Aus dem Department für Nutztiere und öffentliches Gesundheitswesen in der Veterinärmedizin
der Veterinärmedizinischen Universität Wien

Institut für Tierschutzwissenschaften und Tierhaltung

(Leiter: Univ.-Prof Jean-Loup Rault PhD.)

Effect of cooperative care training on physiological parameters and compliance in dogs undergoing a veterinary examination

Diplomarbeit

Veterinärmedizinische Universität Wien

vorgelegt von

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Wien, im Juni 2021

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Danksagung

Ich möchte mich herzlich bei meiner Betreuerin, Frau Dr. med. vet. Christine Arhant, für ihre ausgiebige Geduld und Unterstützung bedanken, sowohl bei Planung und Durchführung der Studie sowie der Erstellung meiner Diplomarbeit. Ebenso gilt mein Dank meinem Ko-Betreuer Jason Yee, PhD. Beide haben mir mit zahlreichen Erklärungen und viel Verständnis das anfänglich doch fremde Gebiet der Herzfrequenzanalyse nähergebracht.

Besonderer Dank gilt allen, die einen großen Beitrag zur Verwirklichung dieses Projektes geleistet haben. Frau Dr. med. vet. Nadja Affenzeller und die Onkologie der Vetmeduni Vienna, dank denen wir eine echte, authentische Ambulanz zur Verfügung hatten. Auch der Gesellschaft zur Förderung Kynologischer Forschung e.V. (GKF), die unser Projekt mit Fördermitteln unterstützt hat, möchte ich meinen Dank aussprechen, ebenso wie Herrn Dr. Günther Schauberger für das zur Verfügung stellen eines Temperaturkalibrators für unser Equipment. Ein riesiges Dankeschön an meine Studienpartnerinnen, Miriam Schützinger und Astrid Böhm, für die großartige Zusammenarbeit beim Mammutprojekt „erste eigene Studie". Und natürlich allen Besitzerinnen und Besitzern, sowie ihren Hunden, die mit ihrer Teilnahme das Projekt möglich gemacht haben.

Größter Dank gilt meiner Familie und meinen Freunden für die Unterstützung während meines gesamten Studiums. Meinem Vater und meiner Schwester, die mir jederzeit sowohl mental als auch finanziell zur Seite gestanden sind. Und meiner Mutter, die meinen Studienweg leider nicht mehr miterleben durfte und der diese Arbeit gewidmet ist.

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1. Introduction

While important to ensure good health and welfare of dogs, a veterinary examination can cause fear and stress and negatively impact the dog's wellbeing (Döring et al. 2009, Mariti et al. 2015, Csoltova et al. 2017). Wellbeing during clinical examinations and treatment is directly influencing the overall welfare (Christiansen und Forkman 2007). According to studies, a majority of dogs seem to experience fear while undergoing those examinations. 60 % (Beaver 1999) up to 78 % (Döring et al. 2009) of dogs at a veterinary clinic could be described as fearful. This is not only of importance for the health and general condition of the dogs themselves but can also hinder the process of the examination (Glardon et al. 2010) and possibly even endanger the veterinary staff (Campbell 1999, Döring et al. 2009). The chance of being bitten, scratched or kicked poses a realistic threat in a veterinary practice (Nienhaus et al. 2005). In Australia, 20 % of animal inflicted injuries are caused by dogs and 85 % out of them are bite injuries (Lucas et al. 2009). As negative experiences can serve as a form of conditioning for the animals for future visits (Simpson 1997), they may subsequently lead to elevated levels of fear and increasingly lower compliance in these situations. This may result in treatments becoming more difficult or only possible while under sedation, as cats and dogs that are difficult to handle have been observed to be more likely to inflict bite wounds (J. Drobatz and Smith 2003, Döring et al. 2009).

As several physiological parameters reflect arousal and affective state in the form of measurable changes, those parameters can be used to assess a dog's welfare. Useful parameters described are heart rate (HR) and heart rate variability (HRV) (Beerda et al. 2000, von Borell et al. 2007) and body temperature including rectal or eye temperature (Travain et al. 2015). Furthermore, the temperature difference between the left and right tympanic membrane might indicate levels of stress as well, as it may conform with differences in cerebral blood-flow due to the tympanic membrane being perfused by the same arterial circuits as the distal cortical structures (Boyce et al. 2002). In cats, an increased temperature of the right tympanic membrane was described to be correlated with increased cortisol levels (Mazzotti and Boere 2009). In addition to physiological measurements, behavioral data can be used to evaluate a dog's emotional state (Beerda et al. 1998, Machado and Silva 2020). In human as well as in animal psychophysiology, a two-dimensional model is commonly used to describe affective state by differentiating valence and arousal; valence determines the effect of a stimulus on affect ranging from positive to negative and the level of arousal can range from low to high (Russell 2003, Mendl et al. 2010). HR and HRV are commonly used metrics that open a window to affective state by assessing autonomic nervous system activation. While HR and "Standard Deviation of the NN Interval" (SDNN) represent the combined influence of all branches, "Root Mean Square of Successive Differences" (RMSSD) is thought to reflect the unique contributions of the parasympathetic branch (Després et al. 2002, von Borell et al. 2007, Kuhne et al. 2014), although this seems not to be strictly the case in all situations (Berntson et al. 2005).

There have been various attempts to lower stress for dogs in a clinical environment. The concept of low stress handling techniques, also known as "Fear Free", has proven to be effective in decreasing fear related behavior in dogs (Scalia et al. 2017). Pharmacological attempts to reduce stress include for example the use of dexmedetomidine oromucosal gel that was found to reduce behavioral signs of stress in dogs during veterinary examinations (Hauser et al. 2020). Another approach, which avoids the use of drugs, is to prepare patients for the examination via training. In the case of cats, a six-week carrier training was able to reduce stress during transport (Pratsch et al. 2018). A four-week desensitization and counter-condition training program on dogs with pre-existing fear of veterinary examinations was found to not influence body temperature, HR or respiratory rate, but trained dogs showed less reduced posture (Stellato et al. 2019b), which is commonly used as an indicator for fear in dogs (Beerda et al. 1998, Döring et al. 2009, Stellato et al. 2019a). Owners also claimed an observable improvement in their dogs, therefore showing the need of further research in this area.

The aim of our study was to investigate the impact of a "cooperative care training" (Howell and Feyrecilde 2018) on stress in dogs, reflected by the physiological parameters heart rate, heart rate variability and tympanic membrane temperature, during a veterinary examination. This form of training introduced the dogs to a target (e. g. a mat located on the examination table). Dogs were trained via positive reinforcement to put both front paws on this target and introduced to common manipulations during veterinary exams. By using positive reinforcement, unpleasant stimuli, like undergoing a medical examination, can become associated with the positive stimulus (e. g. receiving treats) (OHeare 2011), which may ultimately lead to dogs being more willing to participate. The second important aspect of the training was to show the dogs that they will only be manipulated as long as they keep their front paws on the target, thus establishing a “cooperation signal” for them to indicate their willingness to participate (Laule et al. 2003, Coleman et al. 2008). If the dogs wanted the manipulation to be stopped, they simply needed to step off the target, providing an easily understandable way of communication between dogs and humans. This form of communication has already been successfully used in rhesus macaques (Coleman et al. 2008).

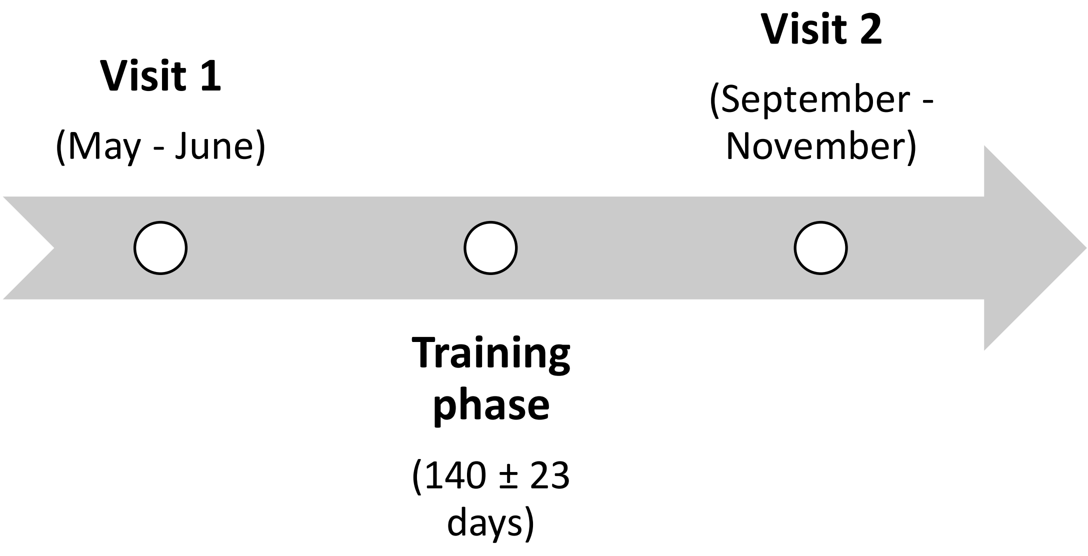
We hypothesized that this type of training leads to dogs showing reduced signs of arousal and an emotional state consistent with resilience to stressors during a veterinary examination. Therefore, we expected a lower heart rate, a higher heart rate variability and a smaller difference in temperature between the right and left tympanic membranes in trained dogs compared to dogs that did not receive this type of training. Furthermore, we expected better compliance in trained dogs.

2. Materials and Methods

2.1. Overview of study design

A blinded semi-randomized controlled trial was used to evaluate the effects of a "cooperative care training" regarding compliance and welfare of dogs during a veterinary examination. The experiment consisted of two veterinary examinations (visit 1 and visit 2), split into three periods (waiting room before the examination, the examination itself in the examination room and waiting room after the examination). The first examinations were conducted in May and June 2019. In the following weeks, the training group (TG) received the "cooperative care training" while the control group (CG) received a different set of exercises unrelated to veterinary procedures to control for increased human-dog interaction in the TG during the training phase. The second examinations took place in September, October and November 2019, 140 ± 23 days (Min: 90; Max: 183) after the first one. The persons conducting the veterinary examination were blinded to the dog's group allocation. This study was discussed and approved by the institutional ethics and animal welfare committee in accordance with GSP guidelines and national legislation (ETK-05/01/2019). Informed consent was obtained of the dog owners taking part with their privately owned dogs.

Fig. 1: Study timeline



2.2. Animals

A total number of 47 dogs were initially enrolled in the study. Participants were recruited via flyers and the social media channels of the Vetmeduni Campus and the Clever Dog Lab. If dog owners showed interest in participating, their dogs were evaluated for suitability in a personal appointment at the Clever Dog Lab. Inclusion criteria for the dogs were the following:

- age 1-10 years
- no current health problems
- no generalised anxiety behavior towards unfamiliar people
- no history of showing severe aggression towards veterinary staff
- up-to-date core vaccinations

After the first veterinary examination, those 47 dogs were semi-randomly assigned into the TG (n = 26) and CG (n = 21). The groups were balanced for age, sex, neuter status, a "fear of veterinary exams score", prior training experience and effort of travel to reach the place of the study. Requests by the owners to participate in a specific group were also considered.

Exclusion criteria included demonstrating aggression during the veterinary examination or developing a health problem during the study period. However, no dog had to be excluded for those reasons. Out of the 47 dogs that underwent visit 1, seven dropped out of the study prematurely due to personal reasons of the owners. Therefore, a total of 40 dogs remained to complete the full study. Those 40 dogs belonged to 16 different breeds as well as various crossbreeds. The average age was 4.8 years (TG 4.9 years; CG 4.7 years). The gender ratio was nearly identical for both groups (TG: 14 females, 8 males; CG: 12 females, 6 males). The prior training experience of both dogs and owners and the "fear of veterinary exams score" was rated by the owners themselves via filling out an initial survey. Training experience was rated on a scale from 1 to 5, with 5 representing very experienced. The average was between 3 and 4 for both dogs and owners in the TG and CG. The "fear of veterinary exams score" was rated from 1 to 5 with 1 meaning frequent occurrence of the behavior and 5 meaning no observations of the behavior at all. In total, 10 behaviors were rated, namely freezing, trembling, panting, trying to hide, seeking comfort, growling, elevating lips, snapping in the air or at persons and involuntary urination or defecation. The average score of the training group was 3.99 and that of the control group 4.18. In none of the parameters balanced during group assignment, significant differences were found (all $p > 0.2$).

2.3. Experimental Procedure

2.3.1. First visit

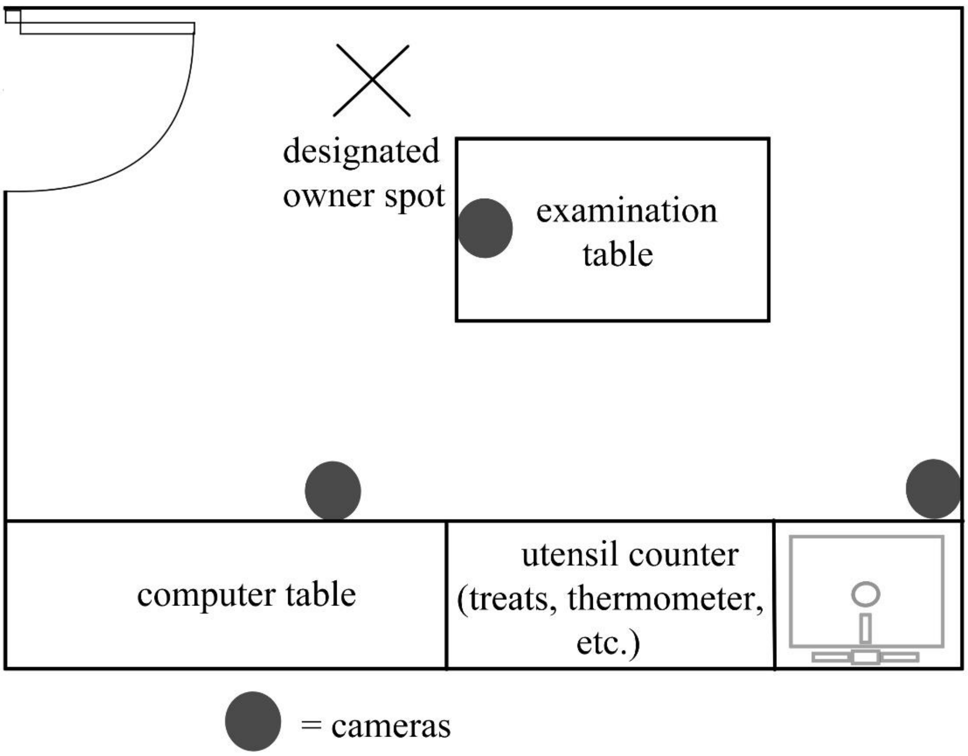
The veterinary exams were held at the oncology department of the Clinical Unit of Internal Medicine for Small Animals of the Vetmeduni Vienna, ensuring an authentic veterinary environment (fig. 2). Exams were only held during the clinics closing hours and windows and glass doors were taped shut in order to prevent any external influences that could differ between participants.

For the examination the dogs were called to the oncology clinic one by one. After being greeted with treats, the dogs were equipped with a Polar electrode belt (fig. 3). A proper connection was ensured with the use of water on the fur and ultrasound gel on the belt itself. Afterwards, the owners and their dogs were left alone in the waiting room for 20 minutes. This period was meant to give the dogs time go get familiar with the belt and the surroundings. During this time owners were free to offer their dogs treats if this was the usual practice for them on regular vet visits. After this acclimatization period of 20 minutes, the first measurement period (waiting room before exam) started. The owners were asked to leash their dog to prevent movement and instructed to avoid interacting with the dog. The owner and its dog were left undisturbed during this period which lasted for five minutes.

Next, the participants were led into the examination room. The dog was given a three-minute acclimatization period to explore the room while a total number of three treats were tossed on the floor by the vet. Then the dog was lifted on the examination table. This was usually done by the owner. The table was 82 cm high and measured 110 cm in length and 70 cm in width. It was covered with a rubber table mat in order to prevent direct contact of the dogs' claws with the metal surface, which could otherwise produce unpleasant sounds. On the front-end of the table, a common bathmat (20 cm length, 65 cm width) had been placed to be used as a front-paw-target after the training. This target was part of the cooperative care training that the training group was yet to receive, but also helped to optimize positioning of dogs for video recordings. Before the exam started, the dog was again offered three treats, this time by the owner. Afterwards a clinical veterinary examination was performed. Order of the different steps and technique were standardized (tab. 1). During the examination the owners stood at a designated spot near the table in view of the dog and were asked not to interact with the dog. At specific steps/points in time they were asked to offer a treat to the dog. Throughout the exam, low-stress handling techniques were used (Yin 2009). The dogs were only slightly restrained by the assistant placing one hand on the chest and the other under the belly to assure safety (e. g. by prevent the dog from jumping off the table). If a dog struggled or tried to escape/jump from the table three times during the same step of the exam (tab. 1), the exam was cancelled. Furthermore, the owners were instructed that they could request a break or the cancellation of the exam at any time. Once the exam was finished or cancelled, the dogs remained on the table for the measuring of the tympanic temperature, with each ear being measured twice. Only in case the dog had attempted to jump/escape from the table, the tympanic temperature was taken on the floor. Afterwards, three treats were offered by the vet and the dogs were put back onto the ground for a three-minute period. During this period, three treats were again offered by the vet by hand. If one or more of those were rejected, they were tossed on the ground instead. Of all treats offered in the examination room it was noted down whether they were accepted or not (tab. 2).

Afterwards, owners and dogs were led back into the waiting room and another measurement period followed (waiting room after the exam; duration: 10 minutes), performed identical to the first.
Roles of the veterinary assistant and veterinarian (for both visit 1 as well as the later visit 2) were performed by Veterinary Medicine students M. Schützinger and L. Wess respectively.

Fig. 2: Examination room plan (only schematic – not true to scale)



Tab. 1: Examination scheme and techniques

Examination	**Body part**	**Duration (seconds)**	**Technique**
Auscultation left lung	Left thorax	15 s	1. Show stethoscope (let dog sniff) 2. Pet from neck to thorax 3. Put on stethoscope 4. Put second hand on dog's back
Auscultation heart		30 s	1. Move stethoscope from thorax to heart
Treat (+ vet and assistant switching sides)			
Auscultation right lung	Right thorax	15 s	1. Show stethoscope (let dog sniff) 2. Pet from neck to thorax 3. Put on stethoscope 4. Put second hand on dog's back
Treat			
Adspection ears	Head both sides	1 s	1. Show hands (let dog sniff) 2. First hand under chin 3. Second hand touches ears
Treat			
Adspection conjunctivae	Head both sides	2 s/eye	1. Show hands (let dog sniff) 2. First hand under chin 3. Second hand opens eyes
Treat			
Adspection oral mucosa/teeth	Head both sides	1 s/side	1. Show hands (let dog sniff) 2. First hand under chin 3. Second hand elevates upper lip
Capillary refill time	Head one side	3 s	1. Fluent transition from second oral mucosa 2. Apply pressure on oral mucosa and watch capillary refill
Treat			
Palpation abdomen	Abdomen	30 s	1. Show hands (let dog sniff) 2. Pet from neck to abdomen two times 3. Apply soft pressure on abdomen three times 4. Perform deep palpation
Feel femoral pulse	Both hind legs	15 s	Fluent transition from abdomen to hind legs
Treat			
Rectal temperature	Rectum	Until signal given	1. Show hands and thermometer (let dog sniff) 2. Pet from tail to flank 3. Pet over tail root 4. Elevate tail 5. Insert thermometer
Treat			

Tab. 2: Number of treats accepted by TG and CG dogs during different time periods (possible maximum in brackets)

	Visit 1				visit 2			
	Before exam (3)	During exam (7)	After exam (3)	Off table (3)	Before exam (3)	During exam (7)	After exam (3)	Off table (3)
TG								
Mean	3	6	3	3	3	7	3	3
Min	0	0	0	3	0	0	0	3
Max	3	7	3	3	3	7	3	3
CG								
Mean	3	5	3	3	2	5	2	3
Min	0	0	0	3	0	0	0	3
Max	3	7	3	3	3	7	3	3

2.3.2. Training

During the training period, the training group received a cooperative care training in the form of group sessions with up to 5 dogs and their owners plus additional individual training at home. On average, a dog received 10 group training sessions, depending on progress and time resources (S.D.: 2; Minimum: 8, Maximum: 12). During the training, the dogs were introduced to a "front paw target", which was represented by a mat. Owners received instructions to train their dogs the use of this target in order to perform a "cooperation signal", which consisted of putting both front paws on the mat. In addition to this, they were introduced to the different kinds of manipulations that made up the veterinary examination. They only had to endure these manipulations as long as they performed the cooperation signal and as soon as they stepped off the target, any manipulation came to an immediate stop. Owner also received aid in recognizing and understanding more subtle body language in their dogs, which they might use to communicate discomfort.

One important goal of the training was to use positive reinforcement as a main motivator for the dogs' participation. In order to achieve this, treats were used as a reward for performing the cooperation signal itself and for enduring the various manipulations. Conversely, ceasing cooperation resulted in both negative punishment due to no longer receiving treats as well as negative reinforcement in the form of the manipulation being stopped. The training was therefore adapted to each dog individually in order to keep the positive reinforcement as the main driving factor. Furthermore, difficulty levels and training duration were increased only in small steps. Just like during the veterinary examination, dogs were always prepared for incoming manipulations by being presented both the manipulators hands and equipment as well as being gently touched/stroked at the respective body part.

The control group received written training instructions that were not connected to the veterinary procedures used in the study and did not include any target training.

The training was carried out by A. Böhm, a more detailed description can be found in her Master's thesis "*Effect of cooperative care training on dog's behavior during a veterinary examination*" (Böhm 2020).

2.3.3. Second visit

After having finished the training period, all participants had to undergo a second veterinary examination. The examination took place in the exact same way as the first veterinary examination. The only difference was the TG being able to make use of the target present on the examination table. Again, the owners were instructed that they could ask for breaks or cancellation at any time. In addition, the owners of the TG learned during the training phase to ask for a break if their dog stepped off the target during the examination. For the CG the target was present but solely used to support positioning of the dog. Both experimenters (veterinarian and veterinary assistant) ignored behavior in relation to the target (e. g. did not react to stepping down from it), as they did not know if a dog was part of the TG or the CG.

2.4. Measurements

In order to assess the dogs' emotional state, both data on behavioral and physiological parameters was collected. Behavior of dogs was video recorded during the examination, behavioral analysis was however not part of this thesis and results are therefore discussed elsewhere (Böhm 2020). Physiological measurements consisted of heart rate data in the form of beat-to-beat recordings and tympanic membrane temperature. In addition, we collected data on the compliance during the exam (whether the exam could be finished or had to be cancelled prematurely) and surveyed perceived training success in the trainer and as well as in dog owners. Details on training success will be presented in "*Evaluierung des Trainingsprozess eines sogenannten "cooperative care training" beim Hund*" (Schützinger in prep).

2.4.1. HR and HRV

HR data in the form of continuous beat-to-beat recording was collected using a Polar heart rate monitor (Polar RS800CX, Polar Eelectro Oy, Finland) and an accompanying electrode belt (fig. 3). First, Polar Precision Performance SW, a software provided by Polar, was used to measure the estimated error count per minute in order to achieve a rough pre-selection of suitable segments. The filter settings were set to "very low". Minutes with very high error rates (>30 %) were excluded. Generally, data quality was better in the veterinary exam period. Therefore, it was possible to choose a suitable three-minute segment for the examination period. For both waiting room periods shorter two-minute segments had to be selected. However, to avoid bias of results due to the difference in sample length, the examination segments were shortened into two-minute segments when analyzed with waiting room periods in the same statistical models. To avoid bias from using different time points within a period, the timing of the minutes was matched between visit 1 and visit 2 for each dog. For the waiting room periods, the first and last minute were avoided where possible due to possible interference caused by the experimenters leaving and entering the room. For the exam period, segments were chosen to be balanced around minute 3 in order to maintain comparability.

Overall, for TG dogs data was available from up to 16 dogs (waiting room before exam: 13, exam: 16, waiting room after exam: 12) and for the CG from up to 17 dogs (waiting room before exam: 11, exam: 17, waiting room after exam: 11).

Interbeat interval (IBI) data of the selected minutes was then transferred into an Excel file and errors were identified manually, either visually or based on differences between IBI values. Identified errors were corrected accordingly (tab. 3). In this step, an error count of <10 % was deemed acceptable. The corrected sequences were then analyzed using the heart rate variability analysis software Kubios HRV Standard version 2.1. Parameters HR (mean heart rate), RMSSD, SDNN and RMSSD/SDNN ratio were calculated.

Fig. 3: Polar RS800CX heart rate monitor and accompanying electrode belt

Tab. 3: Different types of errors and method of correcting them

Type	Description	Identification	Correction	Reference
Type 1	Incorrect beat detection	Single value deviates by over ~40-80 % to neighbouring values	Replace by mean value of neighbouring IBIs	(Marchant-Forde et al. 2004)
Type 2	Single low value followed by single high value	First value noticeable lower than precedent beat + Second value noticeable higher than sucessing beat	Sum up both values and divide by 2, create 2 equal beats	(Gamelin et al. 2006, 2008, Giles et al. 2016)
Type 3	Single high value followed by single low value	First value noticeable higher than precedent beat + Second value noticeable lower than sucessing beat	Sum up both values and divide by 2, create 2 equal beats	(Gamelin et al. 2006, 2008, Giles et al. 2016)
Type 4	Undetected beats	Value 2 to 5 times higher than neighbouring values	Divide by 2-5 and create 2-5 equal beats	(Gamelin et al. 2006, 2008, Giles et al. 2016, Lensen et al. 2017)

Type 5	Falsely detected additional R-waves during a single beat	Several noticeable low values in a row	Sum up and replace by a single beat	(Gamelin et al. 2006, 2008, Giles et al. 2016)
Delete	High number of undetected beats	Value more than 5 times higher than neighbouring values	Delete value	(Schöberl et al. 2015)
Consecutive	Three or more identical values	Value difference of three or more IBIs = 0	Delete all but one value	(Jonckheer-Sheehy et al. 2012)

2.4.2. Tympanic temperature

Tympanic membrane temperature was measured with an ear thermometer (Pet-Temp® Ear Thermometer, Advanced Monitors, USA) (fig. 4) following the instructions given by the manufacturer. Each ear was measured twice with only values >36.9 °C deemed reliable (Pratsch et al. 2018) The higher value was then selected for further analysis. To analyze asymmetric thermic reactions, the temperature difference between the right ear minus the left ear (delta tympanic temperature) was calculated. In addition to tympanic membrane temperature, rectal temperature was measured as well using a rectal thermometer (Microlife® VT 1831, Microlife AG Swiss Corporation, Switzerland) (fig. 5). This was however only done as part of the examination and results were therefore not analyzed.

Fig. 4: Advanced Monitors Pet-Temp® Ear Thermometer

Fig. 5: Microlife® VT 1831 thermometer

2.4.3. Assessment by dog trainer and dog owners

After the completion of the group training sessions the dog trainer was asked to rate training success (the improvement in tolerance of handling by an unfamiliar person) on a 5-point scale ranging from 'no improvement' to 'very good improvement'.

2.5. Statistical analysis

Linear mixed models (LMM) were used to analyze HR and HRV and delta tympanic temperature by using the statistics software R (R Core Team 2018) and the "lme"-function of the “nlme” package (Pinheiro et al. 2013). Fixed effects for the LMM for evaluating training success were the group (training/control), visit (visit1/visit2) and time period (waiting room before examination/examination/waiting room after examination) as well as all interactions. In case of the tympanic temperature, we included the fixed effects group and visit as well as the group*visit interaction. Dog identity was always included as a random effect. To check whether LMM fulfilled the model assumptions, residual plots of all linear mixed effects models were obtained and inspected graphically for normality and homogeneity of variances. If an outlier (> 3x standard deviation) was identified, the analysis was repeated without it. In cases in which the reduced model resulted in different estimates or interactions compared to the original model, both are presented. Non-homogeneity of variances was found in two of the parameters (SDNN and RMSSD), therefore the dependent variable underwent a logarithm transformation. Graphs (boxplots) were generated with the ggplot 2 package of R and are always based on the original, unchanged data.

All other analyses were carried out with IBM SPSS 25. Comparisons of different aspects of training success and compliance were analyzed by using crosstabulations, McNemar’s tests and Chi^2-tests. Training success as assessed by the trainer and relationships with HR/HRV values were analyzed by using Spearman’s rank correlation coefficient.

3. Results

3.1. HR and HRV

Results of both HR and HRV show a significant difference in all 4 parameters (HR, RMSSD, SDNN, RMSSD/SDNN) regarding the time periods. During the examination dogs of both groups (TG and CG) had higher HR (tab. 4) and lower HRV (tab. 5) values compared to the waiting room periods (fig. 6). After correction for multiple testing using the Bonferroni method (four models: p-value considered to be significant 0.0125), no further significant effects were found. However, in the reduced model for HR the visit*period interaction indicates a tendency of both groups having higher HR in the waiting room before the examination at visit 2.

Tab. 4: Effects of training on HR values by group, visit and period (**bold** values represent **significance**, values *in italics* indicate *tendencies*)

	Mean heartrate Complete model		Reduced model[a]	
	Chi^2	p	Chi^2	p
Group	0.039	0.844	21.534	0.142
Visit	0.033	0.856	0.0585	0.809
Period	**442.87**	**<0.001**	**777.596**	**<0.001**
Group*Visit	0.281	0.596	0.1559	0.693
Group*Period	0.147	0.929	33.101	0.191
Visit*Period	43.67	0.113	*77.774*	*0.020*
Group*Visit*Period	12.26	0.542	14.801	0.477

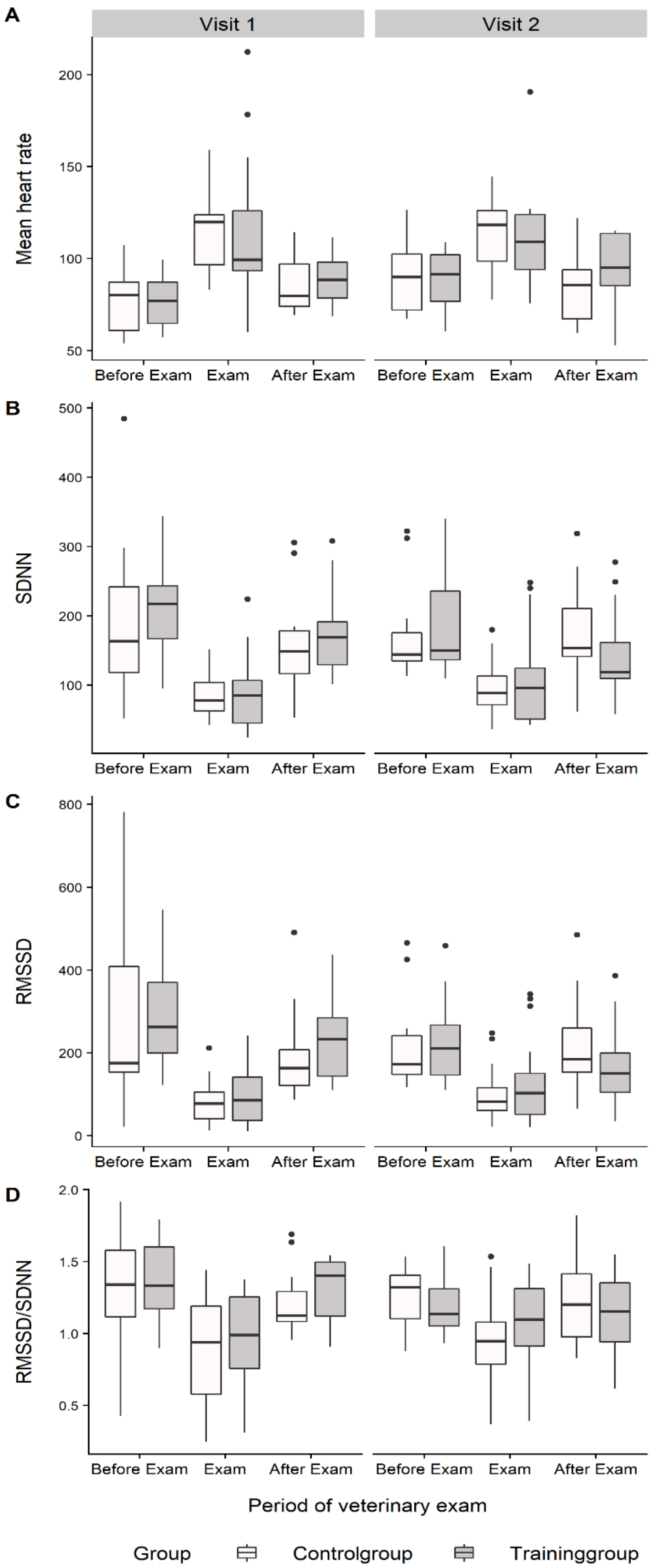
[a] 3 Values > 3 S.D. were excluded

Tab. 5: Effects of training on HRV values by group, visit and period (**bold** values represent **significance)**

	RMSSD[a]		SDNN[a]		RMSSD/SDNN	
	Chi^2	p	Chi^2	p	Chi^2	p
Group	0.015	0.903	0.073	0.787	0.138	0.711
Visit	17.27	0.189	0.743	0.389	0.683	0.409
Period	**410.43**	**<0.001**	**398.62**	**<0.001**	**215.12**	**<0.001**
Group*Visit	0.257	0.612	0.432	0.511	0.232	0.630
Group*Period	18.93	0.388	34.98	0.174	0.389	0.823
Visit*Period	0.836	0.659	0.479	0.787	12.29	0.541
Group*Visit*Period	31.67	0.205	37.08	0.157	22.02	0.333

[a] Dependent variable log transformed

Fig. 6: HR and HRV data of TG and CG from Visit 1 and 2



3.2. Tympanic temperature

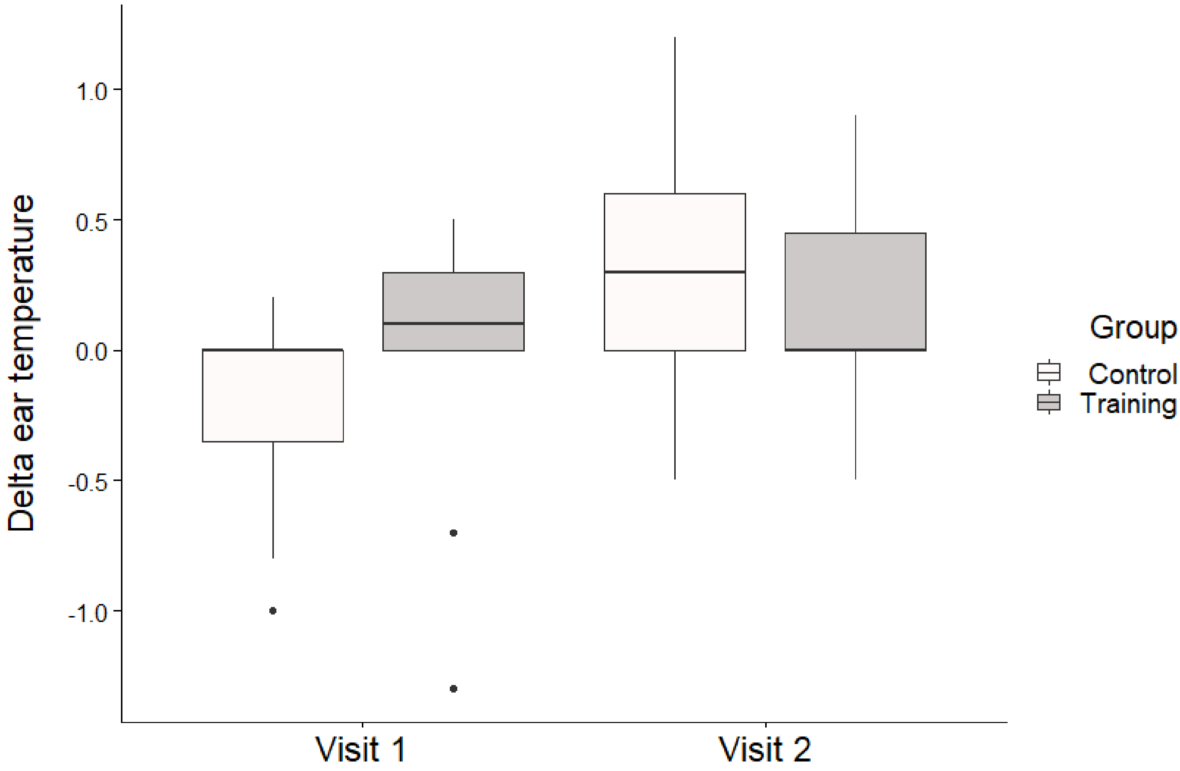
Tympanic temperature values show a significant difference in both main effects as well as in the interaction (tab. 6). The average CG dog showed no difference in tympanic temperature between ears during the first visit but during the second visit a higher temperature in the right ear of 75 % of the dogs was measured. Dogs in the TG on the other hand already showed a higher right-side temperature in 75 % of dogs during the first visit. Values measured during the second visit remained similar, yet the median was slightly lower and the 75th percentile higher.

Tab. 6: Effects of the training on the difference of tympanic temperature between the left and right ear (right minus left) at the end of the veterinary examination (**bold** values represent **significance)**

	Delta tympanic temperature		Reduced model [a]	
	Chi^2	p-value	Chi^2	p-value
Group	2.86	0.091	**57.62**	**0.016**
Visit	**12.36**	**<0.001**	**145.32**	**<0.001**
Group*Visit	**4.30**	**0.038**	**71.12**	**0.008**

[a] 1 value excluded

Fig 7: Delta ear temperature of CG and TG dogs at visit 1 and 2



3.3. Compliance of dogs

Whether dogs endured the total veterinary examination or the exam had to be cancelled because stop criteria were reached (i.e. struggling against examination or attempting to escape more than 3 times during the same examination step, acts of aggression or stop requested by owner) was used as a measure of compliance. During visit 1 there was no difference between the control and training group, whereas during visit 2 in the TG the examination was more frequently cancelled than in the CG (tab. 7). Also, within the training group it was found that during visit 2 the exam was cancelled more often than during visit 1 (tab. 7). In the control group, the frequency of a cancelled exam did not differ between the two examinations.

Tab. 7: Compliance during the examination (cancelled yes/no) and comparison of frequency of cancelling within the groups between visit 1 and visit 2 (**bold** values represent **significance)**

Group	Visit 1	Visit 2 Not cancelled	Cancelled	Comparison Visit 1 and 2 McNemar-Test **p-value**		Comparison Control vs. Training group Chi^2-Test **p-value**
Control	not cancelled	13	1	0.625	Visit 1	0.970
	cancelled	3	1			
Training	Not cancelled	9	8	**0.039**	Visit 2	**0.004**
	Cancelled	1	4			

3.4. Training success as assessed by trainer

At the end of the training (before the second visit) the trainer used standardized questions to assess the success of the training. According to the trainer's assessment, based on the training results of the last training session, almost 77 % of the dogs (17 dogs) showed a moderate to very good progress in training resulting in improved tolerance of handling by an unfamiliar person. 10 dogs (45 %) showed good to very good progress and 6 (27 %) of them very good progress. All 6 dogs with very good progress where classed as difficult to handle at the start of the training (tab. 8). Five other dogs (23 %) showed only slight progress, four of them allowed touching and examination without any problems already at the beginning of the training.

All six dogs in the training group that showed very good improvement in tolerance of handling by an unfamiliar person according to the trainer reached the stop criteria in the 2nd veterinary examination due to their behavior (resistance to the examination/attempts to jump off the table/descending from the target) (tab. 8). In total, only one animal owner had actively taken over the communication with the veterinarian and demanded the veterinary examination to be cancelled. Two other owners asked for breaks. In all other cases, the dogs showed behavior such as struggling or attempting to jump of the table that led to cancellation of the exam by the blinded vet team. This occurred, with one exception, always when measuring the rectal temperature.

Tab. 8: Training success rated by the trainer before the second examination portrayed by cancellation during the second examination

		Improvement in tolerating handling by unfamiliar person as rated by the trainer after the last training session					Total		
		No	slight	moderate	good	Very good		Chi^2	p-value
At start of training									
Difficult to handle	N	0	0	1	0	6	7	21.383	0.002
Inter-mediate	N	0	1	4	2	0	7		
Good to handle	N	0	4	2	2	0	8		
Visit 2									
Exam not cancelled	N	0	3	5	2	0	10	7.365	0.061
Exam cancelled	N	0	2	2	2	6	12		
Total	N	0	5	7	4	6	22		

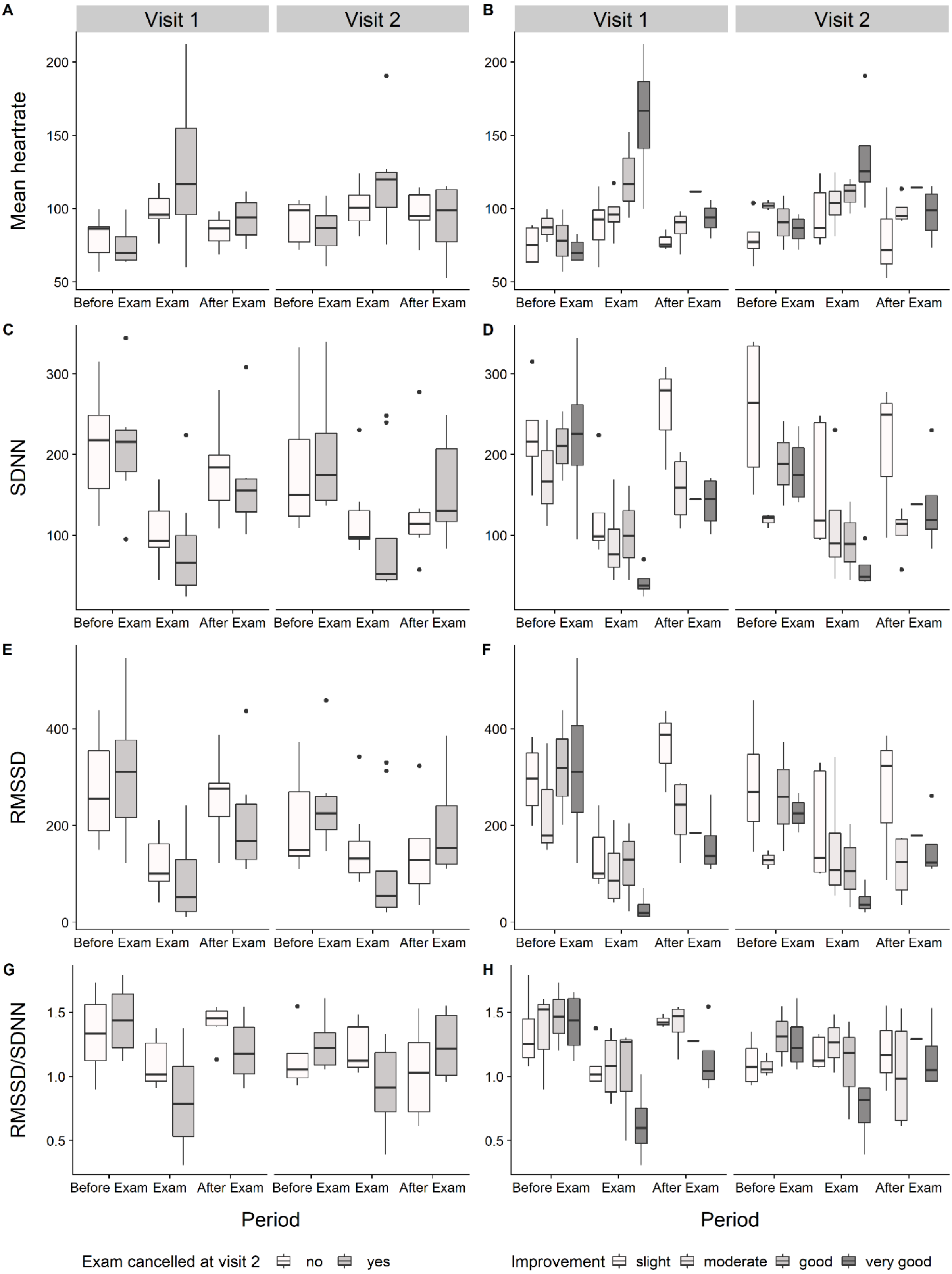
3.5. HR/HRV in the training group by training success and cancellation of exam during visit 2

To explore differences in the development of HR/HRV measures within the training group Spearman correlations with training success (trainer assessment of improvement) were calculated and figures based on training success and the cancellation of the second exam are presented (fig. 8)

The trainer's assessment of the improvement in tolerance of handling was correlated with the HR/HRV measures recorded during the veterinary examination in visit 2 and with the differences in HR/HRV measures between the 1st and the 2nd visit. Dogs with a stronger improvement had a lower HRV in the 2nd examination than dogs with less improvement (SDNN: $r_s = -0.65$, $p = 0.008$, $N = 15$; RMSSD: $r_s = -0.62$, $p = 0.014$, $N = 15$; RMSSD/SDNN: $r_s = -0.41$, $p = 0.130$, $N = 15$). Furthermore, a stronger reduction in HR compared to the 1st exam was found to be related to improved tolerance of handling (Delta HR (visit 2 – visit 1): $r_s = -0.52$, $p = 0.049$, $N = 15$). All other HR/HRV measures during the veterinary examination did not significantly correlate with improvement in tolerance of handling (HR visit 2: $r_s = 0.36$, $p = 0.184$, $N = 15$; Delta SDNN: $r_s = -0.31$, $p = 0.254$, $N = 15$; Delta RMSSD: $r_s = -0.33$, $p = 0.236$, $N = 15$; Delta RMSSD/SDNN: $r_s = -0.03$, $p = 0.911$, $N = 15$).

Looking at the figures depicting HR/HRV within the TG there are some eye-catching differences between visit 1 and 2. For HR, those dogs that did not comply at visit 2 (cancelled the exam prematurely) and in particular those that were assigned a very good improvement in tolerance of handling had reduced HR during the exam at visit 2 (see also results of correlation analyses). Looking at SDNN and RMSSD it is noticeable that dogs that did endure the full exam at visit 2 started at lower base levels and do not show the same increases in SDNN and RMSSD in the waiting room period after the exam compared to visit 1 before training. Changes in HRV after the training phase are also visible in the RMSSD/SDNN ratio where compliant dogs at visit 2 do have an almost inverse pattern with RMSSD/SDNN ratio rising during the veterinary exam. This pattern is present in particular in dogs with only slight or moderate improvement in handling.

Fig 8: HR and HRV data of TG dogs, grouped by cancellation at visit 2 and training success (assessed by trainer)



4. Discussion

4.1. Overview

On the one hand, our results show no clear effect of the cooperative care training on the dogs' HR or HRV during the three periods of the exam, which are measures sensitive to short-term changes in affective state. On the other hand, differences between the right and left tympanic membrane temperature measured after the veterinary examination, thus representing a summary measure of the waiting room period before the exam and the actual exam, point towards initial group differences, differences between the two visits and a different development of the two groups.

Further, HR/HRV results clearly contrast the evaluation done by the trainer and dog owners (Schützinger in prep). The trainer as well as the majority of owners described the outcome of the training with varying degrees of success. This discrepancy between the HR/HRV data and the perception of success by the people involved in the training process leads to the question of where this discrepancy originates from and how these results should be interpreted.

4.2. HR/HRV

First, we would like to discuss a very clear effect of the time period during the veterinary visits. During the examination, dogs of both groups showed a higher HR and a lower HRV compared to both waiting room periods. HR indicates general levels of arousal with any kind of excitement, be it positively or negatively associated, causing it to increase (Inagaki et al. 2004, Giuliani et al. 2008, Lensen et al. 2017, Ogden et al. 2019). HRV on the other hand shows the ability of an organism to cope with stressful situations, for instance a higher HRV value is positively connected to higher levels of flexibility and resilience towards stress in humans (Carnevali et al. 2018). In dogs, higher HRV levels and a stronger vagal activity, represented by RMSSD, as well as RMSSD/SDNN as an overall indicator for the vago-sympathical balance (Electrophysiology 1996, Langbein et al. 2004, Berntson et al. 2005, Wang und Huang 2012, Laborde et al. 2017), have been negatively correlated with the display of appeasement gestures (Kuhne et al. 2014). Conversely lower values and a weaker vagal influence show an overwhelmed organism which lacks the resources to adaptively cope with the situation it finds itself in.

Looking at our results, both groups have a significantly increased HR during the examination room period compared to both waiting room periods, showing a significant higher level of arousal during this time (fig. 6). Furthermore, RMSSD, SDNN and RMSSD/SDNN show significant decrease during the examination. The increase in arousal therefore seems to be linked to a situation they were not able to cope well with and thus likely a negative emotional state. This leads to the conclusion that both groups presumably experienced more pronounced strain while being in the examination room compared to both waiting room scenarios, making the examination the most stressful part of the veterinary visit.

4.3. Tympanic Membrane Temperature

Second, the results of the tympanic membrane temperature data show significant differences in both main effects as well as in the interaction (fig. 7). The temperature of the tympanic membrane can be used as an indicator for brain activity, as brain temperature is majorly determined by cerebral arterial blood temperature (Hayward und Baker 1969) and the distant cortical regions as well as the tympanic membrane share the same arterial circuits (Boyce et al. 2002). Cerebral blood flow is considered to be highly sensitive to arterial hypocapnia (Fan et al. 2008), therefore hyperthermia-induced hyperventilation which leads to subsequent hypocapnia (Wilson et al. 2006, Brothers et al. 2011) severely reduces cerebral blood flow. Such cerebral hypoperfusion is reported to lead to a lack of heat removal from the brain and therefore increase its temperature (Tsuji et al. 2016). This heat removal stems from the fact that the head tends to produce large amounts of heat because it consumes about 30 % of total available energy, while only representing roughly 5 % of body mass. This excess heat is dissipated by radiation through the skull as well as blood circulation. These cooling mechanisms affect the middle ear in the same way, therefore decreasing ear temperature with an increased perfusion (Cherbuin and Brinkman 2004). Conversely, this further underlines the fact that hypocapnia, which can be caused by rapid breathing, can lead to an increased ear temperature that correlates with increased brain activity. Stress-induced panting or hyperventilation could therefore lead to an increase in brain, and subsequently tympanic, temperature which could be used as an indicator for stress. Regarding hemispheric activity, the "Valence theory" suspects both hemispheres of the brain to be responsible for different emotional states respectively (Sackeim et al. 1982). While the left hemisphere seems to be more active during the processing of positive emotions, negative ones seem to be primarily handled by the right hemisphere (Silberman and Weingartner 1986, Canli et al. 1998). This was studied and verified in humans and non-human primates (Parr and Hopkins 2000, Boyce et al. 2002). In humans, studies that tried to associate tympanic temperature asymmetries with behavior in 4-8-year old children found a higher left side temperature to be associated with positive behavior, while a higher right-side temperature was linked to problematic and negative behavioral patterns (Boyce et al. 2002). Furthermore, positive emotions have been observed to be more readily expressed on the right side of the face, whereas negative emotions are more prominent on the left side (Schiff und MacDonald 1990), with the contralateral side of the brain being responsible respectively. Similar to the correlation of cortisol levels, behavior and EEG in rhesus monkeys (Kalin et al. 1998), marmosets and domestic cats with stress related cortisolaemia showed a higher right ear temperature compared to those with lower cortisol levels (Mazzotti and Boere 2009, Pereira et al. 2019, 2020). The Valence theory was further investigated in other non-human vertebrates. The results support the theory's validity in the surveyed species, including dogs, horses, chicken and sheep amongst others (Leliveld et al. 2013). The logical conclusion of these studies therefore seems to be that stress, as well as other negative emotional influences, seem to raise the tympanic temperature in the right ear. On the other hand, it is reported that stress actually leads to an increase of cerebral blood flow in the right prefrontal cortex (Wang et al. 2005), which should, given the prior explained means of cerebral temperature regulation, actually improve the cooling of this region. However, stress does, at least in a long-term measure, lead to an increased body temperature (Nelson et al. 2011). It is therefore possible that the natural cerebral cooling mechanism gets increasingly insufficient as peripheric blood temperature rises, or even contributes to the heat increase as a larger amount of warmer peripheric blood flows through certain areas.

Lateralized differences in the tympanic temperature holds potential for monitoring brain physiology regarding emotional states (Propper and Brunyé 2013). In our study, the average dog of the CG showed no differences in tympanic temperature between the right and the left ear during the first visit. However, in the second visit, around 75 % of dogs had a higher right-side temperature. This could indicate elevated levels of stress in these participants during the second visit, even though both visits happened in exactly the same way. A possible interpretation could be that visit 1 was perceived as negative and served as a form of conditioning for these dogs (Simpson 1997). Visit 2 exposing them to the same, already known situation, could therefore have triggered a higher stress response, as they anticipated what was going to happen already during the waiting period. This interpretation is supported by HR results: here the interaction of visit*period that almost reaches significance ($p = 0.02$; p-value considered significant = 0.0125) indicates that dogs of both groups had a higher HR during the second visit already in the waiting **room** before the exam. In contrast to the CG, dogs of the TG already showed a higher right-side temperature in roughly 75 % of dogs during visit 1. Although behavior scores reported by dogs' owners in the enrolment questionnaire were not significantly different, this result points towards a group difference in how the experience of the first veterinary visit was perceived. Measurements of visit 2 were similar to visit 1 in the TG, however with a slightly lowered median, and a higher 75th percentile pointing towards a mixed development in this group.

4.4. HR/HRV correlation with compliance and training success

Inspection of the associations between HR/HRV and both compliance and training success some patterns possibly explaining the different development of dogs within the TG appear (fig. 8). Six of the dogs that cancelled the second examination also belonged to the group of dogs with the highest training success and least tolerance of handling at the start of the study (fig. 8). While their training success was rated among the highest of all, they simultaneously show the highest stress profile of all TG dogs. Just as in visit 1, HRV values dropped during the examination, indicating their inability to cope with the situation, which then resulted in them jumping off the table. Another reason could be that they experienced frustration which can lead to suppression of parasympathetic activity (Lewis et al. 2004). They may have been frustrated by the differences between the training and the veterinary examination, e. g. the amount of restraint or the vet not stopping the exam when the dogs stepped down from the target. As these dogs showed the least tolerance of handling in the beginning of the study, they would have probably profited from a longer training duration.

Another visually noticeable difference in the development within the TG was that in dogs that did not cancel the examination during visit 2, SDNN and RMSSD values stayed on roughly the same level throughout the whole visit whereas in visit 1 they dropped down during the examination and increased again in the waiting room afterwards. The base level in visit 2 was lower than in visit 1, very likely due to them expecting what was about to happen but did not go down even more which would be expected in the case of them not being able to handle it. While this means that these dogs started with a lower level of vagal activity, it still shows that they were able to maintain it and therefore their ability to adapt to stress and not be overwhelmed by the situation they were negatively anticipating. This resulted in them being able to complete the examination. Furthermore, there is a seeming lack of recovery, as HRV values did not rise again in the waiting room afterwards, although this is most likely caused by the fact that they did not drop significantly to begin with and therefore no real recovery was needed. This aligns well with visible changes in the RMSSD/SDNN ratio of compliant dogs at visit 2 that have an almost inverse pattern with the ratio rising during the veterinary examination. To the extent that a higher RMSSD/SDNN ratio indicates higher vagal tone (Kuhne et al. 2014), this underlines the fact of compliant dogs being able to maintain their vagal activity levels and indicates that dogs that were already good or intermediate to handle at the start of the training (and therefore only had slight or moderate improvement of tolerance of handling) profited from the amount of training provided in this study.

4.5. Compliance and transfer of skills

In general, our HR and HRV data clearly shows that the veterinary examination itself is indeed the biggest source of fear and stress for dogs during a veterinary visit. This is in accordance with prior studies, naming the examination room, more precisely being on the examination table, as the most stress inducing part of a veterinary visit for dogs (Döring et al. 2009, Mariti et al. 2017). While the tympanic temperature measurements indicate that the CG and TG may have been different with regard to stress experienced during the exam from the start, and that the groups underwent a slightly different development between both visits, the analyses of the HR/HRV data do not confirm that a positive effect of the training procedure was consistently transferred to the veterinary context. The training effects could have been not strong enough in particular in those dogs that were more fearful and aroused at the start. Furthermore, HR/HRV data showed high variability and due to errors in IBI recordings that led to reduced sample size, small effects very likely could not be shown to be statistically significant. Further, the transfer of skills from the training to the veterinary context might have been poor. Nevertheless, the tympanic temperature between group analyses and visual inspection of the HR/HRV data within the training group support the idea of the training having differing effects on dogs based on their previous tolerance of handling during a veterinary examination and their progress made during the training.

Regarding changes in the overall compliance, TG dogs even had reduced compliance in the second visit, with more dogs withdrawing from the examination prematurely. Therefore, we cannot confirm our hypothesis that compliance was increased. However, in all dogs the examination was cancelled only at the last step of the procedure, which was taking the rectal temperature. As all other parts of the examination were tolerated this seemed to be the most invasive aspect in our examination. This aligns with results from other studies, in which taking the rectal temperature was linked to a rise in HR and less tolerated compared to other temperature measurement methods in dogs (Lamb and McBrearty 2013, Gomart et al. 2014).

The results of HR/HRV group comparisons and compliance stand in contrast to the training success as assessed by both trainer and owners (Böhm 2020, Schützinger in prep), who claimed to have observed an improvement in the dogs' behavior and tolerance of handling during training. Animal professionals can show a positive bias towards the animals they are working with, as found in the case of search dog handlers who scored their own dogs more favourably (Clark et al. 2020). Regarding owners, the idea that they perceive their pets emotional state in a way that may leave them unable to properly assess the stress level of their pet already came up in a study focusing on the assessment of dog welfare in a veterinary waiting room (Mariti et al. 2015). Another important aspect is the fact that the owners and the trainer of course knew the dog's group allocation. The absence of blinding can alter the results of a study (Gøtzsche 1996) and confirmation bias due to lack of blinding in animal behavior studies could be observed already (van Wilgenburg and Elgar 2013). Therefore, the possibility of bias in the owner and trainer ratings should be considered.

Looking at the physiology data of the TG suggests that there were some changes between visits, putative effects of training could have been dampened by difficulties in transferring the knowledge and skills gained during the training to the veterinary examination. There are a multitude of reasons why a putatively observed improvement during training by the owners and the trainer could not be transferred to the veterinary context. First, it could be caused by the fact that the veterinary examination was strictly standardized, whereas during training as well as in an examination conducted according to recommendations of low-stress handling guidelines the procedure should be adapted towards each individual dog and its behavior (Yin 2009). During training the increase of difficulty and the introduction of new challenges was adapted to individual dogs, but due to standardizing the veterinary examination in our study this was not possible. Furthermore, during the examination, due to the blinding procedure, the veterinary staff paid no attention to any behavior towards the target. If a TG dog wanted to signal a desired break by stepping off the target, this had to be communicated to the veterinary staff by the owner, which did not always happen. This could have led to growing fear or frustration in these dogs, as their attempts to communicate were ignored and they were not able to use their training experience to get more control of the situation (Bassett and Buchanan-Smith 2007). Compliance in the exam was found to be a factor relevant to dog owners regarding training success (Schützinger in prep). This could be a reason why most owners did not signal to the veterinarian to stop when the dog stepped of the target. Other circumstances surrounding the exam that differed from the training context could be further reasons for a worsened training outcome. While the training gave the dogs freedom of unrestrained movement and free choice to step on or off the target, the examination required them to be lifted on a table and to be slightly restrained by an unfamiliar person during the entire exam for security reasons. Being on the examination table is one of the most stressful parts of the examination for many dogs, who are often more comfortable on the floor (Döring et al. 2009, Yin 2009, Mariti et al. 2017). Although training included a table, the dogs were not restrained when on the table during training. This could be a reason for failure to transfer skills and still feeling uncomfortable during the examination. Restraint is highly stressful for animals and hence used as a standardized way to induce stress in laboratory animals (Buynitsky and Mostofsky 2009). Also in dogs restraint caused stress-related behavior and increases in heart rate (Beerda et al. 1998). Therefore, differences in the level of restraint between training and the examination could be a major reason for failure to show a clear training effect between groups. Another important factor that differed from the training was the role of the owners. They were mostly by-standers during the examination, only allowed to interact with their dogs by giving them treats at specific times or petting and talking to them during requested breaks. Owner-dog interactions can improve wellbeing during veterinary examinations, as dogs that are being held by and allowed to interact with their owner have been reported to be less stressed (Csoltova et al. 2017). In this study, signs of stress in dogs were compared during a standardized clinical examination during which the owners were either present but passive or owners held the dog on the table and talked to it. Dogs held by the owners made significantly fewer attempts to jump off the table and exhibited a significantly lower heart rate and a significantly lower eye temperature (measured by a thermographic camera) than dogs held by an assistant, evidencing that the stress level was lower when they were held by their owner than when held by a stranger. The presence of an assistant/veterinary technician could therefore even be seen as a hindrance and counterproductive to achieving a low-stress examination. Owner-dog interactions play an important role in the training procedure, so the sudden lack thereof could have resulted in the dogs not showing trained behavior as well as in increasing the dogs' discomfort to similar levels as experienced in visit 1. Finally, the part of the veterinarian in this study was performed by a male. Animals can perceive androgens and react with a physiological stress response to unfamiliar males (Sorge et al. 2014). Therefore, an unfamiliar male person performing the veterinary examination could have had a major impact on the dogs' emotional state.

4.6. Conclusion

Taking into account all of these points, we conclude that our training approach was received with varying degrees of success. Where some dogs showed a noticeable improvement during their second examination, others did not. The sticking point seems to be how well the dogs were able to transfer their new skills into the context of the veterinary examination, which was also strongly influenced by the level of owner-dog communication. Therefore we propose the idea that an optimized version of the training that involves the veterinary staff and context in the training process as well as an optimized and more personalized version of the veterinary examination, including more freedom to use trained skills and to engage in owner-dog interactions, very likely will lead to a better training outcome and a more comfortable examination situation for dogs.

5. Summary

One way to reduce fear and lack of compliance during veterinary procedures is ‘cooperative care training’, training animals to voluntarily participate in husbandry and medical care. Our hypothesis was that this form of training has observable effects on heart rate (HR), heart rate variability (HRV), tympanic membrane temperature and compliance of dogs in a veterinary examination.

A blinded controlled trial with 40 dogs (training group (TG): 22; control group (CG): 18) was carried out. Dogs and their owners took part in a standardized veterinary visit twice (visit interval: 140 ± 23 days). In between, the TG took part in cooperative care training (10 ± 2 group training sessions, additional training at home).

Results show that HR was higher and HRV lower during the veterinary examination compared to the waiting room, indicating that the examination was more stressful. There were however no significant effects of group in HRV/HRV between both visits, although dogs that started with a high compliance already seemed to be able to improve in maintaining their vagal activity. Tympanic membrane temperature measurements, taken at the end of each examination, on the other hand resulted in significant interaction of group and visit: during the second visit, stress levels in CG dogs appeared increased, whereas in the TG mixed effects were observed. Compliance in TG dogs during the second visit was lower, owners and trainer however claimed observable improvement of behavior during training.

In conclusion, transfer of trained skills to the veterinary examination performed by a team blinded to the group allocation was poor. This seems especially the case for dogs that had a lower compliance to begin with. Further research to optimize training outcomes is needed.

6. Zusammenfassung

Eine Möglichkeit, Angst und mangelnde Kooperation während tierärztlicher Untersuchungen zu reduzieren, ist ein sogenanntes „cooperative care training". Dieses zielt darauf ab, Tiere so zu trainieren, dass diese freiwillig an medizinischen Abläufen teilnehmen. Unsere Hypothese war, dass diese Form des Trainings sichtbare Effekte auf Herzfrequenz (HR), Herzfrequenzvariabilität (HRV), Trommelfelltemperatur und die Compliance von Hunden während einer tierärztlichen Untersuchung hat.

Eine kontrollierte Blindstudie mit 47 Hunden (Trainingsgruppe (TG): 26; Kontrollgruppe (CG): 21) wurde durchgeführt. Hunde sowie ihre Besitzer nahmen an zwei identischen standardisierten Tierarztbesuchen teil (Intervall zwischen beiden Besuchen 140 ± 23 Tage). In der Zeit zwischen diesen Besuchen nahmen TG Hunde an einem „cooperative care training" teil (10 ± 2 Trainingseinheiten, zusätzlich Training zu Hause).

Die Ergebnisse zeigen, dass während der tierärztlichen Untersuchung HR höher und HRV niedriger waren als während des Aufenthaltes im Wartezimmer. Dies deutet daraufhin, dass die Untersuchung deutlich negativer wahrgenommen wurde als beide Wartezimmerperioden. Es gab jedoch keine signifikanten HR/HRV Unterschiede zwischen beiden Besuchen hinsichtlich der Gruppenzugehörigkeit. TG Hunde, die bereits vor dem Training eine bessere Compliance aufwiesen, schienen jedoch in der Lage zu sein, ihr Level an parasympathischer Aktivität während des zweiten Besuches konstanter zu halten. Trommelfelltemperaturwerte, welche jeweils am Ende einer Untersuchung gemessen wurden, hingegen brachten signifikante Ergebnisse: der Stresslevel während des zweiten Tierarztbesuches schien in CG Hunden erhöht, wohingegen in der TG gemischte Effekte beobachtet werden konnten. Die Compliance von TG Hunden war während des zweiten Besuches vermindert, Hundebesitzer und Trainer berichteten jedoch von einer wahrnehmbaren Verbesserung des Verhaltens während des Trainings.

Insgesamt konnten die im Training erlangten Fertigkeiten nicht gut auf die tatsächliche tierärztliche Untersuchung, welche von hinsichtlich Gruppenzugehörigkeit geblindeten Personen durchgeführt wurde, übertragen werden. Dies scheint vor allem auf Hunde zuzutreffen, die zu Beginn eine niedrigere Compliance aufwiesen. Es bedarf daher weiterer Forschung, um den Trainingseffekt zu optimieren.

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8. Figure List

9. Table list

10. Abbreviation list

CG	Control group
TG	Training group
HR	Heart rate
HRV	Heart rate variability
RMSSD	Root Mean Square of Successive Differences
SDNN	Standard Deviation of the NN Intervall
IBI	Interbeat interval