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Home or boarding school for the weekend: which is best for future guide dogs involved in an ongoing training program?

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Abstract: The impact of the housing management approach for future guide dogs during the training program on dogs' welfare is still unknown. During the training period, dogs either go back with their foster family every weekend, or they stay at school. The aim of this study was to compare these two management styles on animals' welfare and performance. Behavioral and physiological parameters were assessed on eighteen dogs over a period of 3 weeks; 9 returned to their foster families on weekends (FF group), and 9 remained at school (Sc group). Results showed that dogs staying at school expressed less stress behavior at rest (GLMM; DF=1; F =10.11; p=0.0018). A visual analogue scale completed by the dog trainer indicated that they were more focused during training sessions (GLMM; DF=1; F=5.42; p=0.0326). Also, dogs were well accustomed to life in the school kennel, with the neutrophil/lymphocyte ratio and levels of serotonin, prolactin, and oxytocin in line with normal rates in both groups. These results suggest that school environment has no negative impact on the dogs and avoiding repeated separations with the foster family could reduce stress and increase their welfare. This should prompt consideration of the housing environment of future guide dogs.

Key Words: animal welfare; assistance dogs; emotional balance; foster family; guide dog school; performance.

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Introduction

Consideration of animal welfare is increasing. Furthermore, since research has shown that welfare could be linked to animal performance, interest in this discipline has grown (Rooney et al., 2009; Waiblinger et al., 2006). Guide dogs are essential for people who choose this type of assistance over a white cane (Mariti et al., 2014), but breeding and training future guide dogs is challenging for institutions. It is expensive (25 000 € per dog) (Mengoli et al., 2017), and many dogs are eliminated from the program due to undesirable traits, such as fearfulness, aggressiveness, or distractibility (Arata et al., 2010; Goddard & Beilharz, 1982). Studies demonstrated that dogs'

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early life experiences can shape their adult behavior. For example, Tiira & Lohi (2015) showed that a good socialization between the age of eight and twelve weeks reduce fearfulness in adult dogs. It is fundamental to understand why dogs develop these kinds of behaviors and if it is possible to reduce the failure rate by adapting the development program of the dogs, with a better understanding and consideration of the animal's needs. Unfortunately, to the best we know, there is a shortage of information about this topic in the literature.

Foster families care for dogs from weaning to the beginning of training, when the dogs are approximately 1 or 2 years old. Since they raise the dogs and care for them during a decisive phase of their development (habituation and socialization), their effects on the dogs are important and could have an impact on the dogs' development and their success as guide dogs (Koda, 2001; Menuge et al., 2021). During the training period, which lasts between 6 and 8 months (Alterisio et al., 2019; Arata et al., 2010), dogs' routines could be different depending on the specific guide dog organization. In some cases, dogs stay at the school for the whole training period (Chur-Hansen et al., 2015), or they return to their foster families every night or every weekend (Dalibard, 2009). These different management types could influence animal stress, welfare, and performance. Stay at school all the time could allow dogs to keep a same routine and stay focused easily. Nevertheless, they would also loose connection with a family environment. In the other options dogs keep a link with family environment but it also could induce separation-related behaviors in dogs. A study evaluated the stress and welfare of dogs who go back with their puppy raisers on weekends. It was observed that on Mondays, which is the day of separation with the foster family, dogs were more stressed and less receptive to new learning than on the other days of the week (Menuge et al., 2021). The objective of this research was to go further these results, and to evaluate if it is better for the dogs to go back with their foster family every weekend or to stay at school, by comparing these two types of management styles. We assumed that it would be more beneficial for dogs to stay at the school during the entire training period to avoid exposure to weekly environmental changes. Indicators of stress, welfare and the performance of dogs participating in a program in which they either returned with their puppy raiser on weekends or stayed at school were assessed to evaluate which organization type was associated with better outcomes in the dogs.

Materials and methods

This project was approved by the French Ministry of Research (APAFIS#24626-202003111326352 2 v2).

Animals

Eighteen potential guide dogs (5 females and 13 males, all neutered) involved in an ongoing training program at the Frederic Gaillanne Foundation (FGF, L'Isle-sur-la-Sorgue, France) participated in this study, in two different groups. The first group (9 dogs–2 females and 7 males) returned to their foster family from Friday evening to Monday morning (FF group), and the second group (9 dogs–3 females and 6 males) stayed at school throughout the training period (Sc group). The guide dogs in training were Labernese (a crossbreed between a Bernese Mountain dog and Labrador), St–Pierre (at least third-generation Labernese) and Labrador. The FF group was composed of 2 Labernese, 6 St–Pierre and 1 Labrador, and the Sc group was composed of 3 Labernese, 5 St–Pierre and 1 Labrador. Before the beginning of the training period, all the dogs lived with their foster families. They were all between 12 and 24 months old at the time of the study and began their training program at school 6 weeks before the start of the experiment. Concerning housing conditions at school, 3 dogs were placed per box. Each box was made up of an interior part of $8m^2$ and an exterior part of $7m^2$, leaving them a total space of $15m^2$. The facility

also had 3 outdoor parks of 100m^2 each to which the dogs had regular access during the day. A bucket of water was available to them ad libitum. They were fed twice a day (morning and evening), and the guide dog organization supplied kibble to the puppy raisers. The school routines during the week were the same for the 2 groups–all dogs followed a preestablish educational program–but their routines were different during weekends. Dogs in the FF group were occupied differently depending on the routines of their foster families. For dogs in the Sc group, an employee of the FGF came 3 × 2 hours per day on Saturday and Sunday to feed, walk and play with the dogs (no training sessions were conducted on weekends).

Experimental design

Data from the FF group was obtained from a recent paper previously published (Menuge et al., 2021). For this group, study was undertaken from 16th of November 2020 to 4th of December 2020, 6 weeks after the beginning of the training period. The same study design was used for the Sc group to ensure comparability, from 5th of July 2021 to 23rd of July 2021, also 6 weeks after the beginning of the training period. During these 6 weeks, dogs were already implied in the housing management of their corresponding groups: Dogs from FF group came back to their foster family on weekends during this period, and dogs from Sc group stayed at school. For the trial, behavioral and physiological parameters were collected during 3 consecutive weeks. The same design as Menuge et al. (2021) was used for the present study, and the parameters were collected on the same hours:

Behaviors: Dog behaviors related to stress, or a positive mental state were reported during a period of rest (Table 1). Dogs were videotaped for 45 min on Mondays, Wednesdays, and Fridays in their kennels between 12:00 p.m. and 1:00 p.m. All behaviors were analyzed using continuous sampling by 2 independent observers using Boris software (Friard & Gamba, 2016).

Table 1. Dogs' behaviors related to stress or a positive mental state assessed during periods of rest on Mondays, Wednesday, and Friday during 45 minutes.

Behavior	Description	References
Locomotion (SE)	Walking, pacing, or running around without exploring the environment or playing.	(Fallani et al., 2007)
Panting (SE)	Rapid shallow breathing (mouth open).	(Beerda et al., 1998)
Social play (SE)	Any vigorous behavior performed when interacting with another dog, including running, jumping and active physical contact.	(Fallani et al., 2007)
Relaxation behavior (SE)	Lying down with the head on ground without any obvious orientation towards the physical or social environment.	(Palestrini et al., 2017)
Head movement (PE)	Characteristic movement sidewards and downwards.	(Schilder & Van Der Borg, 2004)
Withdrawal (PE)	Avoiding interaction with another dog by either moving away, very clearly turning away or looking away.	(Palestrini et al., 2017)

Barking (PE)	"Rough" sound often repeated in quick succession.	(Tod et al., 2005)
Whining (PE)	Throaty, low-frequency sound.	(Beerda et al., 1998; Stellato et al., 2016)
Yawning (PE)	Wide opening of mouth.	(Beerda et al., 1998; Palestrini et al., 2017)
Lip licking (PE)	Portion of the tongue is shown and moved along the upper lip.	(Beerda et al., 1998; Palestrini et al., 2017)
Total stress behaviors	Total of behaviors expressed as point events.	(Palestrini et al., 2017)

SE = State event (duration in second); PE = Point event (frequency).

Performance: A visual analogue scale developed by the experimenter was completed by the professional dog trainer after each dog's morning training session; the dogs received scores ranging from 0 to 10 for eight criteria (focus, stress, agitation, attentiveness, effectiveness, distraction, posture, and a general note from the education session). The same dog trainer completed the visual analogue scale for both groups.

Salivary cortisol levels: Saliva was collected by using a Salivette® system (Salivette®, Sarstedt, Numbrecht, Germany), which is specifically targeted for the determination of cortisol in saliva. The study protocol was repeated on Monday and Friday morning to control for the natural influence of the circadian rhythm on cortisol secretion at the time of the experiment. According to the literature, it takes twenty minutes for cortisol to be secreted in saliva (Dreschel & Granger, 2009; Hernandez et al., 2014). Therefore, sampling was carried out according to this timing: twenty minutes after the time of separation (or at the corresponding hour for non-separation day or group, i.e., between 8:15 a.m. and 9:00 a.m.), after the morning training session (between 9:00 a.m. and 12:00 p.m., and after a period of rest (between 1:30 p.m. and 2:00 p.m.). To prevent sample contamination, the dogs were not allowed to eat for one hour prior to sampling. If the presence of blood was visually observed in the saliva samples, the samples were discarded to prevent contamination with blood cortisol. Immediately after the samples were collected, they were stored at 4°C until they were centrifuged (2 min, 1000 rpm, 4°C). After centrifugation, the samples were frozen at -20°C until the day of the cortisol assay, which was performed using a Salimetrics® Cortisol Enzyme Immunoassay Kit (Kiel, Germany) according to the manufacturer's guidelines.

Blood sampling: Up to 15 ml of blood was collected from the cephalic vein on the forelimb using a 21 G needle to assess the levels of free oxytocin, prolactin, and serotonin and the neutrophil/lymphocyte (N/L) ratio; blood was collected by a veterinarian for welfare monitoring on Wednesday mornings, between 9:00 a.m. and 11:00 a.m. For hormonal analyses, blood was collected into prechilled EDTA-aprotinin tubes (BD® tubes, Elvetec, Pusignan, France) for plasma oxytocin determination and into tubes with a gel separator (Vacuette®, Greiner Bio–One, Alcyon, Paris, France) for serum prolactin and serotonin de-termination. The tubes were immediately stored in an ice box, where they remained at 4°C until centrifugation (1800 g, 12 min at 4°C). Plasma or serum was recovered and stored at -20°C until analysis. The free plasma oxytocin concentration was determined with the commercially available Oxytocin ELISA kit from Cayman Chemical (Arbor Inn, MA, USA) after solid-phase extraction in C18 columns (Hypersep 1 g, Thermo Fisher Scientific, Illkirch, France) following the procedure described in Oliva et al. (2019). Serum prolactin was assayed with a prolactin (canine) ELISA kit (Demeditec, Kiel, Germany) following the manufacturer's instructions. Serum serotonin was assayed with the commercially

available Serotonin ELISA kit from Enzo Life Science (Villeurbanne, France), which has been previously validated in dogs (Chabaud et al., 2018). For the N/L ratio, a complete blood cell count was carried out with an automated LaserCyte from IDEXX (Westbrook, MA, USA) on the day of blood sampling using EDTA tubes provided by the manufacturer (IDEXX VetCollect Tubes). The total white blood cell count, including neutrophil and lymphocyte counts, was recorded, and the N/L ratio was calculated from these data.

Weight: Dogs were weighed at the same time each day, on the morning.

Statistical analysis

Data analysis was performed with SAS 9.4 software (Copyright (c) 2002-2012 by SAS Institute Inc., Cary, NC, USA). The significance threshold was fixed at 5%.

For all parameters, the effect of the educational regime (FF vs. Sc group) was studied for the mean of the 3 weeks and for each individual week of testing, with the inclusion of group, week and the group*week interaction in the models. For salivary cortisol levels, the effect of day was also investigated.

Given that the data were repeated on the same dogs, the dog was assumed to be a random effect using mixed models. For continuous variables (salivary cortisol, prolactin, serotonin, and oxytocin levels; the N/L ratio; state events, and performance scores), general linear mixed models (GLMMs) were applied. The condition of normality of residuals was checked graphically. When this assumption was not met, the GLMM was performed on the transformed data obtained from a Box-Cox transformation.

For discrete data (point events), mixed Poisson regression was applied when the data were appropriately dispersed according to the Pearson chi-square/DF value. When over-dispersion was detected, a negative binomial mixed model was preferred because this sort of model allows a dispersion scale parameter to account for overdispersion problems. For behaviors at rest, the square of the Pearson correlation coefficient was computed to evaluate the interobserver association between the two independent video readers. Few dogs were observed whining, barking, panting, playing, or withdrawing. It was impossible to analyze these behaviors with methods for quantitative data. These data were transformed into binary variables and mixed logistic regression analyses were achieved for whining and barking. Panting, playing, and withdrawal remained not analyzable, even after the binary transformation.

For each model, the best covariance structure was chosen by minimizing the corrected Akaike information criterion (cAIC) and Bayesian information criterion (BIC), and the Tukey-Kramer adjustment was employed for multiple comparisons. When possible, the model was simplified by removing nonsignificant effects step by step as long as the cAIC and BIC criteria decreased.

Results

Behaviors at rest

The interobserver reliability indicated strong associations for all the parameters (>90%). Therefore, the average of the data gathered by the two observers was applied for the remaining analysis.

The Sc group spent more time performing “relaxation behaviors” (GLMM; Num DF=1; Den DF=17.8; F=12.00; p=0.0028), expressed less “yawning” (GLMM; Num DF=1; Den DF=68; F=8.10; p=0.0058), less “head movement” (GLMM; Num DF=1; Den DF=68; F=6.66; p=0.0120), and less “total stress behaviors” (GLMM; Num DF=1; Den DF=132; F =10.11; p=0.0018) than the FF group (Table 2). For the other parameters, no significant differences were found.

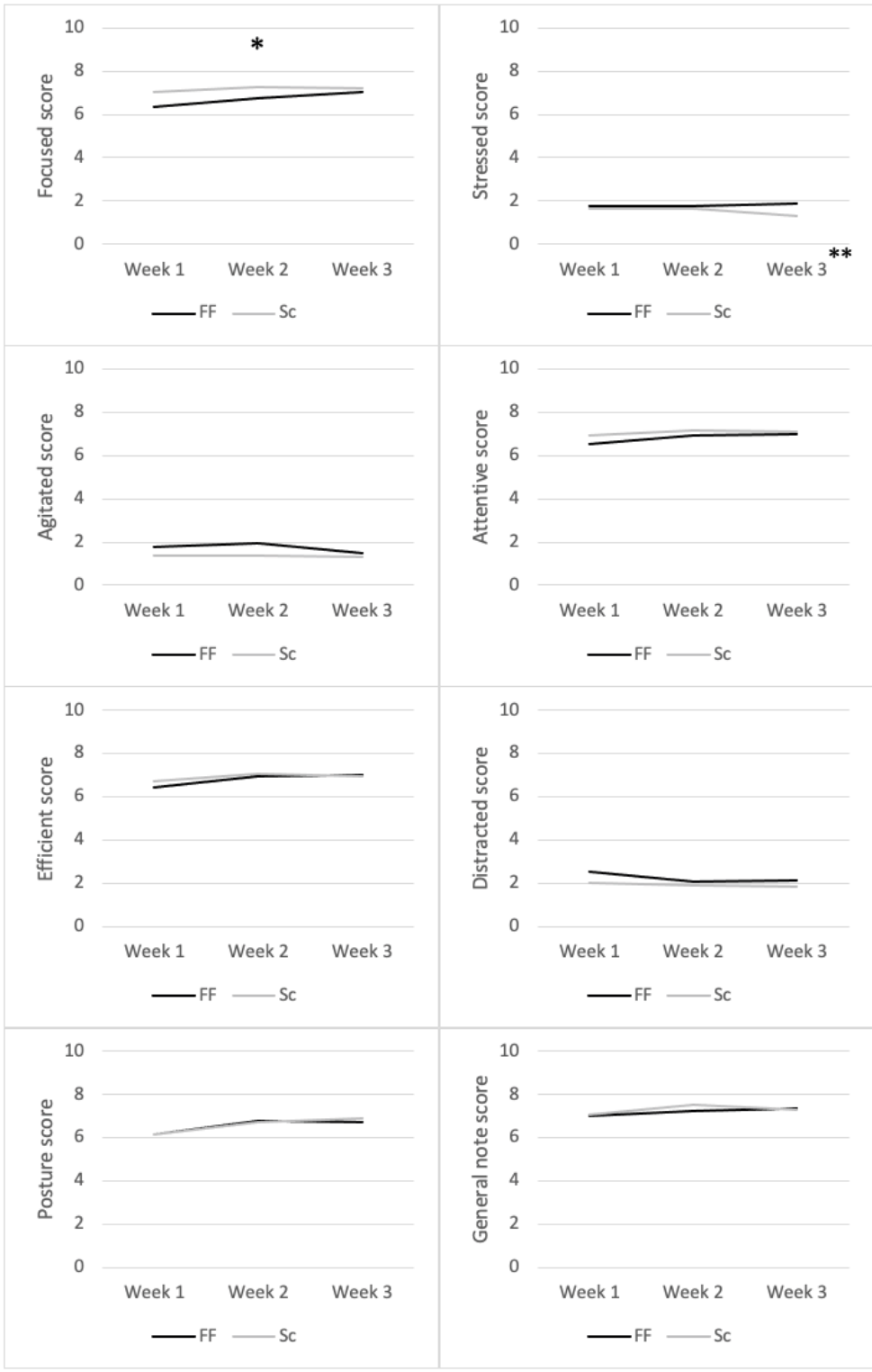

Figure 1. Performance scored by the dog trainer for each criteria. * = $p < 0.05$; ** = $p < 0.01$. FF = Foster Family group; Sc = School group.
* above the curve = presence of a significant result on the general linear mixed model (mean of the three weeks).
* next to the y-axis legend = multiple comparison with significant result.

Table 2. Mean of behaviors expressed per observed day during periods of rest over the three weeks of testing.

N = 81	Foster Family group	School group
mean ± standard error or n (%)		
Locomotion (SE)	13.48 ± 2.05	5.22 ± 0.79
Relaxation Behavior (SE)	2313.34 ± 40.85[a]	2582.79 ± 18.27[b]
Head movement (PE)	23.34 ± 2.06[a]	9.23 ± 1.57[b]
Barking*	15 (18.52)	7 (8.64)
Whining*	18 (22.22)	2 (2.47)
Yawning (PE)	1.14 ± 0.18[a]	0.17 ± 0.06[b]
Lip licking (PE)	3.42 ± 0.53	2.40 ± 0.46
Total stress behavior (PE)	39.03 ± 4.93[a]	12.62 ± 2.02[b]

SE = State event (duration in second); PE = Point event (frequency).
* Variables expressed in binary data (n = number of individuals having exhibited the behavior all observations considered).
[a vs b] Significantly different at a statistical level.

Performance

A group effect was detected for "focused" (GLMM; Num DF=1; Den DF=16.9; F=5.42; p=0.0326) and only for the 3rd week of testing for "stressed" (Tukey-Kramer; DF=21.4; t=2.96; p=0.0074). The Sc group was more focused and less stressed than the FF group. No significant differences were detected for the other criteria (Figure 1).

Salivary cortisol levels

Generally, Sc group secreted more salivary cortisol than FF group (GLMM; Num DF=1; Den DF=16.1; F=6.72; p=0.0196). Moreover, a significant difference was observed between Monday and Friday in the FF group (GLMM; Num DF=1; Den DF=136; F=8.45; p=0.0043) but not in the Sc group (GLMM; Num DF=1; Den DF=135; F=1.57; p=0.2129) (Figure 2).

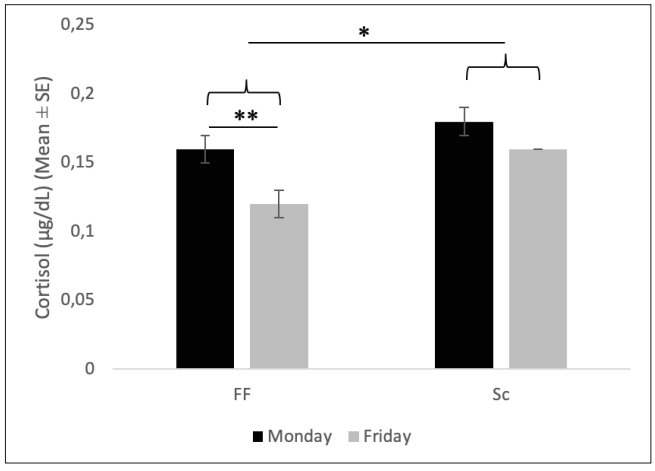

Figure 2. Mean salivary cortisol concentration in the foster family group (FF) and school group (Sc) on Monday and Friday of the 3 weeks of testing. * = $p < 0.05$; ** = $p < 0.01$.

Blood analyses

For prolactin levels, atypical values were observed for two dogs for the 3 weeks of testing (one dog from the FF group and one from the Sc group). These dogs were not included in the statistical analysis for this parameter. Four other samples in the Sc group were below the detection limit of the kit (0.4 ng/ml), suggesting very low or no prolactin secretion in these dogs (one dog over the three weeks of testing and one dog during the second week). An extrapolation of the standard curve using the plate reader software (BioTek Gen5 Data Analysis Software) was performed so that a concentration of prolactin of 0.091 ng/ml was detected for one dog, but not for the others, suggesting the absence or lower levels of prolactin. According to the canine prolactin ELISA kit, normal values are found between non-detectable and 21 ng/ml. Therefore, we replaced these data with 0 to be considered in the statistical analysis.

Although not statistically significant, the serotonin concentrations had a non-significant tendency to be higher in the Sc group compared to the FF group on the first week of testing (Tukey–Kramer; DF=35.2; t=-1.82; p=0.0780) (Table 3). Groups were not different at a statistical level for the concentration of oxytocin (GLMM; Num DF=1; Den DF=16.1; F=2.76; p=0.1162), prolactin (GLMM; Num DF=1; Den DF=14; F=1.62; p=0.2237) or N/L ratio (GLMM; Num DF=1; Den DF=16; F= 2.96; p=0.1044).

Table 3. Mean plasma oxytocin, serum serotonin, and serum prolactin concentrations and neutrophil/lymphocyte ratios (N/L ratios) in dogs in the foster family group (FF group) and school group (Sc group) of the 3 weeks of testing.

Mean ± standard error	**FF group**	**Sc group**
Serotonin (ng/ml)	742.08 ± 41.32 *(n = 27)*	751.43 ± 20.89 *(n = 27)*
Oxytocin (pg/ml)	24.24 ± 1.25 *(n = 27)*	20.08 ± 1.10 *(n = 27)*
Prolactin (ng/ml)	5.57 ± 1.07 *(n = 24)*	3.30 ± 0.52 *(n = 24)*
N/L ratio	2.14 ± 0.50 *(n = 27)*	1.35 ± 0.12 *(n = 27)*

Weight

No significant difference in weight was observed between the Sc (32.61 ± 0.38 kg) and FF (33.55 ± 0.48) groups (GLMM; Num DF=1; Den DF=16; F=0.23; p=0.6347).

Discussion

This project intended to evaluate the different effects between two housing management approaches on the welfare and the performance of potential guide dogs involved in an ongoing training program.

First, behaviors were assessed three times a week during periods of rest to evaluate the capacity of the dogs to relax when they were not solicited by the dog trainer. The comparison between these two groups showed that the dogs in the Sc group expressed fewer stress-related behaviors in general and spent more time in relaxed positions than those in the FF group. These results suggest

that it might be easier for dogs who stay at school all the time to understand the routine of the training sessions and to appropriately adapt their behavior in this context. In fact, when dogs go back to their foster families from Fri-day evening to Monday morning, they seemed to have to readapt themselves to the training environment every week (Menuge et al., 2021), which did not seem to be the case for the dogs that stayed at school.

Regarding the performance of the dogs from the dog trainer's point of view, the dogs in the Sc group were more focused than the dogs in the FF group. This could be the result of a more stable environment, in which dogs could have better anticipated the routines of the training sessions. Additionally, the dogs in the Sc group were less stressed on the third week of the study period than the dogs in the FF group, supporting this hypothesis. Even though differences in the other criteria were nonsignificant, it is interesting to highlight that according to the descriptive data, the Sc group was probably more emotionally balanced than the FF group. In fact, they tended to have lower agitation and distraction scores and higher attentive, efficient, and general note scores.

Salivary cortisol detection allowed us to evaluate the activation of the hypothalamus-pituitary-adrenal axis at key moments in a non-invasive way. For all the samples combined, the cortisol concentration was higher in the Sc group than in the FF group, with a medium-sized effect ($d = 0.375$) (Valentine & Cooper, 2003) and with results in both groups within the normal values described in the literature (Cobb et al., 2016; Colussi et al., 2018). This data wonders on the presence of a real im-pact of this difference on the welfare of these dogs. Regarding the comparison between days, a variation in salivary cortisol concentration was observed in the FF group, with an increase in secretion on Monday (day of separation) compared to Friday (basal value), while data remained stable for the Sc group. Therefore, the stress of the dogs could be tempered by staying at school during weekends.

The neutrophil/lymphocyte (N/L) ratio was evaluated to assess the level of long-term stress (Swan & Hickman, 2014). It was measured each week on Wednesday. Groups were not statistically different, and the results did not show values representative of the presence of chronic stress. Indeed, values were lower than those found by Hodgson et al. (2018) in healthy dogs in both groups. Furthermore, although the difference was nonsignificant, the N/L ratio in the Sc group was lower than that in the FF group (1.35 vs. 2.14, respectively).

Neurohormonal parameters reflecting the emotional states of the dogs were also investigated on Wednesdays. Levels of oxytocin, prolactin and serotonin were assessed. Considering the mean of the three weeks for each group, no significant difference was found, with results in line with published works (Gutiérrez et al., 2019; Mengoli et al., 2021). However, during the first week, the dogs in the Sc group tended to secrete more serotonin than the dogs in the FF group. An increased level of serotonin can be induced by social exposure to other dogs, which has been shown to have positive effects on canine welfare (Alberghina et al., 2019; Rooney et al., 2009). However, this difference probably only has a small effect on the animal behavior and welfare, given the weak effect size ($d = 0.05$). Furthermore, groups were analyzed at two different periods of the year, and we cannot exclude the fact that it could have an impact on these results. However, dogs spent most of their time inside the building, where the temperature was controlled, limiting the impact of external temperature on dogs' hormonal secretion. It is valuable to mention that dogs in guide dog school are subjected to many stimulants, differentiating them from shelter dogs. The latter live in a challenging environment, even if human interaction has a positive impact on their welfare (Coppola et al., 2006; Hennessy et al., 2020).

Finally, the weight of the dogs unveiled no significant difference between groups. Therefore, this result suggest that the housing management approach did not have an impact on the weight of the dogs. All dogs were carefully and frequently checked by staff at the guide dog school, and their weights were balanced and adjusted according to the needs of each dog.

Conclusion

Staying at school during the entire training period does not seem to be detrimental for guide dogs, confirming the observations of a previous study (Menuge et al., 2021), and avoiding repeated separations could reduce stress and therefore increase the welfare of these dogs. Further research would be interesting with a larger number of dogs to compare deeply both housing management styles.

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Casa o scuola per il fine settimana: quale soluzione è la migliore per i futuri cani guida coinvolti in un programma di addestramento continuo?

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Sintesi

L'impatto sul benessere della sistemazione dei futuri cani guida durante il programma di addestramento è ancora sconosciuto. Durante il periodo di addestramento, i cani tornano con la famiglia adottiva ogni fine settimana o rimangono a scuola. Lo scopo di questo studio è stato quello di confrontare questi due stili di gestione sul benessere e le prestazioni degli animali. I parametri comportamentali e fisiologici sono stati valutati su diciotto cani per un periodo di 3 settimane; 9 sono tornati alle famiglie affidatarie nei fine settimana (gruppo FF) e 9 sono rimasti a scuola (gruppo Sc). I risultati hanno mostrato che i cani che frequentavano la scuola esprimevano un comportamento meno stressante a riposo (GLMM; DF=1; F=10,11; p=0,0018). Una scala analogica visiva completata dall'addestratore cinofilo ha indicato che erano più concentrati durante le sessioni di addestramento (GLMM; DF=1; F=5,42; p=0,0326). Inoltre, i cani erano ben abituati alla vita nel canile della scuola, con il rapporto neutrofili/linfociti e livelli di serotonina, prolattina e ossitocina in linea con i tassi normali in entrambi i gruppi. Questi risultati suggeriscono che l'ambiente della scuola di addestramento non ha alcun impatto negativo sui cani ed evitare ripetute separazioni con la famiglia affidataria potrebbe ridurre lo stress e aumentare il loro benessere. Ciò dovrebbe indurre a considerare l'ambiente abitativo dei futuri cani guida.