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Madison J. Pattillo Kennesaw State University, mpattil2@students.kennesaw.edu

Lauren N. Mitchell Kennesaw State University, Imitch60@students.kennesaw.edu

Jessica A. Catchpole Kennesaw State University, jcatchpo@students.kennesaw.edu

Allison L. Martin Kennesaw State University, alm8737@kennesaw.edu

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Cover Page Footnote

Thank you to the staff and volunteers at Mostly Mutts Animal Rescue and Adoption in Kennesaw, Georgia for allowing us to conduct our study at your facility. Thank you to Lauren Faulkner for assistance with ethogram development, Christina Walthers for assistance with study design, and Sarah Barker for interobserver reliability calculations. Correspondence concerning this article should be addressed to Dr. Allison L. Martin, Department of Psychological Science, Kennesaw State University, 402 Bartow Ave NW, MD 2202, Kennesaw, GA 30144. Email: allison.martin@kennesaw.edu.

The Effects of Olfactory Enrichment on Shelter Dog Behavior

Madison J. Pattillo, Lauren N. Mitchell, Jessica A. Catchpole, and Allison L. Martin (Faculty Advisor)

Kennesaw State University

ABSTRACT

Shelter environments are stressful for dogs due to loud noises and unfamiliar surroundings. Previous research showed that exposure to some scents resulted in reductions in activity and vocalizations in shelter dogs. We investigated the effects of two calming (lavender and vetiver) and two stimulating (lemon and rosemary) essential oils on crate position and active, resting, and stress behaviors. There were 8, 5-min observations conducted each week per dog, split between baseline and scent exposure. Our analysis using the Wilcoxon Signed-Rank test found that dogs exposed to lavender (n = 13), lemon (n = 10), rosemary (n = 13), and vetiver (n = 12) did not show a significant difference in crate position or amount of time they engaged in stress, resting, or active behaviors compared to baseline. The lack of behavioral improvement indicates that scent enrichment alone may not be enough to have a significant effect on shelter dog behavior.

Keywords: shelter dog, scent enrichment, essential oil, behavior, activity

Introduction

Approximately 3.3 million dogs are housed in shelters in the United States each year (American Society for the Prevention of Cruelty to Animals [ASPCA], 2019). Dogs housed in a shelter environment are faced with several new changes, including restrictions of their environment, both socially and spatially, and this can lead to stress in the dogs (Beerda et al., 1999). Shelter-housed dogs can develop a variety of abnormal behaviors (Protopopova et al., 2014). The dog's undesirable, or stressrelated, behaviors in the shelter may be caused by the anxiety-provoking environment, and these undesirable behaviors may make them less attractive to potential adopters, which may lead to them staying in the shelters for months. While many studies have examined a variety of methods to improve the welfare of shelterhoused dogs, including social contact with humans or other canines (Hubrecht et al., 1992; Hubrecht, 1993), human contact specifically (Coppola et al., 2006; Kiddie & Collins, 2015), and environmental enrichment (Herron et al., 2014; Kiddie et al., 2017; also see review by Wells, 2004), few studies have been conducted on the effect of olfactory stimulation and how it may benefit the wellbeing of shelter dogs.

Olfactory stimulation, commonly referred to as scent enrichment, has been used in the shelter environment in several ways to increase the welfare of shelter dogs. Nose work activities, where dogs work to find specific scents paired with treats, have been used (ASPCA, n.d; Doyle, 2018; Walker et al., 2006), but there has been little to no scientific research conducted on the effects of nose work on shelter dog behavior. Another method used to increase welfare in shelter dogs is the use of dog appeasing pheromones (DAP). These pheromones are a synthetic replication of the natural pheromone associated with a lactating mother (Hermiston et al., 2018). Tod et al. (2005) found that when diffusing DAP in the shelter dog environment over a seven-day period, there was a significant decrease in both barking amplitude and frequency. Similarly, Hermiston et al. (2018) found a significant decrease in the intensity of barking when the dogs were exposed to the DAP spray, but no significant decrease in stress-related behaviors. There was no placebo included in the study.

In addition to these scent applications, some studies have investigated the influence of essential oils on behavior. Binks et al. (2018) found that when 15 shelter dogs were exposed to coconut, ginger, vanilla, and valerian essential oils on a washcloth, the dogs had decreased levels of vocalizations and movement, and coconut and ginger essential oils increased the amount that the dogs slept. A similar study conducted by Graham et al. (2005) diffused the essential oils into the environment of 55 shelter dogs. They found that essential oils that are typically considered relaxing, such as lavender and chamomile, led to a decrease in vocalizations and movements in the dogs. Essential oils that are considered stimulants, such as peppermint and rosemary, increased vocalizations and movement in the dogs. Further supporting the calming properties of both lavender and DAP, a study conducted by Amaya and colleagues (2020) found that dogs vocalized three to four times less and lied down more when exposed to lavender and DAP compared to the control group which was not exposed to either essential oils or DAP.

Given the success reported in previous studies, we aimed to further evaluate the usefulness of essential oils as scent enrichment in the shelter environment. We compared the behavior of dogs under baseline conditions (no scent) with their behavior when exposed to lavender, rosemary, vetiver, and lemon. We chose to expose shelter dogs to two previously studied essential oils, lavender (Lavandula angustifolia) and rosemary (Rosmarinus officinalis L.) (Amaya et al., 2020; Graham et al., 2005), and two essential oils not previously studied in the shelter environment, vetiver (Vetiveria zizanoides) and lemon (Citrus limonum). We chose vetiver essential oil because it has shown calming and anxiety reducing effects on dogs, and we chose lemon essential oil as it is used to decrease anxiety in dogs (Shelton, 2018). Additionally, Shelton (2018) recommends using lavender, lemon, and vetiver for anxiety and lavender, lemon, rosemary, and vetiver for improving behavioral conditions.

Due to safety concerns related to direct contact with and ingestion of essential oils by animals (see Benson, n.d., Shelton, 2018), we chose to use passive diffusion rather than the active diffusion (Graham et al., 2005; Hermiston et al., 2018) or direct contact (Binks et al., 2018) used in past studies. We placed essential oils onto cotton balls clipped onto the dogs' crates, therefore significantly reducing the risk of the dogs coming into direct contact with or ingesting the essential oils and thus ensuring the safety of the dogs during the study. In addition, we included stress behaviors as well as the previously studied behaviors of crate position, posture, and vocalizations to better measure wellbeing (Binks et al., 2018; Graham et al., 2005). We also chose to do focal animal observations rather than scansampling techniques used in previous

studies (Binks et al., 2018; Ellis & Wells, 2010; Graham et al., 2005), as we believed that this would provide us with more detailed behavioral information.

Based on Shelton's (2018) recommendations as well as past research, we hypothesized that all essential oils included in our study would reduce the amount of time dogs spent engaging in stress behaviors. However, given research showing that rosemary and lemon are stimulating essential oils (Graham et al., 2005; also see review by Wells, 2009), we hypothesized that dogs would be more active, spend more time at the front of their crate, and spend less time resting when exposed to these scents. Given the past research classifying lavender and vetiver essential oils as calming (Graham et al., 2005; Suyono et al., 2020), we hypothesized that dogs would spend more time resting, less time at the front of their crate, and be less active when exposed to these essential oils.

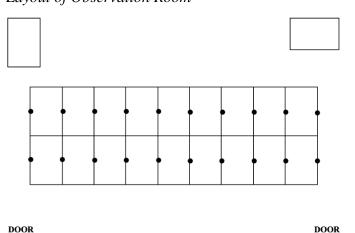
Method

Shelter Setting and Subjects. Prior to the start of data collection, our study was approved by the Institutional Animal Care and Use Committee (ACUP #19-003), and data collection on human activity in the room was approved by the Institutional Review Board (Study #18-538). Dogs in our study were housed in Mostly Mutts Animal Rescue located in Kennesaw, Georgia. Mostly Mutts is staffed by volunteers who walk dogs four times per day and feed dogs two times per day, seven days per week. Staff visit local county animal shelters and select dogs to bring to Mostly Mutts facility to be adopted. Dogs also arrive at the shelter through owner turn in, but this is less common. Not all dogs at Mostly Mutts stay in the shelter before they are adopted, some

dogs are taken by volunteers for short- or long-term fostering.

Dogs at the Mostly Mutts facility are housed in multiple rooms. In the room in which we conducted behavioral observations, dogs were housed in wire crates measuring 54" in length, 36.5" in width and 45" in height. The crates were placed back-to-back in two rows of nine, with two additional crates in the back corners that were not included in the study (Figure 1). There were large plastic dividers that bordered the outside of three out of the four sides of the crates, so the dogs could only see out of the top of their crate and the front. An information card was attached to the top of the crate with various details about the dog, as well as a white board for the volunteers to make notes. Dogs were provided with water, blankets, and toys in their crate unless destructive behaviors with these items were observed.

Figure 1 Layout of Observation Room



We observed a total of 58 dogs over a span of eight months. Behavioral data from dogs who were not observed for a minimum of three observational sessions in both the control and the treatment were excluded from the study. Due to the high turnover typical in dog shelters, of those 58 dogs, 31 met the criteria for inclusion. The dogs included in our analysis ranged in age (1 - 11 years, M = 3.94 years), weight (17 - 56 lbs., M = 34.97 lbs.), sex (11 male, 20 female), origin (27 from shelter, 4 owner surrender), how long they have been in the shelter (6 - 280 days, M = 50.03 days), and their primary breed (Hound = 5, Shiba Inu = 1, Terrier = 2, Chihuahua = 1, Shepherd = 3, Labrador = 11, Retriever = 1, Boxer = 1, Beagle = 4, Australian Cattle Dog = 1, Miniature Pinscher = 1).

Experimental Design and

Procedure. We conducted a within-subjects design with a control and treatment phase. For the control phase, we attached a single cotton ball with no added essential oils to an alligator clip (Figure 2). The clip was then attached to the dividers in between each crate and on the ends of each row, excluding the two crates in the corners (Figure 3). There were a total of 20 cotton balls in the room during the observation period, and they are represented by the black dots in Figure 1. For the treatment, we placed two drops of the essential oil on the control.

Figure 2 Cotton Ball on Alligator Clip

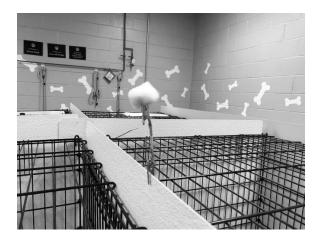


Figure 3 Room Set Up with Cotton Balls



The essential oils (lavender. rosemary, lemon, and vetiver) were each assigned a number. We used a random number generator at the beginning of each four-week block to assign the order in which the essential oils were used. Behavioral observations were conducted eight times per week, focal observations lasted five minutes in duration, and dogs were watched twice per observation block. Observations for the control occurred in the morning of Day 1 and the afternoon of Day 2, and observations for the treatment occurred in the morning of Day 3 and the afternoon of Day 4. All observations were conducted between feeding and walking times when shelter activity was low. Only one dog was watched at a time, and that dog is referred to as the focal dog during data collection. Observations occurred while the observer was sitting on a mat on the floor in front of the focal dog's crate. Prior to the observation, the observer included a threeminute acclimation period, where they just sat quietly in front of the crate for the dog to get used to a person sitting and watching. Some dogs were exposed to the same scent multiple times, and only the data from the first exposure was used.

Our behavioral ethogram (Appendix I) was downloaded in the BORIS app for

Android (Version 0.2.3; Friard & Gamba, 2016; Friard & Gamba, 2018) on Samsung Galaxy Tab 2 (10.1) tablets which we used during our observation sessions. Data were then imported and analyzed into BORIS for Windows (Version 7.4.10; Friard & Gamba, 2016; Friard & Gamba, 2019). Prior to the start of data collection, observers were trained to >85% inter-observer reliability, with ongoing reliability checks during 13.72% of observations (Reliability M = 91.92%).

Data Analysis. For our analysis, we grouped the behaviors from the ethogram into four behavioral categories: Front (crate front), stress (lip lick, yawn, tremble, cratedirected, pacing, panting, vocalizations, selforal, scratch), active (eat/drink, object interaction, move, stand), and resting (lie head down, lie head up, sit). For behavioral categories for which all behaviors were mutually exclusive (resting, front), we calculated the percent duration. Some of our behaviors in our stress and active behavior categories were not mutually exclusive (such as standing and eat/drink), so we calculated the percent of available time the dogs engaged in each behavior category. To calculate the percent of available time, we used the following formula with abbreviations as follows: total duration of time engaged in the category (total beh category dur), session duration (session dur) and the number of nonmutually exclusive behaviors in category (# nonmutual beh):

 $\frac{\text{total beh category dur}}{\text{session dur} \times \# \text{ nonmutual beh}} \times 100$

This allowed us to determine what percent of the time the dogs were engaging in the behaviors in the available time. We conducted our analysis in SAS Studio v. 9.04 software using the Wilcoxon Signed-Rank test (alpha = .05) to compare the difference in the percent of time spent engaging in the behaviors from the control to the treatment. To estimate the effect size for our Wilcoxon Signed-Rank test, we used a Matched-Pairs Rank-biserial r (Table 1) using IBM SPSS Statistics software v. 26.0.

Results

We did not find any significant difference in crate position, active, resting, or stress behavior when dogs were exposed to lemon (n = 10), rosemary (n = 13), lavender (n = 13), or vetiver (n = 12)essential oils as compared to baseline (Wilcoxon Signed-Rank Test, see Table 1). However, we observed nonsignificant trends (.05 for dogs to spend less timethe front of their crates when exposed to rosemary (p = .08, r = -.56) and more time engaging in active behaviors when exposed to lemon (p = .06, r = .67). The majority of effect sizes (r) were small (< .3), but two behavioral conditions had medium effect sizes $(.3 \le x < .5)$ and six behavioral conditions had large effect sizes ($\geq .5$). Refer to Table 1 for details on effect sizes.

Discussion

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Overall, in contrast with our hypotheses, our study did not find a significant difference in crate position, stress, active, or resting behavior when the dogs were exposed to the lavender, lemon, rosemary, and vetiver essential oils via passive diffusion. Regarding the effect on front behavior in the crate, lavender, lemon, and vetiver were not significant. However, during the rosemary condition, time spent in the front of the cage on average decreased and trended close to significance (see Table 1).

Behavior	Control Median	Treatment Median	Median Difference	IQR	S-Statistic	P-value	Matched-Pairs Rank-Biserial r
Front *							
Lavender	25.46	52.01	-0.95	69.16	-2.5	.89	.06
Lemon	51.17	68.19	-10.34	57.59	-1.5	.92	.05
Rosemary	71.04	35.16	25.24	35.63	25.5	.08	56
Vetiver	63.95	66.35	10.47	39.35	17.0	.20	44
Stress^							
Lavender	0.64	0.70	-0.03	0.98	-5.5	.74	.12
Lemon	2.24	1.75	-0.46	2.51	-3.5	.77	.13
Rosemary	1.67	0.99	0.61	2.22	12.5	.41	27
Vetiver	0.46	1.14	0.04	2.23	2.0	.91	05
Active ^							
Lavender	4.20	8.43	-0.58	10.47	-12.5	.41	.27
Lemon	5.39	8.75	-2.67	3.36	-18.5	.06	.67
Rosemary	8.35	2.66	4.97	7.81	23.5	.11	52
Vetiver	6.98	3.33	3.56	9.21	20.0	.13	51
Resting*							
Lavender	87.16	83.63	1.97	14.91	10.5	.50	23
Lemon	82.24	72.44	6.32	13.46	11.5	.28	42
Rosemary	84.06	91.06	-5.94	19.98	-23.5	.11	.51
Vetiver	81.93	90.36	-10.42	18.68	-21.0	.11	.54

Table 1 . Median percent duration (*) or percent of available time (^) in control (no scent) and Image: control (no scent) and
treatment (scent) conditions, results of Wilcoxon Signed Rank Test, and effect size estimates
(Matched-Pairs Rank-Biserial r)

Crate position is noteworthy in that dogs in the front of their crate have more of an opportunity to interact with potential adopters, and previous studies have found dogs that spent more time in the front of their crate were more likely to be adopted (Protopopova et al., 2014).

Rosemary has been shown in previous studies to be a stimulant and to increase movement in dogs (Graham et al., 2005); however, in our study, dogs exposed to rosemary showed trends toward decreasing the amount of time spent in the front of the crate and in displaying active behaviors. Given these conflicting results, more research is needed on the impact of rosemary on canine behavior.

None of the essential oils had a significant effect on stress and resting behavior. For active behavior, lavender, rosemary, and vetiver were not significant, though lemon trended close to significant, in that during exposure to lemon, dogs spent more time engaging in active behaviors, though the increase did not reach statistical significance. Because lemon essential oil has not been previously studied in the shelter environment, there are not precedents on how dog behavior could be influenced by this essential oil. In mice, studies of lemon essential oil showed antidepressant-like properties (Hao et al., 2012). In humans, lemon essential oil has been found to improve concentration, mood, and attention (Akpinar, 2005). A similar behavioral effect could be present when dogs are exposed to lemon essential oil. These data trends warrant further investigation into the use of rosemary and lemon to influence behavior in shelter dogs that is deemed positive.

Previous studies (Binks et al., 2018; Graham et al., 2005) used more active approaches to essential oil diffusion over a longer period of time. Active diffusion

methods can change the concentration or chemical makeup of essential oils, and active diffusion methods result in a stronger concentration of the essential oil in the air. Other studies allowed direct contact with the scent (e.g., Binks et al., 2018). While previous studies allowed direct contact with the essential oils, we had several concerns for the safety of the dogs as essential oils can be toxic to dogs when ingested (Flint & Brutlag, n.d., Tisserand & Young, 2013) Therefore, we chose to do shorter exposure times with a weaker concentration of essential oils to reduce the chance of the dogs becoming overwhelmed by the essential oils and ensured that the dogs had no physical interaction with the cotton balls that held the scent.

In using passive diffusion, there is a possibility that the concentration of essential oil was not strong enough to have a significant effect on the behavior of the dogs in the study, because this method relies on evaporation of essential oils instead of a medium which vaporizes and forces essential oil molecules into the air. Longer periods of exposure and stronger essential oil concentrations used in previous studies may also explain why we found no behavioral difference in our study, but prior studies did. However, dogs have an extremely powerful sense of smell and what may seem weak in comparison to human noses may be strong to dogs. Therefore, differences in prior study results could be due to circumstances other than the concentration of essential oils used.

Future research should attempt to replicate the trends observed in rosemary and lemon in a larger sample. While the results of our study were not significant, we observed several nonsignificant trends with large (r > .50) effect sizes. The effect sizes were large in the trends for activity behaviors to increase during lemon exposure but decrease during rosemary. In addition, large effect sizes were seen in the trends for resting behavior to increase during exposure to rosemary and vetiver and for time spent in front of the crate to decrease during rosemary exposure. The large effect sizes may indicate that the essential oils did influence the dogs' behavior but that our sample size was too small to produce statistically significant results. Future research with a larger sample size should focus on these behavioral conditions and scents as they had the largest effect sizes. A larger sample size would also allow for an examination of how individual characteristics such as sex, time at shelter, size, or breed might influence a dog's response to essential oils.

Given the discrepancies between our findings and those from other studies, future research should systematically compare different diffusion techniques and exposure times to investigate which are the key elements in impacting dog behavior. Furthermore, future research in scent enrichment could investigate whether dogs prefer one scent over another and, if so, whether that scent may have a calming effect on the dogs, even if the scent is typically considered stimulating. In addition, certain essential oils used in this study, such as lavender, may not only have the potential to provide a similar therapeutic effect by providing relaxation, but they can also help with the smell of the shelter environment, which overall improves potential adopters' experience. If potential adopters spend more time in the shelter, it may increase the chance of dogs being adopted. In conjunction with music, essential oils were found to increase the pleasantness of shoppers' experience and made it more likely for shoppers to buy products (Matilla & Wirtz, 2001). Since many shelters already employ the use of calming music as a means

of decreasing stress in shelter dogs, this could be used in conjunction with essential oils to influence the behavior of people in the shelter, potentially increasing the likelihood of adoption. Future studies could look at the connection between essential oils' contribution to behavior in people and the link to adoption rates in shelters.

Our study was limited by several factors, including small sample sizes for each scent and high turnover rates. Research involving animals in applied settings, such as shelters, often involve small sample sizes due to animal availability and high rates of turnover, and our sample sizes are in line with previously published research on scent enrichment (e.g., Binks et al., 2018). While focal observations conducted on each dog allowed for a more detailed observation, the observations were limited in time (only five minutes) and therefore did not represent a full-time budget for the dogs.

While our study did not find significant differences in scent enrichment on dog behavior, there are many factors that influence a dog's behavior, and essential oils may have only played a small part of that. However, because none of the essential oils significantly increased stress behaviors, it appears that there is no negative implication of using essential oils in the shelter. Because there does not appear to be negative implications in the use of essential oils in the shelter environment, continued research should focus on the impact that essential oils could have on the welfare of shelter dogs and its effect on adoption rates.

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Appendix I

Behavioral Ethogram

Front

Behavior	Definition
Front	The majority of the dog's head is positioned in the front half of the crate.

Behavior	Definition		
Crate-Directed	Active, physical interaction with some part of the crate, including the crate pan or crate bars. This includes biting, nose pushing, licking, or pawing behaviors in which the nose, mouth, tongue, or paws is makin physical contact with the crate.		
Lip lick	Dog puts tongue outside its mouth and touches its lips (upper, lower, or both simultaneously).		
Pacing	Full body, patterned, locomotion within the crate (ex. back and forth, circles, diagonal, etc.). Recorded after 3 rotations. Turn off after 3 seconds when the dog is no longer doing the behavior.		
Pant	Deep breaths with open mouth, without retracted lips.		
Scratch*	Dog uses front or back paws to rub against body.		
Self-oral	Dog licks or bites (front teeth only or biting without injury) fur or skin		
Vocalization	 Dog makes audible sounds from throat area. Score this behavior 3 seconds after first vocalization is heard and end 3 seconds after vocalization ends. Barking*: Staccato vocalizations. If barking is accompanied by other vocalizations, barking takes scoring priority. Growling: Low, buzzing sound. Howling: A long drawled, out sound through partially closed jaws. Muzzle is often raised. Whine: A high, sustained pitch. Only score of tone is full pitch (not airy) or if mouth or cheek movement is visible. Cough: Sudden audible expulsion of air through mouth. Other: Vocalization does not fit into any previous category. 		
Tremble	Small vibrations visible in dog's body. Score this behavior 3 seconds after noticeable vibrations in any part of dog's body. End 3 seconds after noticeable vibration stops.		
Yawn	Dog opens mouth widely and inhales.		

Stress

Active

Behavior	Definition	
Eat/Drink	Dog laps water or orally ingests food items.	
Object Interaction	Dog uses its mouth or body to interact with an object in the cage.	
Move	Dog's front two feet change quadrants within the crate. Dog must be bearing weight on feet.	
Stand	Dog's weight is on legs, abdomen or side is not on ground. Dog is stationary.	

Resting

Behavior	Definition
Lie, head down	Dog rests its weight on its abdomen, side, or back. Head rests on surface, including crate, paw, bed, etc.
Lie, head up Dog rests its weight on its belly, side, or back; Head is lifted ground.	
Sit	Front legs are straight and erect, back legs are bent.

* based on Overall (2014)