Comparison of Selected Behavioural Patterns of German Shepherd Puppies in Open-Field Test by Practical Assessment Report

Igor Miňo, Lenka Lešková

Abstract—Over the past 80 years, open-field method has evolved as a commonly used tool for the analysis of animal behaviour. The study was carried out using 50 kennel-reared purebred puppies of the German Shepherd dog breed. All dogs were tested in 5th, 7th, and 9th week of age. For the purpose of behavioural analysis, an open-field evaluation report was designed prior to testing to ensure the most convenient, rapid, and suitable way to assess selected behavioural patterns in field conditions. Onset of vocalisation, intensity of vocalisation, level of physical activity, response to sound, and overall behaviour was monitored in the study. Correlations between measures of height, weight and chest circumference, and behavioural characteristics in the 5th, 7th, and 9th week of age were not statistically significant. Onset of vocalisation, intensity of vocalisation, level of physical activity and response to sound differed on statistically significant level between 5th, 7th, and 9th week of age. Results suggest that our practical assessment report may be used as an applicable method to evaluate the suitability of service dog puppies for future working roles.

Keywords-Dog, behaviour, open-field, testing.

I. INTRODUCTION

PEN-FIELD test has attained the status of one of the most widely used instruments in animal psychology. This test consists of subjecting an animal to unknown environment without the chance to leave. It has served as a simple and widely applicable tool which allows an easy and rapid analysis of selected behavioural patterns. Animal's reactions in openfield test are usually explained in terms of emotionality, fearfulness, temperament, stress-susceptibility, exploration, or copying style [1]. Open-field test was introduced by Calvin Hall [2], [3], who tested behavioural responses of rodents by using defecation in open-field as an index of timidity. In dogs, open-field test was used for the first time by Martinek and Lát [4]; their findings confirmed long-term stability of individual differences in exploratory and other behaviour, and the rate of habituation in dogs. Open-field test was also carried out in dogs to study the effects of some drugs [5]. Studies using open-field test were carried out to assess habituation to novel stimuli in an open-field [6], or to observe activity in an openfield, and the effect of human interaction, age and breed in dogs on their behaviour [7]. Recently, the open-field model has been used to assess sound-induced fear and anxiety-

Igor Miňo is with the University of Veterinary Medicine in Kosice, Slovakia (e-mail: igor.mino@uvlf.sk).

associated behaviours in Labrador Retrievers [8]. When testing dog behaviour, it is important to note that puppies are considered to go through a sensitive period early in their development stages, from approximately three to 12 weeks of age; this may establish some aspects of the behaviour of the dog for the future [9]. In a successful puppy test, the behaviour measured must not be overly influenced by the environment experienced following the testing [10]. Puppy testing typically involves scoring the reactions of young dogs against a series of controlled stimuli and using this information to compare individual differences in behaviour. Assessment must evaluate behaviour that has been consistent within an individual animal over time in similar situations; such behavioural signs reflect an individual's personality. For this reason, puppy tests must detect consistent behaviour, which is the main requirement for personality assessment [11], [12]. Such consistency in behavioural expression has been demonstrated for dogs under one year of age, with measurement items from behaviour assessments achieving average consistency correlations between assessments of 0.34 [13]. However, the majority of studies that assess puppy behaviour [14]-[16] failed to provide evidence of reliability, temporal consistency, or construct validity for designed trials, or included a low number of testing subjects for the results to be meaningfully interpreted [17], [18]. No study has specifically addressed which behavioural responses in juvenile dogs could reflect personality traits. Possibility to select puppies for different purposes should be based on scientific knowledge. Reliable and valid behaviour tests for puppies would be benefitial for working dog organizations, which select dogs at a young age for subsequent training to perform a particular role [19], [20]. The aim of our study was to assess the effect of weight, body measures, age, and gender on the results of behavioural testing. Our hypothesis was that the results of behavioural testing, mainly vocalisation patterns, physical activity, or responses to different stimuli are strongly affected by the afore-mentioned factors.

II. MATERIAL AND METHODS

A. Subjects

The German Shepherd puppies used for testing (n = 50) were all bred at the Slovak Republic Police Breeding Facility. The experimental sample consisted of 32 males and 18 females. They originated from seven different litters (five to nine puppies in each litter). The puppies were marked with

cuts in the fur for identification. All animals were kept in the same conditions and were tested individually at the age of five, seven, and nine weeks. The puppies were always tested before feeding. Only dogs without health problems were involved in testing. Every testing day was preceded by the measurement and weighing of puppies, with the chest circumference, weight, and height at the withers being recorded.

B. Behavioural Testing Procedures

The open-field testing facility consisted of 2.5 meter x 2.5 meter testing arena. The inward-opening door provided access into the arena. The floor of the arena was divided into four squares (125 cm x 125 cm) marked with black waterproof tape for easier movement quantification (locomotor activity was evaluated by counting exceeded squares). Prior to each testing, the floor was cleaned with a disinfectant solution to minimize the disturbing effects of odour on the dogs' behaviour. In one day, only one litter has been tested. The open-field test took five minutes, and the testing and manipulation with the puppies was carried out by the same person. A dog was placed inside the arena and its behaviour was recorded by video camera. Each video record has been archived and properly marked with date and time when the puppy was tested.

C. Evaluation of Recorded Behaviour

The video record was evaluated by three independent ethological observers that provided quantitative measures of locomotion (movement), inactivity (standing or sitting still), grooming (licking, scratching, etc., aimed at the dog's own body), urination, rearing (two front paws lifted off the floor), vocalisation and jumping (all four paws off the floor). At the end of open-field test, the response to sound stimulus (banging on the door) was observed and recorded. During the testing four main areas of interest were evaluated: movement, vocalisation, general response to unknown environment and dog's response to unexpected noise. Attention was paid particularly to the selected parameters of vocalisation, activity, and general evaluation of behaviour. Test results were associated with weight, height, chest circumference, and also with the sex and age of the dogs.

D. Designing of Evaluation Protocol

Behaviour was evaluated by means of a special protocol that was designed for this purpose. Preliminary study on 20 puppies was carried out prior to the testing to search for the most common behavioural patterns in dogs. We assessed the most common reactions and responses of puppies to different stimuli during the open-field test, and according to our results, we have created the scoring that was used for the study. Reactions and behaviour of puppies were either measured in seconds or scored on a scale from 0 to 5 points (more desirable response/behaviour for a future working purpose of a dog was evaluated with the higher score). Final assessment of the behaviour during the open-field test in general was summarized by a description of the most likely response.

E. Statistical Analysis

Statistical analyses were performed using the IBM SPSS Statistics 22 and for all tests p-values of <0.05 were considered significant. Cohen's kappa was used to determine inter-observer reliability with the result $\kappa = 0,698 - 0,770$. Differences between the means were not normally distributed (Shapiro Wilk, P<0.05) and therefore, testing was done using a non-parametric tests. Differences in explored characteristics based on sex were assessed by Kruskal-Wallis test. Correlation between measures of height, weight, and chest circumference, and explored characteristics was explored using Spearman test. Longitudinal change between the age of five, seven and nine weeks was analyzed with Friedman matched-pairs signed ranks test.

III. RESULTS

Practical open-field protocol was designed for the evaluation of selected behavioural patterns in puppies, which would be applicable even in field conditions (Table I).

TABLE I PRACTICAL OPEN-FIELD PROTOCOL FOR EVALUATION OF SELECTED BEH AVIOUR AL PATTERNS IN PURPLES

BEHAVIOURAL PATTERNS IN PUPPIES						
Assessment	Puppy began to vocalise after seconds after					
of adaptation	nsertion into the open-field room.					
Assessment	 intense with a declining trend mild and balanced 					
of vocalisation						
	3 very intense, almost hysterical, had no downward trend					
Assessment	Number of squares crossed by both front limbs was					
of locomotor	counted in every minute individually, and at the end					
activity	total number of crossed squares was calculated.					
Assessment	1 puppy shows enjoyment, jumps on the door					
of response to	2 expressing increasing interest in, sit up					
sound	3 puppy is scared, it tries to run away					
	4 no reaction					
Overall	 intense exploratory behaviour, 					
assessment	 popping up on walls and room doors, 					
of the behaviour	- the ears are usually erect and the tail is up					
during	 puppy does not jump on the walls, 					
open-field test	 average exploratory behaviour, 					
	 ears and tail are released 					
	3 - exploration is associated with chaotic jogging,					
	 puppy has a fear reaction, 					
1	- ears and tail are mostly down					

The purpose of our research moreover was to establish the association between body measures and behavioural responses in puppies. Spearman correlations between measures of height, weight, chest circumference and explored behavioural responses during ontogenesis in 5th, 7th and 9th week of age are displayed in Table II. Most of them were not statistically significant.

The effect of sex on the results of behavioural tests was also studied. An experiment was carried out to evaluate changes in behavioural responses during the early stages of ontogenesis and compared to the impact of age and sex on the results of the open-field test. The Kruskal-Wallis test revealed no statistically significant differences in the explored characteristics based on gender in 5th, ^{7th} or 9th week of age. The results are displayed in Table III.

TABLE II SPERMAN CORRELATIONS BETWEEN CHOSEN CHARACTERISTICS IN THE AGE OF FIVE, SEVEN AND NINE WEEKS AND WEIGHT, HEIGHT, CHEST

	Weight	Height	Chest circumference					
Week 5								
Onset of vocalisation	-0.18	-0.13	-0.20					
Intensity of vocalisation	0.09	-0.19	0.04					
Level of physical activity	-0.21	0.06	-0.02					
Response to sound	-0.10	-0.04	-0.26					
Overall behaviour	0.05	-0.18	-0.14					
	Wee	k 7						
Onset of vocalisation	-0.26	-0.44***	-0.24					
Intensity of vocalisation	-0.04	-0.04	-0.03					
Level of physical activity	-0.15	0.17	-0.08					
Response to sound	-0.29*	-0.13	-0.17					
Overall behaviour	0.04	-0.12	0.15					
	Wee	k 9						
Onset of vocalisation	-0.30*	-0.26	-0.40**					
Intensity of vocalisation	0.12	0.04	-0.02					
Level of physical activity	0.04	0.19	-0.03					
Response to sound	0.07	0.08	-0.22					
Overall behaviour	0.01	-0.11	-0.09					

*p<0.05, **p<0.01, ***p<0.001

TABLE III DIFFERENCES IN CHOSEN CHARACTERISTICS IN THE AGE OF FIVE, SEVEN AND NINE WEEKS BY GENDER

	Male Mean (SD)	Female Mean (SD)	p-value (Kruskal-Wallis test)					
Week 5								
Onset of vocalisation	21.16 (15.94)	27.94 (18.12)	0.21					
Intensity of vocalisation	2.97 (1.15)	3.22 (1.17)	0.47					
Level of physical activity	14.94 (7.21)	17.78 (8.00)	0.23					
Response to sound	3.22 (1.07)	3.17 (0.99)	0.80					
Overall behaviour	2.16 (0.63)	2.11 (0.68)	0.83					
Week 7								
Onset of vocalisation	23.38 (31.31)	24.28 (17.55)	0.21					
Intensity of vocalisation	2.25 (0.88)	1.83 (1.10)	0.04					
Level of physical activity	17.72 (6.51)	15.28 (3.77)	0.21					
Response to sound	2.22 (0.94)	2.00 (0.59)	0.58					
Overall behaviour	1.59 (0.56)	1.67 (0.59)	0.69					
Week 9								
Onset of vocalisation	11.97 (14.61)	15.94 (9.65)	0.02					
Intensity of vocalisation	1.56 (0.91)	1.72 (0.96)	0.43					
Level of physical activity	18.19 (7.28)	25.39 (12.27)	0.05					
Response to sound	1.69 (0.47)	1.67 (0.49)	0.88					
Overall behaviour	1.31 (0.47)	1.22 (0.49)	0.50					

Based on Friedman's test, all explored characteristics differ on a statistically significant level between 5th, 7th and 9th week of age. The results are displayed in Table IV.

IV. DISCUSSION

Our results so far indicate that 1) onset of vocalisation occurs faster with increasing age; 2) intensity of vocalisation decreases with age; 3) physical activity increases and the response to auditory stimuli is snappier with each retesting; 4) behavioural patterns that were studied in general were rated as the most desirable in the ninth week of age compared to earlies stages of ontogeny; this means that exploratory behaviour is more intense, puppies are more active, it is possible observe popping up on walls and room doors, the puppies exhibit less stress reactions, and their ears are usually erect and the tail is up.

TABLE IV CHANGE IN CHOSEN CHARACTERISTICS IN THE AGE OF FIVE, SEVEN AND NINE WEEKS

ININE WEEKS							
	Week 5 Mean (SD)	Week 7 Mean (SD)	Week 9 Mean (SD)	p-value (Friedman test)			
Onset of vocalisation	23.60 (16.90)	23.70 (26.97)	13.40 (13.08)	0.001			
Intensity of vocalisation	3.06 (1.15)	2.10 (0.97)	1.62 (0.92)	0.001			
Level of physical activity	15.96 (7.54)	16.84 (5.75)	20.78 (9.90)	0.01			
Response to sound	3.20 (1.03)	2.14 (0.83)	1.68 (0.47)	0.001			
Overall behaviour	2.14 (0.64)	1.62 (0.57)	1.28 (0.45)	0.001			

Our assessment tool is intended for use as a measure of behavioural changes. It assesses behavioural responses over time, throughout similar situations, and is designed in a way where each individual has an equal opportunity to display the full range of responses. In an open-field test, subjects are placed in a relatively novel environment and observed for a fixed period of time. The novelty of the test situation is likely to affect behavioural activity. We can suggest that locomotor responses to a novel open-field reflect behavioural reactivity rather than differences in motor activity. In our study, we have observed a statistically significant increase in the physical activity (p-value in Friedman test was 0.01). Furthermore, according to a general assessment of behaviour during the entire testing, also statistically more significant improvement with the increasing age of the puppies (p-value in Friedman test was 0.001) was observed. Our results are consistent with other studies showing increasing physical activity and exploratory behaviour in an open-field during this period of life [21]. We have hypothesized that exploratory behaviour could be with the most probability related to the development of some cognitive processes. It can be assumed that the level of activity will increase until the completion of puberty. In the later period with increasing age, a gradual decrease in physical activity has been shown, as it was reported in the study of [7]. Behavioural sex differences in dogs have so far scarcely been reported. However, Scott and Fuller [22] found males to be dominant over females in competitive situations at the age of eight weeks [23]. In our study considering the intensity of the vocalisation, the seven-week-old males showed a significantly higher score than the females. Statistically significant differences were also found in the onset of vocalisation in the 9th week when the males started to vocalize much earlier than the females. On the other side, we have noticed the opposite situation in the level of physical activity in the 9th week when females dominated over males with a higher rate of locomotor activity. This difference was due to the different method of testing. While Wilson and Sundgren [23] tested puppies

together during a competitive situation, in our test, each puppy was tested individually in unknown environment.

In our study, we also tried to find out at what age it is possible to observe the biggest individual differences in puppy behaviour. Although many important life stages may be observed in canine ontogeny, the key sensitive period in a puppy's development is generally accepted to be the 10-week period from three weeks to three months of age. This 10-week period is the primary sensitive period of canine development. This time period is generally referred to as the 'socialisation period', for this is when species-typical social skills are learned most rapidly [24]. Our study confirmed that puppies are able to learn very quickly in this period of time and differences may be noticed week after week. During each testing, we observed statistically significant differences in every category of behaviour (onset of vocalisation p = 0.001; intensity of vocalisation p = 0.001; level of physical activity p = 0.01; response to sound p = 0.001). Definitely the biggest individual differences in our study were found during testing in the 9th week of age. However, the behaviour of puppies continues to develop; therefore, it would be useful to monitor behavioural changes also in the following weeks as well. Some authors argue that behavioural testing should be carried out up to 12 weeks of age. Yet, we focused on behavioural changes that take place before the puppies are affected by the changing of their environment and of their owner.

Many tests in young dogs have been performed in order to evaluate which components of temperament are the most important and may be reliably observed in early age. We agree with the statement of Svartberg [25] that the most important temperament characteristics are fearfulness and aggression. Goddard and Beilharz [14] suggested that fearfulness, being one of the most common reasons for the rejection of dogs, could be evaluated as early as 12 weeks of age. However, we believe that fearfulness, as well as reactivity of the puppies, can be reliably assessed as early as the 9th week of the puppies' development by our practical assessment report [26]. We believe that specifically fearfulness and physical activity in general are the most important indicators of future behaviour because exploratory measures in open-field activity test are correlated with cognitive function [7], [8]. Fearfulness in our study was evaluated according to dog's response to a specific sound stimulus. The results of our study were similar to the study of "Emotional Reactivity Test" [27]. It suggests that the response to the sound stimulus may be viewed as a useful measure for selection or rejection of a service dog. Further performance testing is need to determine if an openfield test to sound stimuli might predict dog response to the loud sounds that occur in combat situations [8]. This is an important area for future research that will result in further scientific progress to understanding the differences in behaviour during the adolescence of puppies.

A. Compliance with Ethical Standards

All ethical standards considering the treatment of living animals during behavioural testing were kept.

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