a role in immunologic defense. Cilia, on the apical surface of respiratory epithelial cells lining the airways from the larynx to the bronchioles, beat in an organized and synchronous fashion, propelling mucus out of smaller airways to the trachea and then up to the larynx to be swallowed or expectorated.<sup>1,2,3</sup> Mucociliary clearance is essential for the removal of airway secretions, pathogens, debris, and particulate matter from the respiratory tract.<sup>4,5</sup> Determination of tracheal mucociliary clearance rate (TMCCR) provides information regarding the functional integrity of the mucociliary clearance mechanism.

The rate of mucociliary clearance has been investigated in a number of species, and reference values have been established in humans,<sup>6,7,8,9</sup> horses,<sup>2,3,4,10</sup> calves,<sup>11,12</sup> dogs,<sup>5,6,13-15</sup> rats,<sup>6,16</sup> ferrets, and rabbits.<sup>6</sup> To our knowledge, no published reports have determined TMCCR in cats.

Effective mucociliary clearance is dependent on an efficient interaction between respiratory epithelium and mucus. Cilia must beat in a coordinated, effective manner and mucus must possess physical properties that optimize its clearance.<sup>5,6</sup> The TMCCR can be altered by environmental factors, exercise, disease, and pharmacologic agents. Theophylline, a methylxanthine, has been shown to increase TMCCR in humans<sup>17</sup> and dogs.<sup>16</sup> Its effect on TMCCR in cats has not been evaluated. The objectives of this prospective, randomized, blind study were to establish reference values for TMCCR in healthy anesthetized cats and to evaluate the effect of orally administered theophylline on TMCCR in healthy anesthetized cats.

## Materials and Methods

**Animals**—Six healthy, purpose-bred, vaccinated domestic shorthair cats, ranging in age from 1 to 3 years, were obtained for our study. All cats were housed in a facility approved by the Canadian Council on Animal Care. No evidence of systemic or respiratory disease was found in any cats, and abnormalities were not detected on physical examination, CBC determination, serum biochemical analysis, urinalysis, ELISA testing for feline leukemia antigen and feline immunodeficiency virus antibody, thoracic radiography, bronchoscopic evaluation of the airways, and a tracheal wash for cytology and aerobic bacterial culture.

**Study design**—The TMCCR was evaluated 6 times in each cat with a minimum of 7 days between measurements. On 3 occasions, cats were randomly assigned to receive placebos, consisting of an empty gelatin capsule, 8 hours prior to imaging. On 3 occasions, cats were assigned to receive sustained-release theophylline<sup>a</sup> (25 mg/kg) 8 hours prior to imaging. All cats were fed a commercial dry food ad libitum, but food was withheld for 12 hours before each scan.

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**Scintigraphic evaluation**—An IV cephalic catheter was placed in each cat, and a bolus of propofol<sup>b</sup> (8 mg/kg) was administered slowly. A light plane of anesthesia lasting 15 to 20 minutes was obtained. A 3.5-F nasal oxygen catheter was inserted into a nostril to provide humidified oxygen at a rate of 200 ml/min throughout the procedure. The anesthetized cats were placed in sternal recumbency, and a bronchoscope<sup>c</sup> was inserted through the larynx into the trachea and positioned at the carina. The scope was inserted without contacting the larynx or the wall of the trachea, as this could induce a cough reflex and possible bronchospasm. A 10-ml droplet of technetium Tc 99m macroaggregated albumin (3.3 mCi/mL) was deposited at the carina through a 0.58-mm-diameter polyethylene tubing<sup>d</sup> inserted down the 1.2-mm-diameter port of the scope. The images were acquired by use of a gamma camera<sup>e</sup> on a 128 × 128 matrix without zoom. A small field view with an all-purpose collimator was positioned to image the cervical and cranial thoracic regions. Dynamic acquisition at 20 s/frame began immediately following droplet deposition and continued until it reached the level of the larynx. The TMCCR was measured by computer analysis<sup>f</sup> by a nuclear medicine physician blinded to which treatment had been administered (theophylline or placebo). The rate was calculated by multiplying the number of pixels between the origin and the location of the stops of the droplet. After acquisition, all 20-second images were superimposed to view the path of the radioactive droplet. The distance traveled divided by the time of acquisition yielded the rate of migration in millimeters per minute. Once recovered from anesthesia, the cats were housed in a safety-approved radiation facility for 24 hours.

Each scan lasted 8 to 12 minutes. During imaging, each cat had a blood sample taken from the lateral saphenous vein to measure serum theophylline concentrations. Samples were analyzed by a fluorescence polarization immunoassay.<sup>g</sup>

**Statistical analysis**—Calculation of the means and SD was performed for the placebo and theophylline-treated groups. A paired *t* test was used to assess significant differences following treatment with placebo and theophylline. The differences between the means for TMCCR in both groups were determined to be normally distributed on the basis of a comparison of means, medians, and a Wilk-Shapiro-Rankit plot, therefore permitting the use of a paired *t* test. A value of *P* < 0.05 was considered significant.

## Results

The mean TMCCR determined for the placebo treated cats was 22.2 ± 2.8 mm/min. The mean TMCCR determined for the theophylline treated cats was 21.8 ± 3.5 mm/min. No significant difference was found between the 2 groups (*P* = 0.817). Serum theophylline concentrations measured during scintigraphy ranged from 11 to 19 µg/ml. All cats tolerated the procedure well and did not have signs of discomfort afterwards.

## Discussion

Reference values for TMCCR in cats anesthetized according to this protocol were 22.2 ± 2.8 mm/min. These rates are considerably higher than those reported in humans, dogs, and horses in which TMCCR have been estimated at 15.5 mm/min, 12.7 mm/min,<sup>6</sup> and 19.3 mm/min,<sup>10</sup> respectively. The TMCCR has been shown to be positively correlated to tracheal surface area,<sup>6</sup> which would have predicted a slower TMCCR in cats. Mucociliary clearance is a result of complex interactions between mucus

rheology, density and beat frequency of cilia, and airway size.<sup>3,4,6,7,18</sup> The TMCCR cannot, therefore, be predicted in a species without direct measurement. A number of environmental factors (temperature, humidity, head position) have been documented to affect TMCCR.<sup>4,11</sup> It is equally known that viral infections, through their destruction of cilia and stimulation of mucus production, slow TMCCR, which facilitates bacterial colonization of the airways.<sup>3</sup>

Results of studies<sup>5,10,15</sup> of TMCCR in humans and horses have revealed that significant variations normally exist among individuals of the same species. The relatively small SD identified in our study indicates that there was little variation in TMCCR between the individual cats. This may be attributable in part to the homogeneity of our population and the use of a consistent anesthetic protocol and imaging technique.

Theophylline has been used for more than 50 years in the management of human asthma and is also used in people suffering from chronic bronchitis and cystic fibrosis.<sup>17,19,20</sup> It has long been considered an effective bronchodilator, but more recently, its anti-inflammatory properties have received a considerable amount of attention.<sup>17,19,20,21</sup> Theophylline has been used in veterinary medicine to treat animals presumed to have airway disease and impaired mucociliary clearance, such as horses with chronic obstructive pulmonary disease,<sup>22</sup> dogs with chronic bronchitis,<sup>22</sup> and cats with asthma.<sup>23,24</sup> Theophylline has been reported to increase TMMCR in healthy humans and dogs<sup>16,17</sup> through a number of proposed mechanisms. It may have a direct stimulatory effect on ciliary beat amplitude and frequency and improve mucokinesis.<sup>3,16</sup> Our results revealed no significant differences in TMCCR in cats administered theophylline and those administered placebo. It is possible that no effect was seen because these healthy cats had rapid, near maximal TMCCR at the onset. It is unlikely that our anesthetic protocol masked differences between the treatment and placebo groups, because the same protocol was consistently used and would have been expected to affect TMCC equally in both groups. The effects of propofol on TMCCR have not been reported.

The therapeutic range of theophylline has been well established in humans and determined to be between 10 and 20 mg/ml. The serum concentration required to cause bronchodilation in cats has not been determined but is assumed to be similar to that in humans.<sup>25,26</sup> Measurements in our study confirmed that all cats had serum theophylline concentrations within the therapeutic range at the time of their scan. Sustained-release theophylline was administered because its pharmacokinetics is predictable and have been well established in cats.<sup>26</sup> A dose of 25 mg/kg was chosen because it has been shown to provide therapeutic serum concentrations during a 24-hour period.<sup>26</sup>

This paper describes a clinically useful and well-tolerated procedure to determine TMCCR in cats. Our study determined TMCCR in healthy anesthetized cats and found that it was not influenced by the administration of theophylline. Further studies are needed to evaluate the effects of disease, such as feline asthma, on TMCCR and to evaluate pharmacologic manipulations of TMCCR in disease states and in health.

<sup>a</sup>Slo-Bid, Rhone-Poulenc Rorer Canada Inc, QC, Canada.

<sup>b</sup>Rapinivet, Malinckrodt Veterinary Inc, Ajax, ON, Canada.

<sup>c</sup>Olympus LF-1, Carsen Group Inc, Markham, ON, Canada.

<sup>d</sup>Becton, Dickinson & Co, Sparks, Md.

<sup>e</sup>Siemens LEM, Hoffman Estates, Ill.

<sup>f</sup>Pagasys, ADAC Laboratories, Milpitas, Calif.

<sup>g</sup>Abbott AxSYM System, Abbott Laboratories, Abbott Park, Ill.

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