2019

Evaluation of antimicrobial efficacy in shelter cats with upper respiratory infection: a prospective study

Jillian R. Broadhurst
University of New Hampshire, Durham

Sarah E. Proctor DVM
University of New Hampshire, Durham

Evaluation of antimicrobial efficacy in shelter cats with upper respiratory infection: a prospective study

Authors:

Jillian Broadhurst

Peter S. Erickson, PhD

Sarah E. Proctor, MPH, DVM

Affiliation for all authors:

Department of Agriculture, Nutrition, and Food Systems, College of Life Sciences and Agriculture, University of New Hampshire, Durham, NH 03824.

Corresponding author: Sarah E. Proctor, sarah.proctor@unh.edu

Abstract

Objective: To evaluate the efficacy of antimicrobial treatment in feline upper respiratory disease (URD) through comparison of illness severity and duration in shelter cats treated with and without antimicrobials.

Design: Randomized prospective clinical trial.

Sample: 38 cats with URD.

Procedures: Cats with symptoms of URD were randomly assigned to 2 groups based on an alternating group assignment model. The treatment group received antimicrobial treatment and the control group did not. The severity of each cat's oculonasal symptoms was scored daily on a numerical scale (0 = no symptoms to 4 = severe symptoms). Duration of clinical symptoms was recorded. Aerobic bacterial culture and PCR testing was performed to identify possible bacterial causes of URD in 14 of the 38 cats, 7 from each group.

Results: Upper respiratory disease duration did not differ between groups (treatment group = 6.99 days, control group = 6.28 days, P=0.61). URD severity score did not differ between groups (treatment group = 3.93, control group = 6.69, P=0.26). 4 of 14 cats who underwent diagnostic testing had positive PCR tests. Positive PCR results included *Mycoplasma, Bordetella,* and *Chlamydia spp.* or a combination of these. Aerobic bacterial culture results showed positive growth across all tested cats. All organisms detected were considered normal oral flora.

Conclusions and Clinical Relevance: No significant difference in the severity or duration of URD was found in cats who did and did not receive antimicrobial treatment. These findings suggest that antimicrobial treatment did not improve outcomes of URD at this shelter.

Abbreviations

C: amoxicillin/clavulanic acid

D: doxycycline

FHV: Feline Herpesvirus

FS: female spayed

FVRCP: feline viral rhinotracheitis, calicivirus, panleukopenia

ISCAID: International Society for Companion Animal Infectious Diseases

MN: male neutered

OS: owner surrender

S: stray

SU: sucralfate

T: tobramycin

URD: upper respiratory tract disease

Upper respiratory tract disease (URD) is one of the most predominant causes of feline morbidity and mortality in North American animal shelters and a frequent reason for euthanasia.[1,2] Feline herpesvirus (FHV) and calicivirus account for approximately 80% of all cases of URD, but secondary bacterial infections are common, especially in shelters where stress and disease pressures are high.[1,3,4] Bacteria implicated as primary causes of URD include *Chlamydophila felis*, *Bordetella bronchiseptica*, and *Mycoplasma felis.*[1,5-8] Diagnosis and treatment of URD is based primarily on clinical symptoms, and while testing for pathogens is available, it is not often performed. Causative agents of URD are often detected in cats with and without symptoms, so correlation between a positive test and the true cause of symptoms in any individual is often unclear.[4-9] Many cats with URD symptoms are empirically treated with antimicrobials, particularly in shelter environments.[1,4,10]

With growing recognition of antimicrobial overuse and its relationship to antimicrobial resistance, it is important to consider both the benefits and the costs of antimicrobial therapy for individual patients. Antimicrobials can alter the microbiome, induce gut bacterial overgrowth, and may alter the immune response to viruses.[10] Furthermore, the act of medicating cats may compound their stress and may thus prolong viral URD.[11] The International Society for Companion Animal Infectious Diseases (ISCAID) guidelines suggest that mucopurulent oculonasal discharge is not sufficient to warrant antimicrobial therapy in an otherwise healthy, non-febrile cat.[12] Other sources also recommend reserving antimicrobial treatment for cats exhibiting clear evidence of secondary bacterial infection with systemic involvement, and suggest that antimicrobials should not be used for every cat with symptoms of URD.[1] Human medicine uses similar guidelines for the judicious use of antimicrobials in upper respiratory tract infections.[13]

However, antimicrobial use in clinical practice and recommendations in other published guidelines are often less judicious than ISCAID guidelines.[4,14,15] In a shelter setting where there is often less veterinary oversight, there is even greater chance for inappropriate antimicrobial use. A survey of US shelters found that 35% of shelters use non-medical staff to make decisions about URD treatment.[16] The decision to initiate antimicrobials was based on symptoms like sneezing in 46% of shelters, rather than the presence of mucopurulent oculonasal discharge.[16]

We do not know of any studies directly comparing clinical outcomes of cats with URD being treated with and without antimicrobials. Litster et. al. compared various antimicrobials in treating URD but did not have an untreated control group.[17] Zirofsky et. al. found that PCR results for *Mycoplasma spp.* and FHV had poor predictive values for URD symptoms and for URD outcomes in cats, but they did not compare the outcomes between groups treated with antimicrobial versus antiviral medication.[9]

The aim of this study was to compare the duration and severity of URD in cats treated with and without antimicrobials in a shelter environment. We hypothesized that the two groups would not differ in their outcomes. We also tested cats for common URD pathogens to determine if there was an underlying primary bacterial cause for URD in this shelter that would suggest a need for antimicrobial therapy.

Materials and Methods

This non-matched case-control study evaluated cats at an animal shelter in New Hampshire from June 2018 through August 2019. The study protocol was approved by the University of New Hampshire Institutional Animal Care and Use Committee.

The shelter's feline intake procedure is as follows. All cats received an intranasal modified-live FHV and calicivirus vaccine[a] the day they entered the shelter. All cats without a documented current FVRCP vaccine were also vaccinated with a subcutaneous modified-live FVRCP vaccine[b] within a few days of entry, the date varying according to staff schedules. All cats at that time were also tested for FIV antibody and FeLV antigen[c], given oral anthelmintic[d] and topical insecticide[e] medication. Intact cats were spayed or neutered according to veterinary staff schedules.

Cats that developed symptoms of URD were moved to an isolation room until symptoms resolved. Cats were housed in stainless steel cages, varying in size from 18 x 24 to 48 x 30 (width by height in inches). Facing cage banks were 5-6 feet apart. Cages contained a litterbox, bedding, food and water bowls, and a hiding box if space allowed. The isolation room was within hearing of the dog kennels. Once in isolation cats were handled only by veterinarians or veterinary technicians and assistants.Gowns and gloves which were changed between each cat and shoe covers were used

Study participants were selected from the population of cats housed in the isolation room. To be eligible, cats had to exhibit one or more of these symptoms: sneezing more than two times per day, clear or colored ocular discharge (excluding cats with black crusted discharge only), conjunctivitis or chemosis, nasal congestion that could be heard when the cat was at rest, and clear or colored nasal discharge. Cats with a rectal temperature of 103°F or higher or with lethargy or anorexia were excluded from the study so they could be treated according to the shelter's normal URD protocol (Appendix).

The shelter's treatment protocol for URD was adapted from one published by the UC Davis Koret Shelter Medicine Program (Appendix).[14] Cats over 6 months old with moderate nasal congestion, colored nasal discharge, or severe chemosis and conjunctivitis were treated with doxycycline oral solution[f] (10 mg/kg PO q12h for up to 14 days). Doxycycline solution was prepared by combining 30ml suspending liquid[g] and 1/3 scoop doxycycline powder (1500mg). The solution was stored in amber bottles and refrigerated for up to 2 weeks. Cats under 6 months old with similar symptoms were treated with amoxicillin/clavulanic acid oral solution[h] (12.5 mg/kg PO q12h for up to 14 days). If a cat's symptoms did not improve after 4-5 days of doxycycline or amoxicillin/clavulanic acid, their treatment was changed to azithromycin[i] oral solution (10 mg/kg PO once, then 5 mg/kg PO q24h for 4 days). Cats with colored ocular discharge, severe chemosis or conjunctivitis were treated with 0.3% tobramycin[j] ophthalmic drops (2 drops q12h for up to 14 days). If a cat with ocular symptoms did not improve after 4-5 days on tobramycin and the cat was not already on an oral antimicrobial, doxycycline or amoxicillin/clavulanic acid solution was started as described above, according to the cat's age. A treatment was stopped when associated symptoms fully resolved or at 14 days, whichever occurred first. The shelter veterinarian determined further treatment if symptoms remained after day 14 of treatment. Additional non-antimicrobial treatments were prescribed by the staff veterinarian according to individual cat symptoms.

Study participants were placed into one of two groups. The treatment group was treated according to normal shelter protocol including antimicrobials. The control group was treated according to normal shelter protocol with the exception of antimicrobials. An alternating group assignment protocol was established to randomize treatment group assignment.

Cats in the control group who developed lethargy, anorexia, or rectal temperature of 103ºF or higher during the study period were removed from the study and started on antimicrobials according to the shelter's regular URD treatment protocol. Data collected on all cats in the study were: date entering the shelter, source (stray or owner surrender), estimated age, sex, neuter status (date of spay/neuter if within 1 week of onset of URD), body weight, current medical conditions, date of first symptoms of URD, date of URD symptom resolution, treatments for URD.

Each cat in the study was evaluated daily by one of the authors or shelter veterinary medical staff using the shelter's existing monitoring protocol (Appendix). The protocol assigns two daily numerical scores on a scale of 0 to 4 for ocular symptoms and 0 to 3 for nasal symptoms. This protocol was created by one of the authors who trained all parties in cat care and record-keeping. For this study, daily symptom scores were summed to calculate an overall severity score for each cat. The start date for URD was defined as the first day of symptoms. The URD endpoint was defined as the second day with zero symptoms, which ensured that subtle symptoms were less likely missed in recovering cats. URD duration in days was calculated by subtracting the start date from the end date.

A Fisher exact test was performed to evaluate the following variables between study groups: sex, neuter status, age, source (stray, owner surrender), and presence of recent surgery or concurrent medical conditions. Statistical analysis was performed on URD duration and severity scores as a randomized complete block design using the MIXED procedure of SAS 9.4[k] according to the following model:

$$Y_{ijk} = \mu + B_i + T_j + S_k + T_j \times S_k + \beta X_{ijk} + e_{ijk}$$

where Y_{ijk} = the dependent variable, B_i = the random effect of the ith block (I = 1-24); T_j = the jth treatment effect; Sk = the kth effect of source (owner surrendered or stray); $T_j \times S_k$ = the treatment by source interaction; β = the regression (covariate coefficient); X_{ijk}= the covariate measurement; and e_{ijk} = the residual error. The covariate used was the presence of any other conditions (concurrent medical condition, or surgery within 2 weeks of illness). Degrees of freedom were calculated using the Kenward-Roger option of the MIXED procedure. Any variable that was 2.5 SD from the mean was removed from the data. If the covariate analysis resulted in $P > 0.25$, it was removed from the model. For all variables, significant treatment and interaction effects were noted as $P \leq 0.05$ and trends were noted at $0.05 < P \leq 0.10$.

When possible, study participants were tested for bacterial pathogens via aerobic culture and PCR testing of pharyngeal swabs. Samples were collected by the authors before any treatments were administered. To collect samples, sterile swabs were rubbed along each cat's oropharynx. Collected swabs for PCR were moistened with sterile saline then placed into sterile test tubes. Collected swabs for aerobic bacterial culture were placed into tubes containing Amies agar gel.[1] All swabs were then refrigerated until transported to the testing laboratory. Swabs for PCR were packed with ice packs in carboard mailers and shipped overnight to the Cornell Animal Health Diagnostic Center. PCR testing was performed for *Bordetella, Chlamydia,* Influenza virus matrix, *Mycoplasma cynos* and *felis,* and Pneumovirus. Swabs for culture were driven to the New Hampshire Veterinary Diagnostic Laboratory and placed in an outdoor drop-box.

<u>Results</u>

45 cases of URD were evaluated among 38 cats. 3 suspected cases of URD were eliminated from the study because they did not meet study criteria upon veterinary examination. Of the remaining 42 cases, 25 were assigned to the antimicrobial treatment group and 17 to the control group (Table 1). In 7 cases a cat was deemed cured according to the study protocol and left isolation, then at a later date developed URD symptoms and entered the study a second time as a separate case. The interval between episodes of URD for these cats was 2 days for 3 cats, 12 days for one cat, 75 days for one cat, and 84 days for 2 cats. 3 of the cats with a second episode of URD changed from the control group to the antimicrobial treatment group. The other 4 cats remained in the same treatment group for both episodes, 2 cats in the treatment group and 2 in the control group.

The only factors that differed between groups were sex and neuter status (Table 2). The treatment group consisted of 10 neutered males, 10 spayed females, and 5 intact females. The control group consisted of 12 neutered males and 5 spayed females. Mean age for cats in the treatment group was 5.1 years (SD=4.5). Mean age for cats in the control group was 5.4 years (SD=4.5).

The least squares mean for duration of illness was 6.99 days for the treatment group and 6.28 days for the control group (*P*=0.61). The least squares mean for severity scores was 3.93 for the treatment group and 6.69 for the control group (*P*=0.26).

The duration of URD was not dependent on or affected by cat source (*P*=0.29), nor was there a effect of antimicrobial administration on duration of URD (*P*=0.7) (Table 3). There was no effect of antimicrobial treatment on severity score (*P*=0.34). However, there was a trend for an interaction on severity scores (p=0.06). Stray cats in the treatment group had a lower severity score than owner surrendered cats in that group (2.49 and 6.07 respectively); whereas stray cats in the control group had a higher severity score than owner surrendered cats in the same group (10.03 and 2.72 respectively).

Aerobic bacterial culture and bacterial PCR were performed on 14 cats in the study, 7 in each group. Shelter staff instituted treatment when cats entered isolation, and often before authors arrived, limiting the number of patients available to test. Of these cats, 11 tested negative for primary URD bacteria and 4 tested positive. Positive PCR results included *Mycoplasma*, *Bordetella*, and *Chlamydia* species or a combination of these. Aerobic bacterial culture results showed normal oral flora and various bacteria not considered primary causes of URD across all tested cats (Table 4).

Discussion

This study found no statistically significant difference in mean illness severity scores and mean duration of URD between cats treated with and without antimicrobials, suggesting that antimicrobial treatment did not improve outcomes.

The normal URD protocol for this shelter directed staff to administer antimicrobials when cats showed colored ocular or nasal discharge, based on the expectation that colored discharge indicates bacterial infection is present.[12] Colored discharge was noted in cats in both groups in this study, however, our results suggest that antimicrobials were not beneficial in the treatment of URD in this shelter. International Society for Companion Animal Infectious Diseases guidelines recommend delaying antimicrobial use in feline URD even with mucopurulent oculonasal discharge unless the cat exhibits fever, lethargy, or anorexia, because many cats will recover within 10 days without antimicrobial therapy.[12] These guidelines target treatment of privately-owned cats, but our results suggest that this recommendation may be used in certain shelter environments as well.

The two treatment groups differed significantly in the proportion of males and females (P=0.012) and in the proportion of neutered and intact cats (P=0.03). Low numbers made it difficult to calculate a relevant comparison between intact females, spayed females, intact males, and neutered males in the two treatment groups. Prior studies provide conflicting information about neuter status and sex as risk factors in URD.[7,18,19] Other factors that may have more impact on a cat’s URD susceptibility like age, source (stray vs. owner surrender), and co-morbidities or recent surgery were similar between groups. We theorized that cats under 1-year old, stray cats, and cats with a concurrent medical condition or recent surgery would have longer and more severe URD due to poor immunity than cats over 1-year old, owner surrendered cats, and otherwise healthy cats respectively. Since those factors were similar between groups, they should not have created bias in the results.

The trend for an interaction between cat source (owner surrender vs. stray) and severity score is worth consideration. Previously owned cats are more likely to have a complete immunization history and may be healthier than strays. Stray cats may therefore be more susceptible to secondary bacterial infection than owned cats.[18] That may explain why severity scores were higher in stray cats than owned cats when not treated with antimicrobials but also why severity scores were lower in stray cats when treated. If stray cats were more prone to secondary bacterial infection than owned cats, they would not only have more severe URD without antimicrobials but would also benefit more from them than owned cats. Another possible explanation is that previously owned cats might harbor more resistant microbes due to past access to veterinary care and higher likelihood of previous antimicrobial therapy.[20-22] Reduced treatment efficacy would be expected with higher rates of antimicrobial resistance, leading to higher severity scores in owned cats when antimicrobials were used.

It is unclear how often feline URD involves bacteria.[1,4-7]. Feline herpes virus is considered the most common primary causative agent of feline URD.[1,4,7,23-25] Many cats likely have co-infections, and many are suspected of developing primary viral infection with secondary bacterial infection.[23-25] Less certain is whether cats with secondary bacterial involvement can actually benefit from antimicrobial therapy.[12,16,17] Bacterial culture and PCR testing is rarely helpful in diagnosing and treating individual cases of feline URD due to the poor correlation between symptoms and positive test results and due to the difficulty in culturing bacteria like *Chlamydia* and *Mycoplasma*.[8,9,26,27] In shelter settings especially, this testing is rarely performed since it is not cost-effective. The role of diagnostic testing in shelters is more often to establish baseline pathogen prevalence in the population, and to assist with URD protocol development.[27]

This study attempted to test all cats with aerobic bacterial culture and bacterial PCR to establish the background presence of bacteria in the population and to compare results between study groups. However, sample collection was limited. Of the subjects who underwent testing, the majority had a negative PCR result. While the ambiguity of these test results must be noted, the generally negative PCR results suggest that viral infections were likely the primary cause of illness in this population, consistent with prior published works.[1,4,7,23,25] It is important to note that both groups had the same rate of positive bacterial PCR with two cases each, which suggests that there was a comparable low prevalence of primary bacterial infection between groups. Interestingly, three of the four positive PCR tests were positive for Pneumovirus, an emerging pathogen of unknown pathogenicity in cats.

Due to the limitations of testing for URD pathogens, empiric selection of antimicrobials for URD is common practice. The shelter protocol in this study used oral doxycycline and ophthalmic tobramycin as first line drug choices. In 5 cases amoxicillin/clavulanic acid was used instead (due to the patient’s young age in two cases, because the patient was already on the medication prior to URD symptoms in two other cases, and for unknown reasons in one case). Additionally, some cats were treated with doxycycline, some with tobramycin, and some received both, according to the shelter protocol. We elected to group all antimicrobial treatments together to increase the overall sample size. However, the variation in treatment within the study groups could have impacted outcomes.

This study has other limitations. It suffered from small sample size; thus, there may be a true difference between groups that was not detected. Staff turnover occurred during the study period. Although all staff members involved in scoring were trained by the same author and scoring was defined by specific clinical symptoms, error and bias may have occurred due to variations in staff. Personnel recording cat severity scores were also responsible for feeding, cleaning, and medicating study cats so they were not blinded to the cats' study group. Additionally, in several cases a staff member started a newly ill cat on antimicrobials before the authors arrived, which disrupted the alternating treatment group assignment. If staff believed these cats had more severe illness that warranted antimicrobials, that may have skewed the results, possibly weighting the antimicrobial treatment group with more severe cases. With more control over these variables, we would have more confidence in the validity of the study results. However, the disruption in group assignment occurred in only 4 cases so there may not be a significant effect on final results. Eliminating cats with evidence of systemic disease (fever, anorexia, lethargy) from the study likely minimized the effect of this bias by making the study participants more similar across groups. Conversely, study results can't predict the benefit of antimicrobials in cats with URD that includes systemic symptoms.

Compounded doxycycline powder used in the study was mixed with an oral suspension liquid[g] and kept refrigerated for up to 2 weeks. This is common practice in some animal shelters but may deliver less than desired doses of the drug.[28-30] FDA-approved doxycycline formulations are recommended to ensure appropriate and consistent drug doses are administered.[28,29] An FDA-approved doxycycline product may have created more significant improvements in URD than this study detected.

Calculating a severity score was complicated by the fact that illness duration impacted a cat's total severity score. Because all daily severity scores were summed to create each cat's overall severity score, more days of illness led to a higher severity score even if daily severity scores were continuously low. We opted to use the severity sum score rather than a mean severity score per cat that would then have been averaged to determine the group average. That could have created a condition known as Simpson's paradox, where a trend in individual data disappears when individual data sets are grouped together.[31]

This study may not be generalizable to other shelters with different environmental and disease pressures, nor to privately owned cats. *Bordetella* and *Chlamydia* were not detected in culture or PCR testing, and only 3 cats tested positive for *Mycoplasma*. If there truly were no primary bacteria involved, that may explain the lack of benefit from antimicrobials.. Several cats in both groups, however, showed mucopurulent oculonasal discharge, suggesting that bacteria were present. Regardless, less than half the study cats were tested, so few conclusions can be made on the basis of test results.

There is evidence that antimicrobials are overprescribed in veterinary medicine despite widespread information about antimicrobial stewardship.[15,16] URD treatment protocols vary widely between shelters. Many do not have written treatment protocols, many use antimicrobials before mucopurulent discharge occurs, and some rely on non-medical staff to make individual patient medication decisions.[16] Although we cannot rule out the benefit of antimicrobial therapy for URD in shelter cats, this study did not detect any and thus supports more judicious use of antimicrobial therapy for cats in shelter settings.

The study population had veterinary professionals in charge of medical care following a standard treatment protocol, housing designed to reduce stress, and other measures that improved overall cat health. It is important to address disease prevention measures like these prior to considering a change to antimicrobial use.

Further work in this area may include a similar study with a larger sample size and more standardized medication protocols. Working in a different shelter environment would be important to compare outcomes among shelters with varied cat populations and differing management and environmental conditions. Additional evaluation of the efficacy of antibiotic usage is worthwhile for shelters due to the high cost of medication administration and the stress it places upon cats. Antimicrobial stewardship is important to the future of animals, humans, and the environment, further warranting more research on the use of antimicrobials in the treatment of feline URD. This study's results support ongoing research in this area.

Acknowledgements

Supported by a grant from Maddie's Fund. The authors report no conflict of interest relating to this study.

Footnotes

a Feline Ultranasal FVRC, Heska, Loveland, CO.

b Fel-O-Guard 3, Elanco, Greenfield, IN.

c SNAP FIV/FeLV Combo test, IDEXX Laboratories Inc, Westbrook, ME.

d Pyrantel pamoate 50 mg/ml, Columbia Laboratories, Lexington, KY.

e Advantage Multi for cats, imidacloprid and moxidectin, Bayer Healthcare, Shawnee, KS.

f Doxycycline powder CONC??, Rood and Riddle Veterinary Pharmacy, Lexington, KY.

g Vehicle for oral suspension, USP-NF (Ora-Plus), OraMedix Inc, Lancaster, Calif.

h Clavamox Drops, Pfizer Animal Health, New York, NY.

i Tobramycin ophthalmic solution USP 0.3%, (generic), Bausch & Lomb, Bridgewater, NJ.

j Azithromycin for oral suspension USP 200mg/5mL (generic), Teva Pharmaceuticals, North Wales, PA.

k PROC MIXED, SAS, version 9.2, SAS Institute Inc, Cary, NC.

l BBL culture swab, Collection and Transport system, BD Diagnostics, Sparks, Md.

m Sucralfate 1g tablets (generic), Teva Pharmaceuticals, North Wales, PA.

n Mirtazapine 15mg tablets (generic), Watson Laboratories, Parsippany, NJ.

References

1. Scarlett, JM, Feline upper respiratory disease in: Miller, L, Hurley, K, eds. *Infectious Disease Management in Animal Shelters*, 1st ed. Ames, Iowa: Wiley-Blackwell, 2009;125-146.

2. Contreras, ET, *Assessment of novel strategies for the prevention and treatment of feline upper respiratory tract infections in shelters and feline herpesvirus-1 in laboratory settings* [dissertation]. Fort Collins, CO: Colorado State University; 2019.

3. Pedersen NC, Satop, R, Foley JE, et al. Common virus infections in cats, before and after being placed in shelters, with emphasis on feline enteric coronavirus. *J Feline Med Surg* 2004;6:83–8.

4. Thiry, E, Addie, D, Belak, S, et al. Feline herpes virus infection, ABCD guidelines on prevention and management. *J Feline Med Surg* 2009;11:547-555.

5. Garbal, M, Adaszek, L, Lyp, P, et al. Occurrence of *Bordetella bronchiseptica* in domestic cats with upper respiratory tract infections. *Pol J Vet Sci* 2016;19(2):353-358.

6. Egberink, H, Addie, D, Belak, S, et al. *Bordetella bronchiseptica* infection in cats, ABCD guidelines on prevention and management. *J Feline Med Surg* 2009;11:610-614.

7. Bannasch, MJ, Fley, JE, Epidemiologic evaluation of multiple respiratory pathogens in cats in animal shelters. *J Feline Med Surg* 2005;7:109-119.

8. Foley, JE, Rand, C, Bannasch, MJ et al. Molecular epidemiology of feline bordetellosis in two animal shelters in California, USA. *Prev Vet Med* 2002;54:141-156.

9. Zirofsky, D, Rekers, W, Powell, C, et al. Feline herpesvirus-1 and *Mycoplasma* spp. conventional PCR assay results from conjunctival samples from cats in shelters with suspected acute ocular infections. *Top Companion Anim Med* 2018;33:45-48.

10. Pesavento, PA, Murphy, BG, Common and emerging infectious diseases in the animal shelter. *Vet Pathol* 2014;51(2):478-491.

11. Tanaka, A, Kass, PH, Martinez-Lopez, B, et al. Epidemiological evaluation of cat health at a first-response animal shelter in Fukushima, following the great East Japan earthquakes of 2011. *PLoS One* 2017;12(3):e0174406.

12. Lappin, M, Blondeau, J, Boothe, D, et al. Antimicrobial use guidelines for treatment of respiratory tract disease in dogs and cats: antimicrobial guidelines working group of the International Society for Companion Animal Infectious Diseases. *J Vet Intern Med* 2017; 31:279–294.

13. Wong, DM, Blumberg, DA, Lowe, LG, Guidelines for the use of antibiotics in acute upper respiratory tract infections. *Am Fam Physician* 2006;74(6):956-966.

14. UC Davis Koret Shelter Medicine Program. Sample Treatment Protocol. Available at www.sheltermedicine.com/library/resources/?r=uri-sample-treatment-protocol. Accessed Feb 25, 2020.

15. Schmitt, K, Lehner, C, Schuller, S, et al. Antimicrobial use for selected diseases in cats in Switzerland. *BMC Vet Res* 2019;15:94.

16. Spindel, ME, Slater, MR, Boothe, D, A survey of North American shelter practices relating to feline upper respiratory management. *J Feline Med Surg* 2013;15(4):323-327.

17. Litster, AL, Wu, CC, Constable, PD, Comparison of the efficacy of amoxicillin-clavulanic acid, cefovecin, and doxycycline in the treatment of upper respiratory tract disease in cats housed in an animal shelter. *J Am Vet Med Assoc* 2012;241:218–226.

18. Dinnage, JD, Scarlett, JM, Richards, JR, Descriptive epidemiology of feline upper respiratory tract disease in an animal shelter. *J Feline Med Surg* 2009;11:816-825.

19. Helps CR, Lait P, Damhuis A, et al. Factors associated with upper respiratory tract disease caused by feline herpesvirus, feline calicivirus, *Chlamydophila felis* and *Bordetella bronchiseptica* in cats: experience from 218 European catteries. *Vet Rec* 2005; 156:669–73.

20. Leite-Martins, M, Prevalence of antimicrobial resistance in faecal enterococci from vet-visiting pets and assessment of risk factors. *Vet Rec* 2015;176(26):674–674.

21. Coetzee JF, Magstadt DR, Sidhu PK, et al. Association between antimicrobial drug class for treatment and retreatment of bovine respiratory disease (BRD) and frequency of resistant BRD pathogen isolation from veterinary diagnostic laboratory samples. *PLoS One* 2019;14(12): e0219104, doi: 10.1371/journal.pone.0089593.

22. Goossens, H, Ferech, M, Vander Stichele, R, et al. Outpatient antibiotic use in Europe and association with resistance: a cross-national database study. *Lancet* 2005;365:579-587.

23. Maddie's® Shelter Medicine Program, University of Florida, College of Veterinary Medicine. Feline Respiratory Infections in Animal Shelters, Available at https://sheltermedicine.vetmed.ufl.edu/files/2017/01/Feline-respiratory-infections-in-shelters.2018.pdf. Accessed Mar 1, 2020.

24. McManus, CM, Levy, JK, Andersen, LA et al. Prevalence of upper respiratory pathogens in four management models for unowned cats in the Southeast United States. *Vet J* 2014;201(2):196-201.

25. Berger, A, Willi, B, Meli, ML, et al. Feline calicivirus and other respiratory pathogens in cats with feline calicivirus-related symptoms and in clinically healthy cats in Switzerland. *BMC Vet Res* 2015;11:282.

26. Burns, RE, Wagner, DC, Leutenegger, CM, et al. Histologic and molecular correlation in shelter cats with acute upper respiratory infection. *J Clin Microbiol* 2011;49(7):2454-2460.

27. Cohn, LA, Feline Respiratory Disease Complex. *Vet Clin Small Anim* 2011;41:1273–1289.

28. KuKanich K, KuKanich B, Slead T, et al. Evaluation of drug content (potency) for compounded and FDA-approved formulations of doxycycline on receipt and after 21 days of storage. *J Am Vet Med Assoc* 2017;251(7):835-842.

29. Nahata, M. Stability of levothyroxine, doxycycline, hydrocortisone, and pravastatin in liquid dosage forms stored at two temperatures. *Int J Pharm Compd* 2015;19(5):428–431.

30. Papich, M, Davidson, G, Fortier, L. Doxycycline concentration over time after storage in a compounded veterinary preparation. *J Am Vet Med Assoc* 2013;242(12):1674-1678.

31. Malinas, G, Bigelow J, Simpson's Paradox. Edward N. Zalta. (ed.) The Stanford Encyclopedia of Philosophy (Fall 2016 Edition). Available at: https://plato.stanford.edu/archives/fall2016/entries/paradox-simpson. Accessed Sep 1, 2020.

URD Treatment Decision Tree and Scoring Chart. Used by shelter medical staff and authors when selecting treatments for cats with URD and when scoring URD cats' daily symptoms.

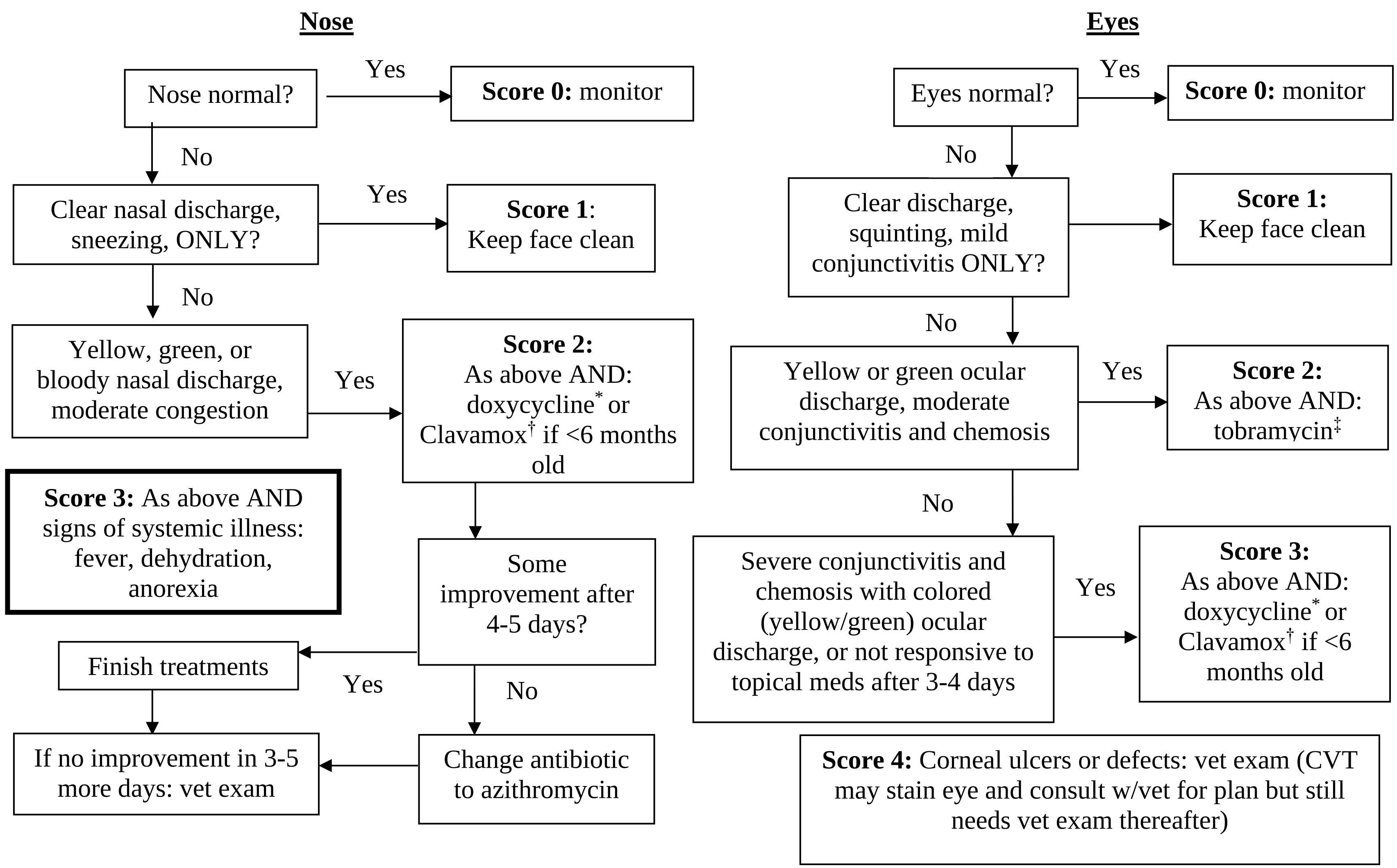

Table 1: Characteristics of study participants, duration of URD, severity score, and antimicrobial treatments used*

Group 0: No Antimicrobials								
Cat	**Age**	**Sex/ neuter status**	**Source**	**Weight in pounds**	**Other condition**	**URD duration in days**	**Severity Score**	**Interval[†]**
1	15 y	MN	S	7.8	hyper-thyroid	5	6.5	n/a
2	6y	MN	OS	13.7		7	3.25	n/a
3	2 y	FS	OS	7.7	recent spay	2	2	n/a
4A[‡]	5 mo	MN	S	4.5		5	6.5	n/a
5A[‡]	5 mo	MN	S	4.7		3	5	n/a
6A[‡]	5 mo	MN	S	3.9		3	4.5	n/a
7A[‡]	10 y	MN	OS	6	hyper-thyroid	8	14	n/a
7B[§]	10 y	MN	OS	6	hyper-thyroid	3	2.5	75 d
8	12y	FS	OS	7.8		4	3	n/a
9	3 y	FS	S	8.8	recent spay	7	6.5	n/a
10	12 wk	MN	OS	3		7	1	n/a
11	12 wk	MN	OS	3		7	1	n/a
12	4 y	MN	S	11		9	12.75	n/a
13A[‡]	9 y	FS	OS	10		6	7	n/a
13B[§]	9 y	FS	OS	10.4		7	0.5	12 d
14	6 y	MN	OS	10.8		23	22.25	n/a
15	3 y	MN	OS	13.4		5	3	n/a

	Group 1: Antimicrobials								
Cat	**Age**	**Sex/ neuter status**	**Source**	**Weight in pounds**	**Other condition**	**Days with URD**	**Severity Score**	**Interval[†]**	**Treatment**
4B[§]	5 mo	MN	S			6	2	2 d	D, T
6B[§]	5 mo	MN	S	3.9		2	0.5	2 d	D, T
5B[§]	5 mo	MN	S	4.7		14	4	2 d	D, T
16	4 y	MN	OS	11.6	recent neuter	3	2.5	n/a	D, T
17	6 y	MN	OS	10.8		16	9	n/a	D, T, SU
18	6 y	MN	S	11.6	puncture wound	8	4	n/a	T

19	7 y	MN	OS	14.5		5	2.25	n/a	T
20A[‡]	4 wk	F	S	1.3		8	20	n/a	C, T
20B	18 wk	F	S	4.1		3	1	84 d	D
21	10 y	FS	OS	6.9	pyometra	4	3	n/a	C, T
22	adult	F	OS	12		12	19	n/a	D, T
23A[‡]	4 wk	F	S	1.3		8	20	n/a	C, T
23B[§]	18 wk	F	S	4.8		3	1	84 d	D
24	8 wk	FS	OS	2.		5	4.5	n/a	T
25	10 y	FS	OS	6.8		4	3	n/a	D, T
26	1.5 y	MN	S	7.7		9	1.5	n/a	C, SU
27	12 y	FS	OS	11.8		11	9.75	n/a	D
28	12 y	FS	S	6.7	dental surgery	6	4.5	n/a	C, T, SU
29	15 y	FS	OS	5.2		12	19.75	n/a	D, T, SU
30	2 y	FS	S	9.2		19	41.5	n/a	D, azithromycin mirtazapine[n]
31	8 y	FS	OS	8.5		4	2.75	n/a	D
32	4 y	MN	stray	8.8		12	11.75	n/a	D
33	8 y	FS	OS	11.1	FIV +	10	7.25	n/a	T
34	8 y	FS	OS	11.1		10	5.25	n/a	D, T
35	6 y	MN	OS	9.9		9	22.5	n/a	D, T

*cats are listed in numerical order, not necessarily the order they entered the study. †- interval in days between first and second episode of URD. ‡ - first episode of URD in cat who entered study twice. § - second episode of URD in cat. (Treatments: D = doxycycline, T = tobramycin, C = amoxicillin/clavulanic acid, SU = sucralfate[m]) (Source: S = stray, OS = owner surrender, Sex/neuter status: MN = male neutered, F = female intact, FS = female spayed)

Table 2: Variation between treatment groups across several variables

	Group 0	Group 1	Fisher exact test p value
Sex			
Female	17	4	0.012[a]
Male	9	13	
Spay/neuter status			
Intact	7	0	0.03[a]
Altered	19	17	
Age			
<1 year	11	5	0.53
1-7 years	16	13	
>7 years	9	6	
Source			
Owner surrender	9	11	0.21
Stray	17	7	
Recent surgery or concurrent condition			
Yes	5	20	0.71
No	5	12	

[a]Values differ significantly (P<0.05) between groups.

Table 3: URD duration and severity score summary values according to treatment group and cat source (owner surrender or stray), least squares mean, and standard error of the mean reported.

	Least squares mean							
	Group 0		Group 1			P values		
	Owner surrender	Stray	Owner surrender	Stray	SEM †	Group 0 vs. Group 1‡	Owner surrender vs stray§	Interaction‖
URD duration	4.02	8.97	6.62	7.42	2.15	0.70	0.26	0.19
Severity Score	2.72	10.03	6.07	2.49	3.05	0.34	0.57	0.06

†Standard error of the mean

‡Main effect of antimicrobial use vs no antimicrobial use

§Main effect of owner surrendered cats vs. stray cats.

‖Interaction of main effects.

Table 4: PCR and bacterial culture results for 14 cats with URD

Group 0: No Antimicrobial treatment		
Animal	**PCR results**	**Bacterial culture results**
7A	-	*Staphylococcus felis, Pasteurella dagmatis, Pasteurella multocida*
7B	-	*Staphylococcus felis, Pasteurella dagmatis, Pasteurella multocida*
8	-	*Pasteurella multocida*
9	-	*Pasteurella multocida*
11	n/a	*Pasteurella multocida, E. coli*
12	Pneumovirus	*Staphylococcus aureus*
14	*Mycoplasma*	*Pasteurella multocida, Rothia nasimurium, Bergeyella zoohelcum*
Group 1: Antimicrobial treatment		
Animal	**PCR results**	**Bacterial culture results**
17	*Mycoplasma*, Pneumovirus	n/a
18	-	*Escherichia coli*
21	-	*Pasteurella multocida, Enterococcus faecium*
24A	-	*Enterococcus faecium, Staphylococcus lentus, Pasteurella multocida*
24B	-	*Enterococcus faecium, Staphylococcus lentus, Pasteurella multocida*
26	-	*Pasteurella multocida, Pasteurella stomatis, Neisseria zoodegmatis*
35	*Mycoplasma*, Pneumovirus	*Pasteurella multocida, Bergeyella zoohelcum*