

Adaptive mechanisms in dogs adopted from shelters: a behavioral assessment of the use of a synthetic analogue of the canine appeasing pheromone

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Abstract: Adaptation to a new socio-environment might represent a very hard step for sheltered dogs, because of a higher level of difficulty in coping with unfamiliar conditions. The adaptation process can be logically and scientifically related to the concept of stress and welfare, limiting the success rate of rehoming. A synthetic analogue of the Dog Appeasing Pheromone (Dog Appeasing Pheromone, DAP) is reported to have a reassuring effect in puppies and adult dogs in a wide variety of stressful situations. The aim of the current study was to investigate the effects of DAP (Adaptiil[®] Ceva Vetem S.p.A.) in dogs re-homed from rescue shelters. The study was designed as a prospective open-label clinical trial.

Significant decreases were observed in adult dogs for wandering in the house restlessly ($p=0.022$) and hiding fearfully in protected corners ($p=0.033$), whereas in puppies treatment with DAP significantly ($p<0.05$) improved the reaction towards unfamiliar dogs ($p=0.048$) and wandering in the house restlessly ($p=0.022$). In both adults and puppies a significant improvement in interaction with owners was observed. In particular, “looking continuously for the owners” and “following the owners everywhere like a shadow” were significantly improved ($p=0.0012$ and 0.0016 respectively) in adult dogs. Separation reactions revealed a significant decrease ($p<0.05$) and in puppies the tendency to vocalize in absence of the owner was also significantly reduced ($p=0.0029$). Both adults and puppies showed a decreased tendency to wake suddenly in the night ($p=0.024$ and $p=0.026$ respectively) and wander around the home ($p=0.012$ and $p=0.026$ respectively). In contrast, for house-training no significant difference was reported in adults, whereas for puppies there was a significant decrease ($p<0.05$) in the mean scores for urination and/or defecation wherever in the house and after coming home. Data regarding the overall assessment suggested a significant improvement in all the efficacy variables considered in the study. The analysis of owners’ degree of satisfaction at the final visit showed that DAP treatment was considered successful by 84.4 % of owners.

Results suggest that DAP might improve dogs’ adaptability throughout the first weeks following adoption and can be considered a useful tool for reducing stress in re-homed dogs.

Key Words: sheltered dogs; pheromones; dog appeasing pheromone; adoption; adaptation.

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Introduction

A prolonged stay in shelters may influence the behavior of dogs, since social isolation and spatial restriction can affect dogs adversely (Hubrecht et al., 1992; Wells & Hepper, 1992) with severe consequences on animal welfare (Hubrecht & Turner, 1998; Taylor & Mills, 2007a). There are many issues involving pet relinquishment and improvements may be achieved with better shelter facilities, thorough training of the animal and addressing the expectations of the owner before adoption (Marston & Bennett, 2003; Mondelli et al., 2010). Satisfactory information about the management and policy of rescue associations might improve the number of successful adoption processes (Shore, 2005). Nevertheless, adoption often represents a stressful event for the animal, mainly when the dog is emotionally compromised based on previous detrimental events and on the shelter experience (Wells & Hepper, 2000).

Adaptation to a new socio-environment might represent a very hard step for sheltered dogs (Van der Borg et al., 1991; Beata, 2006), because of the difficulty in coping with unfamiliar conditions (Tod et al., 2005). As far as puppies and young subjects are concerned, even a short lifespan in the shelter might significantly influence their behavior and physiological responses because of the crucial phase of their psychosomatic growth. Furthermore the owners are not always aware of the behavioral problems shown by dogs adopted from a shelter (Gazzano et al., 2009). A destabilization phase related to the challenge of coping with the new environment and lifestyle is sometimes observed, although a specific psychological disease is not likely to be diagnosed. The importance of this adaptation period should not be underestimated as it could represent the origin of severe behavioral disorders, as reported by different authors (Wells & Hepper, 2002; Hennessy et al., 2001). Moreover, it can represent one of the reasons for returning the dogs to the shelter after adoption. In fact Luescher & Medlock (2009) have observed that about 20% of adopted dogs are re-relinquished to the shelter, primarily due to behavioral problems. Chewing, digging, or scratching at objects was the most common behavior problem for dogs 1 month after adoption (Lord et al., 2008). This was similar to the findings of a previous study (Wells & Hepper, 2000), in which destructiveness was reported for 25% of dogs 1 month after adoption. The high frequency of problems associated with chewing, digging, and scratching at objects among dogs in that study (Lord et al., 2008) may also have been related, in part, to anxiety. In the same paper it has been reported that previous studies have shown that dogs obtained from animal shelters show an increased risk of separation anxiety, and separation anxiety often manifests as destructive chewing and scratching behaviors.

Chemical communication has been widely explored in animals (Wyatt, 2009; Wyoscki & Preti, 2009). Canine pheromones are molecules secreted by different glands that allow intra-specific olfactory communication (Pageat & Gaultier, 2003a). Dogs might perceive pheromones with their vomeronasal organ (VNO), a paired structure situated above the hard palate which opens to the incisive canal. The stimulation of specific receptors localized on the membranes of the VNO nervous cells allows information to be transmitted to the accessory olfactory bulb (Halpern, 1987). Efferent connections in turn stimulate different structures within the limbic system, thus altering the emotional state of the individual (Tirindelli et al., 2009). Combined anatomical, physiological and behavioral studies provided new information that furthered our understanding of the chemo-architecture of the VNO and the pheromonal system (Halpern & Martinez-Marcos, 2003). In the dog it has been discovered that specific pheromones are secreted by the mammary complex of bitches from 3 to 4 days after parturition and persist up to 2-5 days after weaning; as they show appeasing actions on puppies they are also known as "appeasines" (Pageat & Gaultier, 2003a).

Dog Appeasing Pheromone (DAP) is a synthetic mixture of simple compounds that have been identified from the secretions of the sebaceous glands in the inter-mammary sulcus of bitches shortly after parturition (Pageat, 1999). This pheromonal analogue of the appeasing pheromone secreted by nursing bitches has shown efficacy in the management and control of behavioral stress and fear/anxiety related signs in dogs (Pageat & Gaultier, 2003b; Taylor & Mills, 2007a). In particular, DAP helps to establish the puppy in the new family, stops or prevents fear and stress related signs in puppies and adult dogs and comforts the dog in stressful environments. Research has actually shown that the reassuring properties of the appeasing pheromone persist even into adult age; thus they appear to modulate both the emotional state and the social interaction of the dog throughout its life (Sheppard & Mills, 2003; Mills et al., 2003; Mills et al., 2006; Gaultier et al., 2005; Tod et al., 2005; Estelles & Mills, 2006; Graham et al., 2007; Levine et al., 2007).

An increasing concern for the welfare of dogs housed in rescue shelters has been reported (Pageat & Gaultier, 2003b; Taylor & Mills, 2007a). It is well known that one of the most effective ways to improve the long-term welfare of a sheltered dog is to ensure that the animal is adopted and to reduce the number of dogs returning to the shelter after adoption. From this point of view, the dog's behavior after the introduction in the family can determine whether or not the animal will be regarded as desirable by the owners.

The aim of the present study was to investigate the use of DAP (Adaptil® Ceva Vetem S.p.A) in dogs re-homed from rescue shelters. In line with the clinical developmental process adopted in human medicine, an open, uncontrolled trial was proposed to assess the pheromone's potential efficacy in supporting the adoption step in re-homed dogs. Such studies, also called initial phase 2 studies, are often used before investing in the resources required to conduct a placebo-controlled trial (Reid et al., 1996). This type of study was also justified because of the need to safeguard the welfare of the dogs in the care of the rescue association shelter (Sheppard & Mills, 2003).

Contrary to other published trials performed with DAP, which investigated the use of pheromones as a therapeutic option for controlling undesirable behavior in dogs (as critically reviewed by Frank et al., 2010), in the present study pheromones have a preventive value, trying to optimize the adaptation phase to the new home, and not a curative role. This evaluation in effectiveness prevention has been done similarly to Taylor & Mills (2007b) and Gaultier et al., 2008.

Materials & methods

Animals

Thirty-two, 16 puppies and 16 adults, out of 80 dogs were finally included in this prospective open (non-blinded) label clinical trial. Dogs were recruited among animals housed in a metropolitan private animal shelter (ENPA, Turin, Italy).

To be suitable for inclusion in the trial, the dogs had to be healthy, of any breed or mix-breed, aged between 1 and 7 years for adults, 2 and 6 months for puppies, and between 2.5 and 80 Kg of weight.

All the dogs were health checked when they entered the shelter and they received a high standard of veterinary health care throughout their stay in the humanitarian facility. Clinical data were obtained from medical records including a standard complete physical examination performed by the shelter veterinarian. Among the population of 80 dogs 72 dogs remained, since 8 dogs were ruled out for severe clinical problems (cardiological 4, neurological 2, orthopedic 2). It is not ethical to involve unhealthy animals in a behavioral trial unless strictly necessary. In addition, this group of dogs underwent a preliminary behavioral (Van der Borg et al., 1991-modified- reported in Bergamasco et al., 2010) evaluation performed by a certified ECAWBM-BM examiner (MC.O.). Twelve dogs were ruled out for aggressive behaviors towards people and excessively fearful reactions. Additional exclusion criteria were: treatment with psychotropic drugs within the last 60 days before the beginning of the study, presence in the shelter for less than 2 months for adults.

Out of these 60 dogs, 32 dogs adopted within the experimental time were finally included in the study, after the new owners' acceptance. Some dogs were adopted but the owners did not want to participate in the study (n=10), some dogs were not adopted (n=9), 9 dogs left the study before it ended (n=2 did not begin the study, n=7 left the study after the first month).

No dogs in the 32 included returned to the shelter, according to a phone follow up at one month, two months and 6 months after adoption.

Study design and data collection

At adoption (V_0) a diffuser with DAP (Adaptil® Ceva Vetem S.p.A) was given to the owners who agreed to participate in the study; they were briefed verbally about the procedure, although no behavioral consultations were given throughout the study time. In particular, no instructions were given to the new owners to prevent behavior problems in their newly adopted dog or puppy. They used the standard method in the shelter, that is, general and not professional advice. If the problem was too severe, the study had to be interrupted immediately.

Owners that agreed to participate in the study were asked to sign an informed consent. The DAP diffuser (Adaptil[®] Ceva Vetem S.p.A) had to be plugged in the room in which the dog was likely to spend most of its time during the day, and it had to cover an area of up to 70 m². Since one diffuser lasts approximately 4 weeks, at V₁ a refill was given. At V₁ (28 ± 3 days after the beginning of treatment) and V₂ (56 ± 3 days after the beginning of treatment) the owners were interviewed during a control visit of the dog at the shelter, to assess the owners' compliance and reliability. At V₂ treatment was stopped.

Only one investigator, a graduating student in veterinary medicine, was trained in interviewing techniques and asked the questions without any suggestion to the owner about the answers. The behavioral variables considered to assess DAP efficacy were: degree of general activity, socio-environmental interaction, separation reactions, sleep/wake cycle and house training.

Based on a behavior check list for reference (Table 1) developed by the investigators, each owner was expected to provide the following information about his dog's behavior during both V₁ and V₂ visits: a rating (using the four-point frequency scale) of the frequency of the behavioral signs displayed (0 never, 1 rarely, 2 frequently, 3 always); a rating of the evolution of each behavioral sign from V₁ to V₂, on a five point change scale (-2 much worse, -1 slightly worse, 0 same, +1 slightly better, +2 much better); a rating of the dog's overall response using a four-point change scale (resolved, improved, unchanged, worse). All the owners were required to answer to this last question, even in the case of no problem detected.

Table 1. Behaviors included in the assessment checklist.

Socio-environmental reactions
<ul style="list-style-type: none"> • Wandering in the house, panting and trembling, hiding in protected places • Reaction to unfamiliar people, telephone and door ringing, unfamiliar noises
Human-dog interaction
<ul style="list-style-type: none"> • Interaction and play with the owners • Attachment degree • Emotional urination
Separation reactions
<ul style="list-style-type: none"> • Vocalization • Urination, defecation, vomiting, diarrhea • Destructiveness
Sleep/wake cycle
<ul style="list-style-type: none"> • Restless before and/or during sleeping
Housetraining
<ul style="list-style-type: none"> • Urination and defecation indoor

Moreover, the owners' degree of satisfaction was considered. The owners were also asked to rate their degree of satisfaction with the pheromone treatment (very satisfied, satisfied, dissatisfied, mainly dissatisfied) as an indication of the clinical significance of any response to the treatment (Sheppard & Mills, 2003).

Statistical analysis

The data sets were not normally distributed. Non parametric Wilcoxon rank sum tests were therefore used to compare the baseline ratings (V₁) of each behavior with the final ratings (V₂);

the Wilcoxon match paired test and the Kruskal Wallis test were used to analyze the ratings on the change scale at the final assessments. The owners' overall assessment and degree of satisfaction were analyzed by means of the Chi-square test or Fisher exact test according to the theoretical values obtained.

The significance limit is set at $P < 0.05$. Statistical analysis was performed using SAS Institute Inc software, version 8.2.

Results

The data for adult dogs and puppies are presented in Table 2 and 3 respectively.

Table 2. Evolution, over time, of behavioral parameters in adult dogs treated with DAP (mean values \pm SD; n=16). V1 = 28 days after the beginning of treatment, V2 = 56 days after the beginning of treatment. $P < 0,05$; n.s. = not significant.

Parameters	V1	V2	p value
Socio-environmental reactions			
Wander in the house restlessly	0.81 \pm 1.11	0.19 \pm 0.40	0.022
Pant/tremble, resting in a corner	0.38 \pm 0.72	0.06 \pm 0.25	n.s.
Hide in protected places	0.88 \pm 1.02	0.13 \pm 0.34	0.033
React to unfamiliar people	1.13 \pm 1.26	0.56 \pm 0.63	n.s.
React to telephone and door ringing	1.19 \pm 1.22	0.56 \pm 0.63	n.s.
React to unfamiliar dogs	1.13 \pm 1.09	0.50 \pm 0.63	n.s.
Human-dog interaction			
Interact inappropriately with the owners	0.63 \pm 0.81	0.25 \pm 0.58	n.s.
Play inappropriately with the owners	0.69 \pm 0.79	0.31 \pm 0.60	n.s.
Look continuously for the owners	2.47 \pm 0.83	1.20 \pm 0.56	0.0012
Follow the owners everywhere like a shadow	2.50 \pm 0.82	1.31 \pm 0.70	0.0016
Release urine during exuberant play	0.56 \pm 0.89	0.19 \pm 0.40	n.s.
Separation reactions			
Vocalize in the owners' absence	1.07 \pm 1.16	0.31 \pm 0.48	n.s.
Urinate in inappropriate places	1.33 \pm 1.29	0.19 \pm 0.40	0.0073
Defecate in inappropriate places	1.33 \pm 1.29	0.25 \pm 0.45	0.014
Destroy furniture and objects	1.36 \pm 1.22	0.53 \pm 0.83	n.s.
Vomit and/or diarrhea	0.33 \pm 0.49	0.13 \pm 0.34	n.s.
Sleep/wake cycle			
Refuse to calm down before sleeping time	0.69 \pm 1.01	0.19 \pm 0.54	n.s.
Wake suddenly in the night	0.88 \pm 1.09	0.13 \pm 0.34	0.024
Wander around the house in the night	0.94 \pm 1.06	0.13 \pm 0.34	0.012
Housetraining			
Urinate and/or defecate wherever in house	0.44 \pm 0.81	0.19 \pm 0.54	n.s.
Urinate and/or defecate in the sleeping area	0.13 \pm 0.34	0.00	n.s.
Urinate and/or defecate after coming home	0.19 \pm 0.40	0.00	n.s.

Table 3. Evolution, over time, of behavioral parameters in puppies adopted from a shelter and treated with DAP (mean values \pm SD; n=16). V1 = 28 days after the beginning of treatment, V2 = 56 days after the beginning of treatment. P<0,05; n.s. = not significant.

Parameters	V1	V2	p value
Socio-environmental reactions			
Wander in the house restlessly	0.62 \pm 1.02	0.13 \pm 0.34	0.022
Pant/tremble, resting in a corner	0.56 \pm 0.89	0.13 \pm 0.34	n.s.
Hide in protected places	0.63 \pm 0.89	0.13 \pm 0.34	n.s.
React to unfamiliar people	1.25 \pm 1.24	0.63 \pm 0.89	n.s.
React to telephone and door ringing	1.56 \pm 1.46	0.44 \pm 0.51	n.s.
React to unfamiliar dogs	1.63 \pm 1.41	0.50 \pm 0.52	0.048
Human-dog interaction			
Interact inappropriately with the owners	1.32 \pm 1.30	0.94 \pm 1.12	n.s.
Play inappropriately with the owners	1.44 \pm 1.36	0.81 \pm 0.98	n.s.
Look continuously for the owners	2.31 \pm 1.08	1.88 \pm 1.09	n.s.
Follow the owners everywhere like a shadow	2.31 \pm 1.08	1.81 \pm 1.11	n.s.
Release urine during exuberant play	1.50 \pm 1.15	0.44 \pm 0.51	0.011
Separation reactions			
Vocalize in the owners' absence	2.30 \pm 1.18	0.67 \pm 0.49	0.0029
Urinate in inappropriate places	2.00 \pm 1.29	0.47 \pm 0.52	0.0067
Defecate in inappropriate places	1.15 \pm 1.14	0.27 \pm 0.46	0.033
Destroy furniture and objects	1.08 \pm 1.32	0.40 \pm 0.51	n.s.
Vomit and/or diarrhea	0.25 \pm 0.45	0.13 \pm 0.35	n.s.
Sleep/wake cycle			
Refuse to calm down before sleeping time	0.69 \pm 1.01	0.31 \pm 0.87	n.s.
Wake suddenly in the night	0.81 \pm 0.83	0.25 \pm 0.58	0.026
Wander around the house in the night	0.81 \pm 0.83	0.25 \pm 0.58	0.026
Housetraining			
Urinate and/or defecate wherever in house	2.13 \pm 1.20	0.44 \pm 0.63	0.0013
Urinate and/or defecate in the sleeping area	0.14 \pm 0.36	0.00	n.s.
Urinate and/or defecate after coming home	1.20 \pm 1.20	0.13 \pm 0.35	0.009

For socio-environmental interactions a decrease in the mean scores between V₁ and V₂ was observed for all the following variables: wander in the house restlessly, pant/tremble, remaining in a corner, hide in protected places, react to telephone and door ring and react to unfamiliar dog. In both adults and puppies a significant improvement in interaction with owners was observed. The analysis of separation reactions revealed a significant decrease (p<0.05) in the mean scores between V1 and V2 for the following variables: urinate in inappropriate places; defecate in inappropriate places, in adult dogs as well as puppies. The behavior of adopted dogs related to the sleep/wake cycle significantly improved after treatment. In contrast, for house-training no significant difference between V₁ and V₂ was reported in adults, whereas for puppies there was a significant decrease (p<0.05) in the mean scores for the following variables: urinate and/or defecate wherever in the house and urinate and/or defecate after coming home. Data regarding the overall assessment in puppies and adults suggested a significant improvement in all the efficacy variables considered in the study (Table 4).

Table 4. Overall assessment of different efficacy parameters over time in dogs adopted from a shelter and treated with DAP (mean values \pm SD; n=32). V1 = 28 days after the beginning of treatment, V2 = 56 days after the beginning of treatment. $P < 0,05$, n.s. = not significant.

Parameters	V1	V2	p value
Socio-environmental reactions	0.59 \pm 0.50	1.31 \pm 0.78	<0.0001
Human-dog interaction	0.65 \pm 0.61	1.45 \pm 0.57	<0.0001
Separation reactions	0.64 \pm 0.68	1.58 \pm 0.62	<0.0001
Sleep/wake cycle	0.31 \pm 0.54	0.84 \pm 0.88	0.011
House training	0.28 \pm 0.46	1.00 \pm 1.04	0.0043

The analysis of owners' degree of satisfaction at V2 visit showed that DAP treatment was considered successful by 84.4% of the owners against 53.1% in V1, while the percentage of unsatisfied owners 46.9% in V1 decreased to 15.6% in V2 ($p < 0.0001$) (see Fig. 1).

Discussion

The results of the current study suggest a significant positive effect of treatment with DAP on the behavior of dogs newly adopted from the shelter. The authors suggest that DAP might be interpreted as a valid adjuvant in the adaptation phase, since there are no negative effects on adoption and even a positive one: a reduction in the rehoming rate which is usually around 10% (Unpublished data). Of course it might be objected that the data reported could very well have been the result of habituation to the new environment, desensitization and positive interactions with the owners if the latter actively rewarded the dogs (with petting, praising or use of food or toy/play rewards) or, in the case of the puppies, the result of normal ontogeny. A similar conclusion was reached by Schneider et al. in 2009, where 11 dogs were available for analysis, using meantime as a treatment of canine compulsive disorders with behavioral modification, at different doses, without placebo. This might be an alternative explanation, but different authors referred that the number of dogs adopted from shelters showing behavioral problems in the first phase after adoption is very high and the primary reason for returning a dog to the shelter has been reported to be a behavior problem (Wells & Hepper, 2000; Mondelli et al., 2004; Shore, 2005; Luescher & Medlock, 2009). Furthermore in the first period of the rehoming the dog can create an attachment bond with the owner, who can become a secure base for the dog (Mariti et al., 2013).

Our data show a significant improvement in some behaviors related to socio-environmental reactions. In particular, positive effects on behavioral signs of fear, such as hiding and reacting defensively to unfamiliar dogs, were observed. Canine anxieties and phobias, especially separation anxiety and noise aversions, are common behavioral disorders in pets (Sherman et al., 2008). Fear related behaviors might be an adaptive response, especially when the stimuli are potentially indicative of impending threat or danger; fear is actually an adaptive emotional response and important for the survival of the individual. However, when the danger or threat is a perceived one, fear can become maladaptive, causing a phobic state with serious behavioral signs (Heath & Bowen, 2003). In 2003 a research team from the University of Lincoln reported the results of an open uncontrolled trial to assess the product's efficacy in treating the fear of fireworks in pet dogs in the absence of any specific behavioral modification (Sheppard & Mills, 2003); in the abovementioned study the DAP diffuser

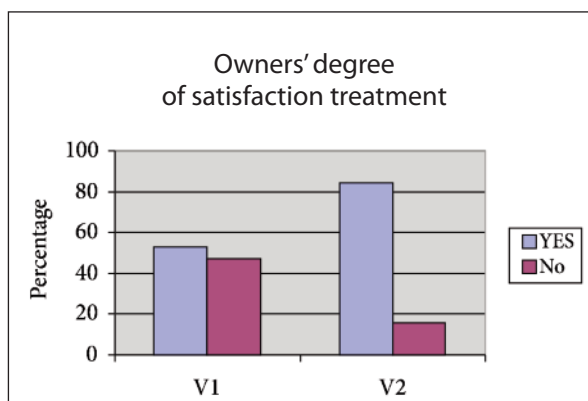


Fig. 1. Evolution of positive response according to time.

was positioned as close as possible to the dog's usual resting place at least two weeks prior to the predicted firework event and recordings of behavioral signs associated with a fear of fireworks were recorded both as a baseline measure, at the start of the trial, and a final assessment, after three to five weeks of treatment. The results published (Sheppard & Mills, 2003) suggested a reduction in the measures of overall severity of the behavioral signs after treatment with DAP in comparison with baseline responses, and a significant reduction in measures of the frequency of nine of the fourteen most common behavioral signs displayed by dogs. However, no studies investigating the effects of DAP on general activity behaviors in dogs adopted from a shelter had ever been published to date. Moreover, the analysis of the effects of environmental management strategies was not considered, since no instructions were given to the new owners to prevent behavior problems. The present study focuses on the effects of DAP on the behaviors shown by the dogs in their two months following adoption, and not on the treatment of behavioral problems in sheltered dogs after adoption. The benefits as a short-term treatment strategy are consistent with the enhanced quality of these hideouts and the consequent success of the animal's natural coping strategy. A similar effect might explain the positive results in the current study. The efficacy of pheromones for the treatment of fireworks, combined with desensitization and counter-conditioning programs, has been investigated recently (Levine et al., 2007, 2008).

But the current study suggests that in dogs treated with DAP significant modifications in human-dog interaction efficacy parameters occurred. In particular, pheromones were effective in reducing excessive contact-seeking behaviors, especially in adults. This was assessed by a significant decrease in the two following parameters: "Looking continuously for the owners", and "Following the owners everywhere like a shadow". Studies investigating the efficacy of DAP for the treatment of separation-related disorders had already demonstrated that pheromonotherapy is able to reduce undesirable behaviors such as hyper-attachment to the owners (Gaultier et al., 2005). In our study, the efficacy was observed particularly in adults.

Adoption is widely recognized as being stressful for all sheltered dogs, but especially puppies. In fact the puppy's maternal bond is sometimes traumatically broken in rescue animals or, more simply, the youngest never experience a mother-puppyhood relationship. The first two months of life are crucial since the puppy is moved to his/her new social and physical environment with new rules (Serpell & Jagoe, 1995); for this reason the puppies might show more difficulties in the coping process. The importance of providing behavioral advice to puppy owners has been demonstrated by Gazzano et al. (2008).

As in many dogs over attachment is strongly correlated with signs of distress in dogs suffering from separation-related behavioral problems (Appleby & Pluijmakers, 2003), we may conclude that pheromone treatment improves dogs' welfare. Furthermore pre-adoption counseling was found to be not effective on the prevention of separation anxiety in newly adopted shelter dogs (Herron et al., 2014). Interestingly, DAP reduced vocalization in puppies during their owners' absence. Gaultier et al. (2008) investigated the value of DAP in reducing signs of distress in puppies adopted from a pet shop, especially when socially isolated and/or during the night. The Authors found that puppies with a DAP collar stopped nuisance activities before the third night of treatment. In young dogs, vocalization is considered as the main sign of social stress (Hetts et al., 1992). This behavior can become annoying for neighbors and owners. Reducing the tendency to vocalize during periods of social isolation is a way of improving the integration of puppies into the family and neighborhood. Furthermore, our results are consistent with those of Taylor and Mills (2007b), who found that DAP prevents prolonged disturbance at night at the adoption stage in puppies. According to the authors (Taylor & Mills, 2007b), pheromone treatment is useful in helping puppies to settle into their new home by reducing the frequency of disturbed nights and maintaining the puppy's emotional stability.

House training efficacy parameters showed that treatment was able to reduce inappropriate elimination, especially in puppies; this action is generally associated with a high degree of owner compliance. Disturbance and house-soiling during the night in recently adopted puppies are behaviors

that appear to reflect a mismatch between the developmental status of the puppy and its new environment. It has been suggested that DAP may help the puppy to settle into the new home by continuing the provision of the maternal appeasing pheromone, according to a double-blind, placebo-controlled trial with DAP conducted in 60 pedigree puppies as they entered their new home (Taylor & Mills, 2007b). The effects of temporary exposure to synthetic dog appeasing pheromone on levels of arousal in puppy classes were positively reported (Graham et al, 2007) as well as on anxiety and fear in puppies during training and long-term socialization (Denenberg & Landsberg, 2008).

As regards the results of the sleep/wake cycle, overall assessment and owners' degree of satisfaction, they suggest a positive response to DAP. Our data are consistent with those of Gaultier et al. (2008), who found a significant positive effect on dogs' behavior at night.

An assessment of the owner's level of satisfaction showed that owners were more satisfied at V2 than at V1. The satisfaction of the owners increased in over the time after adoption suggesting a significant improvement in terms of long-term management of adopted dogs.

According to Stephen & Ledger (2003), owners are considered reliable observers of their own dog's behavior and the assessment of owners' perception of behaviors has been used in a number of clinical studies concerning the effects of DAP (Sheppard & Mills, 2003; Mills et al., 2006).

In addition, one limitation that should be discussed regarding the present trial is that no initial assessment providing baseline behavioral data was performed before the treatment phase with the DAP diffuser was implemented. This would have helped to assess the exact improvements due to the positive effects of DAP, and to differentiate them from the results of habituation to the new environment, desensitization or even positive interactions with the owners. In the particular case of puppies, the observed improvement can also partly be due to normal development, in addition to the effect of DAP.

Conclusions

The precise mechanism of action of most pheromones is still unknown, but in mammalian species they might induce some modifications in both the limbic system and the hypothalamus, thus affecting emotional state and behavioral response in the animal, as reception of pheromones creates an input and sets off internal and physiologic reactions (Pageat & Gaultier, 2003a). If they are prescribed, provided and used in the right way, pheromones show a significant effect and can help the new owner to prevent and/or solve many problems in a totally safe way. "Pheromonotherapy" is the word suggested to describe the use of pheromones applied to behavioral disorders; because pets are living in a world full of odors, this particular clinical approach seems interesting and is considered acceptable and pleasant by owners (Pageat & Gaultier, 2003a). Sherman et al, (2008) advised pheromone use in the new modalities of treatment of canine anxieties and phobias in their updates of these conditions.

The preliminary results of this study show that the use of pheromones when animals enter the new environment might improve their adaptability throughout the different adoption steps. As a consequence this could ultimately help to decrease the return rate to the shelter. Depending on their characteristics and properties, it seems that the range of action of pheromones could actually reduce stress and improve welfare. Their efficacy has been generally assessed in some specific behavioral problems, even if highly criticized by Frank et al. (2010), but no study has yet been published on the specific issue of the probable positive effects in the adoption process. It has to be highlighted that without invasive neurophysiological analysis it might be impossible to detect some effects of DAP on the central nervous system, but this is not acceptable in pets, as it requires the sacrifice of the dogs.

Nevertheless, the study was performed as a prospective open label experimental design comparing the evolution over time of an experimental group of 32 dogs adopted from a shelter and treated with DAP (Adaptil[®] Ceva Vetem S.p.A). Double-blind, controlled clinical studies, which would es-

establish with greater certainty the relative efficacy of this product compared to a placebo, could not be applied without a strong reaction about animal rights and experiments on shelter animals. Further investigations would be required to test these potential benefits; since it would be extremely difficult in the context of the Italian rescue associations to apply a placebo-controlled study, a survey for evaluating the support offered by behavioral advice with or without DAP support once the animal has been re-homed, might be the next step for the authors.

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Meccanismi adattativi in cani provenienti da canili: valutazione comportamentale dell'uso di un analogo sintetico del feromone di appagamento canino

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Sintesi

L'adattamento ad un nuovo ambiente sociale può rappresentare una prova molto ardua per un cane da canile, per la difficoltà di fare a condizioni di vita che non gli sono familiari.

Il processo di adattamento è correlato, in termini sia scientifici sia logici, con il concetto di stress e benessere e da questi fattori dipende il successo o meno dell'adozione del cane proveniente da un canile.

È stato dimostrato da numerose ricerche che un analogo sintetico del feromone di appagamento del cane (DAP) ha un effetto calmante sui cuccioli ed i cani adulti in una vasta gamma di situazioni strassanti.

Lo scopo della presente ricerca è stato quello di valutare gli effetti del DAP (Adaptil® Ceva Vetem S.p.A.) su cani adottati da canili rifugio.

Per la presente ricerca sono stati utilizzati 32 cani, 16 cuccioli e 16 cani adulti, provenienti dal canile rifugio ENPA di Torino. All'adozione del cane, i nuovi proprietari ricevevano un diffusore con DAP che avrebbero sistemato nell'appartamento e mantenuto attivo per 56 giorni.

I risultati hanno rivelato che il trattamento col DAP provoca una riduzione statisticamente significativa nei cani adulti dei comportamenti di "vagare senza sosta in casa" ($p=0,022$) e "nascondersi spaventati in angoli protetti" ($p=0,033$), mentre nei cuccioli diminuisce, in modo significativo, la "reattività verso cani sconosciuti" ($p=0,048$) e "vagare senza sosta in casa" ($p=0,022$).

Sia nei cani adulti, sia nei cuccioli è stato osservato un significativo miglioramento nelle interazione con i proprietari. In particolare, nei cani adulti, "cercare continuamente il proprietario" e "seguire il proprietario ovunque come un'ombra" si sono ridotti significativamente ($p=0,0012$ e $0,0016$ rispettivamente).

Le reazioni alla separazione mostrano un significativo decremento ($p<0,05$) e nei cuccioli la tendenza a vocalizzare in assenza del proprietario si è ridotta significativamente ($p=0,0029$).

Sia i cani adulti sia i cuccioli mostrarono una diminuita tendenza a svegliarsi improvvisamente di notte ($p=0,024$ e $p=0,026$ rispettivamente) e a vagare in casa ($p=0,012$ e $p=0,026$ rispettivamente).

Per quanto riguarda l'educazione ad una corretta eliminazione, nei cani adulti non è stata evidenziata nessuna differenza significativa, mentre nei cuccioli si è riscontrato un significativo decremento ($p<0,05$) nei valori medi relativi al comportamento di urinare/o defecare in casa o dopo il rientro a casa.

L'analisi del livello di soddisfazione dei proprietari alla visita finale, ha mostrato che il trattamento con il DAP è stato considerato positivo dall'84,4% dei proprietari.

Questi risultati suggeriscono che il DAP potrebbe migliorare le capacità di adattamento dei cani provenienti dai canili nelle prime settimane dopo l'adozione e può rivelarsi un utile strumento nel ridurre lo stress post-adozione.