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Is the addition of spray homeopathic in feed able to change dog behavior?

A adição de spray homeopático na ração é capaz de alterar o comportamento de cães?

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ABSTRACT: This study aimed to determine if the homeopathic formulation based on *Natrum muriaticum*, Matricaria, *Chamomilla*, and *Ignatia amara* was effective for stress control in dogs in order to prevent and/or reduce the frequency of unwanted behaviors. Ten male beagle dogs were divided into two groups, with five animals per group. The control group was animals receiving 0.5 mL/day of placebo via spray in the feed for 30 consecutive days as a preventive. The treated group received 0.5 mL/day of tested drug spray in the feed for 30 consecutive days as a preventive. Behavioral tests were performed at two time points, before and after receiving the tested drug. We performed the following behavioral tests: a) persons known and unknown in the internal and external environment; b) interaction with toys; c) interaction with an unknown dog, d) test with a leash; and e) firecracker test. There was more engagement in digging and scaling the fence in the control group than in the treatment group ($P < 0.05$). The animals that received homeopathic medicine sat 2.54 times longer than the control ($P = 0.0269$). Dogs that received homeopathic showed 4.49 times more interest in general at D30 than at D1 ($P = 0.0214$). Animals in the treated group showed an 83% less chance of urinating than the control group in the tests involving displacements. We conclude that the preventive addition of homeopathic medicine containing *Natrum muriaticum*, *Chamomilla*, and *Ignatia amara* reduced undesirable behaviors related to anxiety and increased the incidence of interest behavior.

KEYWORDS: Anxiety; Animal behavior; Canine; Homeopathy.

RESUMO: Este estudo visou determinar se a formulação homeopática baseada em *Natrum muriaticum*, Matricaria, *Chamomilla*, e *Ignatia amara* foi eficaz para o controle de stress em cães, a fim de prevenir e/ou reduzir a frequência de comportamentos indesejados. Dez cães beagle machos foram divididos em dois grupos, com cinco animais por grupo. O grupo controle recebeu 0,5 mL/dia de placebo via spray na ração durante 30 dias consecutivos, como preventivo. O grupo tratado recebeu 0,5 mL/dia de homeopático via spray na ração durante 30 dias consecutivos, como preventivo. Os testes comportamentais foram realizados em dois momentos, antes e depois de receber o fármaco testado. Foram realizados os seguintes testes comportamentais: a) pessoas conhecidas e desconhecidas no ambiente interno e externo; b) interação com brinquedos; c) interação com um cão desconhecido, d) teste com a guia; e teste com fogos de artifício. Houve um aumento nos comportamentos cavar e escalar a grade no controle ($P < 0,05$). Os animais que receberam o homeopático sentaram-se 2,54 vezes mais do que o grupo controle ($P = 0,0269$). Os cães que receberam homeopático mostraram 4,49 vezes mais interesse em geral no D30 do que no D1 ($P = 0,0214$). Os animais do grupo tratado mostraram uma probabilidade 83% menor de urinar do que o grupo controle nos testes envolvendo deslocamento. Concluímos que a adição preventiva de medicamentos homeopáticos contendo *Natrum muriaticum, Chamomilla*, e *Ignatia amara* reduziu os comportamentos indesejáveis relacionados com a ansiedade e aumentou a incidência de comportamentos de interesse.

PALAVRAS-CHAVE: Ansiedade; Cães; Comportamento Animal; Homeopatia.

INTRODUCTION

Gray wolves (*Canis lupus*), the ancestors of dogs, are responsible for bringing dogs close to humans. Since pet ownership expanded in Brazilian homes in the 19th century, man-animal interactions have contributed to human happiness (CROSSMAN, 2017). Brazil ranks fourth for pet ownership worldwide (IBGE, 2013). In 2018, Brazilian homes housed 55.1 million dogs (INSTITUTO PET BRASIL, 2019). In contemporary society, pet dogs are increasingly left alone within restricted environments, which means they are

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unable to express natural behaviors (CROSSMAN, 2017). Isolation can result in undesirable behaviors such as anxiety, aggression, fear, excessive barking, and destructive behavior. As a result of these undesirable behaviors, owners may eventually abandon their dogs. Homeopathy companies have been producing medicines designed to temper these behaviors. Homeopathy is already used in human medicine for this purpose (MITTRING-JUNGHANS et al., 2021; MISHRA et al., 2021), and the concept has been extrapolated to pets.

Homeopathy is a system of curing all curable diseases, whether in humans or animals, by agency of small doses of medicines. When these medicines are exhibited in large and repeated doses, they are capable of producing in a healthy body, symptoms similar to those produced by the disease in a sick body, in brief this principle is expressed as similia similibus curantur i.e. like is cured by like (RUSH 2007). Homeopathic medicines include substances from plants, animals or minerals that have known therapeutic effects for particular purposes (HIGUERA-PIEDRAHITA et al., 2020). These substances are diluted several times and are indicated for preventive and curative uses, with the general objective of developing immunity and inhibiting the disease (ŞENEL, 2019). Homeopathy has been an alternative to antimicrobials, as it does not leave residues in animal tissues or the environment (BENEZ et al., 2004).

In this context, among the homeopathic medicines we can mention *Natrum muriaticum*, which is sodium chloride; has been used in humans to treat behavioral disorders (SAREMBAUD, 2017). In addition, *Chamomilla* is a medicinal plant used to relieve emotional stress (PINTO et al., 2008) and as a tranquilizer, anti-inflammatory, and aromatherapy (UBESSI et al., 2019). Another substance used as homeopathic medicine is *Ignatia amara* is a phytochemical extracted from the seeds of the plant *Strychnos ignatii*. It is used for symptoms related to anxiety in humans, in addition to being a sedative (MARZOTTO et al., 2012). It also showed positive effects in the therapy of respiratory diseases (ROSSI et al., 2009). Against this background, the objective of the present study was to determine whether a homeopathic formulation based on *Natrum muriaticum*, *Chamomilla,* and *Ignatia amara*, indicated to reduce stress in dogs, would prevent or reduce the frequency of undesirable behaviors.

MATERIALS AND METHODS

Homeopathic product

The medicine Orgacalm was formulated according to the recommendations of the Homeopathic Pharmacopeia. It is intended to improve adaptive conditioning in the face of stressful situations. The homeopathic medicine includes *Natrum muriaticum* 30CH, *Chamomilla* 30CH, *Ignatia amara* 30CH, and vehicle 30 mL (aqueous).

Animals, accommodation, and feed

Ten male beagles with an average weight of 10.2 ± 0.64 kg and two years of age were used. They were distributed in a completely randomized design for two groups. The dogs were purchased from a commercial breeder when they were 60 days old and were subsequently housed in an experimental kennel at the State University of Santa Catarina-UDESC. These animals were closely related, as they are the pups of the same father, but were the mothers different. The difference in delivery dates was 3 days. This was done to reduce the variability between groups.

During the day and night, the dogs were divided by treatment in two collective kennels (3 x 4 m), housed twice a day in individual kennels (1 x 1 m) for feeding. The collective kennels were connected to external shaded spaces with gravel surfaces where they accessed during the day.

The dogs received water ad libitum and 300 grams of commercial dog food divided twice a day (7:30 am and 5:00 pm). The experimental period lasted 30 days. The homeopathic medicine and placebo were sprayed on the food at 0.5 mL/day divided twice a day. For the control (CO), five dogs received a placebo via spray in the feed for 30 days. The treatment group (TRA) included five dogs that received Orgacalm (registered patent 916829944) via spray in the feed for 30 days.

Behavioral variables

Behavioral analyses were performed at two time points: before receiving the homeopathic medicine (D1) and 30 days after receiving the homeopathic medicine (D30).

Known and unknown person in the kennel's internal environment

Cameras for recording behavior were installed inside the kennel. The dogs were kept in individual kennels (n = 10) for testing in an indoor environment. First, a known person (who fed the animals) passed in front of the individual kennels without stopping in front of the kennel, talking, making sounds or gestures, and not engaging in physical contact with the animals. This procedure was carried out for two minutes, allowing the person to pass an average of 11 times in front of each kennel. After this first stage, this person performed the same actions in the indoor environment, this time calling the dogs' names (eye and physical contact) for two minutes, passing in front of the same kennel three times. Then, an unknown person walked the same path in front of the kennels for two minutes (11 times in front of each kennel) without engaging in visual or physical contact with the animals.

The following behaviors were analyzed: lying down, resting on the fence, jumping, jumping to another kennel, licking, digging, barking, crying, wagging the tail, climbing the fence, urinating, yawning, sitting, sticking the tongue out,

and sniffing. These behaviors were recorded on video for detailed analysis.

Unknown dog in the kennel

The fourth indoor test was performed using an unknown castrated female, who circulated in front of the individual kennels for two minutes. The following behaviors were analyzed: supporting on the fence, jumping, jumping to another kennel, licking, digging, barking, crying, wagging the tail, climbing the fence, urinating, yawning, sitting, sniffing, sticking tongue out, and attacking.

Interaction of dogs with toys

The dogs were allocated to the collective kennels in the external environment. Each group received five 600 ml pet bottles (n = 10, one per animal) to observe the dogs' interaction with the toys. This test was filmed for ten minutes from the moment the pet bottles were made available. It is important to note that, during this period, no person was in view of the dogs.

The following behaviors were analyzed: urinating, yawning, sitting, sniffing, and attacking. In this case, the interest in the toy was measured on a scale, and the approach time to the toy was measured.

Unknown person in the external environment

After 30 min of testing with the toy, other tests were carried out with the dogs remaining in the external environment: A) Two unknown people passed in front of the kennel without making any noise. This test was recorded for two minutes. We waited 30 minutes and started another test. B) The same people passed again in front of the kennel, this time making noise to attract the dogs' attention. The event was also filmed for two minutes. In both situations, the following behaviors were analyzed: leaning on the railing, digging, barking, crying, sitting, sniffing, attacking, wagging the tail, sticking the tongue out, interest in the action, and walking through the kennel.

Test with a known person in the external environment

This test was similar to that described in section 2.2.4. However, now two well-known people passed in front of the kennel without making any noise. This test was filmed for two minutes. Again we waited 30 min, and these same people would pass in front of the kennel, talking and interacting with the dogs. This event was also recorded. The following behaviors were analyzed at both times: leaning on the fence, digging, barking, crying, sitting, sniffing, wagging the tail, sticking tongue out, climbing the fence, interest in the action, and walking through the kennel.

Test with dogs on a leash

For this test, an adaptation period was first performed with the dogs, as they had never been led on leashes. After this adaptation period, each animal individually used the leash to walk 25 meters in a straight line. At the height of 20 m, there was another dog (an unknown castrated female) restrained by a leash with its guardian. The animal's behavior was recorded during the journey, and we recorded the time it took for each animal to make this journey. The following behaviors were recorded: lying, jumping, barking, crying, urinating, attacking, interest in the other dog, pulling on the leash, fear, running, playing, and the length of the walk.

Sound test: use of a firecracker

Two tests were carried out using firecrackers, sound stress on dogs that is prohibited in many Brazilian municipalities because of the stress it causes. In the first test, the animals were housed inside the kennel in the collective pens. We waited 10 minutes for the dogs to be calm, then a firecracker was exploded at a distance of 20 meters from the kennel; for five minutes, the dogs' behavior was filmed with the internal cameras. We waited for approximately 1 h, and this same procedure was performed outdoors. The dogs' behavior in both situations was grouped into characteristics defined as "indifferent," "afraid," "brave," and "agitated," as detailed in the Supplementary Material.

Statistical analysis

Various analyses were used depending on the variables' characteristics. All analyses were performed using SAS OnDemand (2012). Two continuous variables were analyzed using analysis of variance (interaction time seconds and walk duration). In contrast, other discrete variables (supporting on the fence, jumping, licking, digging, barking, crying, climbing the fence, attacking, and playing) were measured, such as counts. They were analyzed using Poisson regression because these values were not distributed normally. The treatment, the time of measurement, and interaction between treatment and time were considered fixed effects, while the test was used as a random effect, in addition to considering repeated measures for animals whenever necessary.

Categorical variables (hanging tongue out, sitting, and sniffing) were analyzed using logistic regression, always considering the probability of the behavior occurring with the non-use of homeopathic as a reference category for treatment and Day 1 as a reference category for the moment. Variables measured on a Likert scale (wagging tail, urinating, walking through the kennel, pulling on the leash, and displaying interest) were analyzed using ordinal logistic regression, which uses a cumulative logit model that looks like a binary logistic regression model (AGRESTI, 2007) separating the scores (ordinal data) into two categories: lower and higher

score values. We considered the probability of higher score values using the reference categories for treatment and time as in the logistic regression described above.

The variables related to the animals' behavior with the firecracker test (with firecracker) were analyzed using the Chi-square test, which verifies the existence of an association between categorical variables. When more than 20% of the cells in the contingency table counts of less than 5, the assumption of the Chi-square analysis, Fisher's exact test was applied, and when significant, the standardized adjusted residues for each cell were used to verify the differences between proportions (MEYERS et al., 2013).

Analysis of normality of the residues was performed using PROC UNIVARIATE of SAS together with the presence of outliers. Analysis of variance, Poisson regression, and logistic regressions were analyzed using PROC GLIMMIX from SAS using the appropriate distribution for each variable. Fisher's exact test was performed using the PROC FREQ of the SAS. Statistical significance was considered at the level of 5% probability.

RESULTS

No significant interactions between time and treatment were found for any of the variables displayed in Table 1. The treatment group was significant for "Jumping" (P = 0.0083; Table 1); that is, these animals jumped more often. For the "Digging" behavior (P = 0.0246; Table 1), the animals in the control group dug more often. For the "Scaling Fence" behavior (P = 0.0350; Table 1), the control group's animals climbed more often. For "Crying" (P <0.0001; Table 1), the animals at D30 cried more often than at D1.

No interaction between time and treatment was found for any of the behavioral variables displayed in Table 2. For the "Sitting" behavior, the treatment was significant (P = 0.0269; Table 2), suggesting that the animals in this group sat 2.54-fold or 154% more often than those who did not receive the homeopathic medicine. For the "Sniffing" behavior, the timepoint was significant (P = 0.0025; Table 2), suggesting that, at D30, this behavior was 8.78-fold more or 778% that of D1.

An interaction was observed for the variable "Interest" (P = 0.0214; Table 3), in which treated animals at D30 were 4.49 times more likely or 349% more likely to be interested than at D1. The treatment was significant for the variable "Urinating" (P = 0.0023; Table 3), in which animals in the treatment group were 83% less likely to urinate than the control group. The timepoint suggested that the animals walked 3.19 times more or 219% more often at D30 than at D1.

There was no general association between behavior and treatment (P = 0.1463; Table 4), behavior and treatment for the external firecracker test (EF) (P = 0.1949; Table 4) and internal firecracker test (IF) (P = 0.3713; Table 4), behavior and treatment for D1 (P = 0.1949; Table 4) and D30 (P = 0.3498; Table 4), and for test and behavior (P = 0.2186; Table 4). Only the timepoint (P = 0.0349; Tab 4) when the tests were applied resulted in an association with behavior; at D30, the animals were more indifferent, while at D1, they were braver.

DISCUSSION

Homeopathic medicine is provided to its patients based on the clinical symptoms of each individual; thus, homeopathy seeks to understand the patient's history, environment, and

Table 1. Adjusted means ± standard error of the mean for comparisons between treatments and moments using Poisson regression.

Variable	N	Treatment		Timepoint		P-value		
		No	Yes	D1	D30	Treat.	Time	Treat. x Time
Support Grade	160	4.2 ± 1.00	3.8 ± 0.93	3.7 ± 0.91	4.3 ± 1.03	0.4917	0.2304	0.3925
Jumping	120	1.5 ± 0.70	3.4 ± 1.41	2.4 ± 1.03	2.2 ± 0.95	**0.0083**	0.7585	0.5731
Licking	100	0.2 ± 0.10	0.3 ± 0.12	0.3 ± 0.12	0.2 ± 0.10	0.8221	0.5665	0.3914
Digging	160	0.18 ± 0.088	0.03 ± 0.023	0.07 ± 0.052	0.07 ± 0.045	**0.0243**	0.9648	0.4354
Barking	180	10.1 ± 4.96	10.4 ± 5.11	11.1 ± 5.44	9.5 ± 4.65	0.8237	0.2579	0.8170
Cryining	180	0.3 ± 0.16	0.4 ± 0.26	0.2 ± 0.10	0.8 ± 0.44	0.1282	**<0.0001**	0.3096
Scale Scale	140	1.0 ± 0.26	0.3 ± 0.14	0.6 ± 0.22	0.6 ± 0.19	**0.0350**	0.9395	0.4007
Attacking	60	0.2 ± 0.28	0.3 ± 0.43	0.3 ± 0.44	0.2 ± 0.27	0.1148	0.0756	0.9905
Interaction Time	20	160.6 ± 62.87	68.1 ± 62.87	69.2 ± 62.87	159.5 ± 62.87	0.3136	0.3249	0.6360
Throwing	20	0.7 ± 0.49	1.2 ± 0.64	1.0 ± 0.59	0.8 ± 0.56	0.5453	0.8735	0.8735
Tour Duration	20	27.0 ± 3.83	31.0 ± 3.83	29.5 ± 3.83	28.5 ± 3.83	0.3615	0.8173	0.9631

Note 1: Means and standard errors are retransformed values which can lead to values different from those calculated on the original data, except for "TempoInteracaoSeg" and "DuracaoPasseio", in which ANOVA was performed. The interpretation is the same as the comparison of traditional averages made by ANOVA. No significant interaction was found between time and treatment for any of the variables. Treatment was significant for "Jumping" (p = 0.0083) (animals that received the homeopathic jumped more often), "Digging" (p = 0.0246) (animals that did not receive the homeopathic digging more often) and "EscalarGrade" (p = 0.0350) (animals that did not receive the homeopathic climbed more often), while the moment was significant for "Crying" (p <0.0001) (at D30 the animals cried more often).

Table 2. Adjusted odds ratios and 95% confidence intervals obtained through logistic regression for treatments and timing.

Variable	N	Treatment	Odds (95% CL) [1]	Time	Odds (95% CL)[1]	P-value		
						Treatment	Time	Trat. X Mom.
Tongue out	160	Yes x No	0.92 (0.46 – 1.82)	D30 x D1	1.58 (0.79 – 3.14)	0.8029	0.1919	0.8029
Sitting	160	Yes x No	2.54 (1.15 – 5.63)	D30 x D1	1.84 (0.83 – 4.06)	**0.0269**	0.1165	0.7772
Sniffing	160	Yes x No	1.08 (0.34 – 3.42)	D30 x D1	8.78 (2.77 – 27.84)	0.8837	**0.0025**	0.0641

[1]Always modeling the chance of the behavior happening ("YES") with the category "No" being a reference for treatment and "D1" being a reference for the time. Note 2: When "Odds" reads probability or chance of something occurring (in case the behavior has happened). Here I will always be modeling the chance of the behavior occurring using the categories described at the bottom of the table as a reference for calculating the Odds ratio. No interaction between time and treatment was found for any of the variables. For "Sitting", the treatment was significant (p = 0.0269), indicating that the animals that received the treatment sat 2.54 times more (or 154% more chance of sitting, as another way of interpreting) compared to those that did not receive, that is, this behavior increases with the use of homeopathic. For "Smelling" the moment was significant (p = 0.0025), indicating that at D30 this behavior was 8.78 times or 778% concerning D1.

Table 3. Adjusted odds ratios and 95% confidence interval obtained through ordinal logistic regression for treatments and moments.

Variable	N	Treatment	Odds (95% CL)[1]	Time	Odds (95% CL)[1]	P-value		
						Treatment	Time	Treat. X Time
Wag your tail	160	Yes x Not	1.43 (0.21 – 9.79)	D30 x D1	1.10 (0.59 – 2.04)	0.7161	0.7645	0.8949
Urinate	140	Yes x Not	0.17 (0.06 – 0.53)	D30 x D1	0.47 (0.16 – 1.38)	**0.0023**	0.1677	0.1714
Walking through the kennel	60	Yes x Not	0.98 (0.22 – 4.40)	D30 x D1	3.19 (1.12 – 9.10)	0.9762	**0.0307**	0.9494
Force the guide to travel	20	Yes x Not	0.55 (0.03 – 10.89)	D30 x D1	0.41 (0.04 – 4.14)	0.6460	0.3900	0.4934
Interest	100	Yes x Not (D1)	0.42 (0.12 – 1.45)	D30xD1 (Yes)	4.49 (1.55 – 13.00)*	0.9982	0.0901	**0.0214**
		Yes x Not (D30)	2.39 (0.68 – 8.39)	D30xD1(No)	0.79 (0.28 – 2.18)			

[1] Always modeling the chance of obtaining higher score values, with the category "NOT" being a reference for treatment and "D1" for the moment. * Significant at 5% probability. Note 3: There was interaction for "Interest" (p = 0.0214), and the split indicated that the effect occurred only for the animals that received treatment, where at D30 these animals had 4.49 times more chance (or 349% more chance) of presenting higher values score (that is, having greater interest) compared to D1. The treatment was significant for "Urinating", and the animals that received the homeopathic had a 17% chance (0.17) to urinate or 83% less chance to urinate compared to not using the treatment (here we also read more chance of presenting higher score values). The moment was significant for "AndarPeloCanil" (p = 0.0307), indicating that the chance of animals presenting higher score values for this variable was 3.19 times (or 219%) in D30 compared to D1.

daily routines (ADLER et al., 2013). This approach is based on highly diluted extracts, components, and minerals. Homeopathy has been used in humans to treat diseases and disorders such as respiratory problems, dermatitis, depression, and stress, among others (DOSSETT et al., 2018). However, there are few studies in this area related to animal behavior.

Landsberg et al. (2018) reported that the most frequent signs related to separation anxiety in dogs are destructive behaviors, vocalization, excessive salivation, defecation, and inappropriate behavior when urinating. These researchers observed that dogs treated with fluoxetine had lower incidences of urination behavior than the placebo group. The authors attributed this effect to stress reduction, especially for dogs left alone for long periods (CHUNG et al., 2016). Dodman et al. (2018) also observed an increase in the frequency of urination behavior in dogs that were alone longer and had owners with emotional instability.

Marzotto et al. (2012) observed that *Ignatia* reduced anxiety and fear in mice. The authors suggested that *Ignatia* modulates emotional responses and possibly regulates urination behavior, a non-linear mechanism of regulation in the animals' central nervous system. *Ignatia*'s mechanisms of action in the central nervous system may involve the centers that control pain and anxiety, possibly through interaction with glycinergic receptors (MARZOTTO et al., 2012). These findings suggest that the homeopathic medicine may use both mechanisms, reducing the anxiety in the dogs in the face of stressful environments, consequently reducing the frequency of urination.

Several behaviors in this study are related to separation anxiety, including jumping, urinating, climbing a fence, crying, and sitting. These behaviors can occur as a result of inadequate socialization. In addition, experimental dogs are limited in their ability to socialize or become familiar with new situations and social and non-social stimuli (DODMAN et al., 2018) because these animals do not benefit from animal-owner bonds.

The homeopathic medicine tested was associated with less digging and fence-climbing behaviors that may be related to anxiety. The dogs that received the homeopathic medicine remained seated longer during the tests, which may reflect

Table 4. Absolute (and relative) frequencies and Fisher's exact test for the variables moment, treatment and tests related to the animals' behavior using firecrackers.

Variable	With Foguete Behavior				P-value[1]
	Abs. frequence (relative freq.%)				
General (n = 40)	Restless	Indifferent	Afraid	Brave	0.1463
Treatment					
No	0 (0.0)	8 (20.0)	9 (22.5)	3 (7.5)	
Yes	1 (2.5)	11 (27.5)	3 (7.5)	5 (12.5)	
FE Test (n = 20)	Restless	Indifferent	Afraid	Brave	0.1949
Treatment					
No	0 (0.0)	2 (10.0)	5 (25.0)	3 (15.0)	
Sim	1 (5.0)	5 (25.0)	3 (5.0)	3 (15.0)	
IF Test (n = 20)	Restless	Indifferent	Afraid	Brave	0.3713
Treatment					
No	0 (0.0)	6 (30.0)	4 (20.0)	0 (0.0)	
Yes	0 (0.0)	6 (30.0)	2 (10.0)	2 (10.0)	
D1 (n = 20)	Restless	Indifferent	Afraid	Brave	0.1949
Treatment					
No	0 (0.0)	3 (15.0)	5 (25.0)	2 (10.0)	
Yes	1 (5.0)	3 (15.0)	1 (5.0)	5 (25.0)	
D30 (n = 20)	Restless	Indifferent	Afraid	Brave	0.3498
Treatment					
No	0 (0.0)	5 (25.0)	4 (20.0)	1 (5.0)	
Yes	0 (0.0)	8 (40.0)	2 (10.0)	0 (0.0)	
Test (n = 40)	Restless	Indifferent	Afraid	Brave	0.2186
EF	1 (2.5)	6 (17.5)	6 (15.0)	6 (15.0)	
IF	0 (0.0)	12 (30.0)	6 (15.0)	2 (5.0)	
Time (n = 40)	Restless	Indifferent	Afraid	Brave	**0.0349**
D1	1 (2.5)	**6 (15.0)***	6 (15.0)	**7 (17.5)***	
D30	0 (0.0)	**13 (32.5)***	6 (15.0)	**1 (2.5)***	

[1]Teste exato de Fisher. Note 4: *Comparisons of proportions based on adjusted standardized residues, with values outside the range of -1.96 to 1.96 were considered significant based on the statistics of Z scores. EF: external firecracker; IF: internal firecracker

reduced anxiety. In addition, the animals in the treated group were more interested in knowing what was happening, and this can be associated with reduced fear. This quality is desirable for dog owners. The homeopathic medicine tested was also associated with a greater capacity for socialization with people, another desirable behavior.

King et al. (2014) found that individual personality types and life experiences can modulate how dogs react in stressful environments or situations, resulting in undesirable behaviors. Dogs are usually fearful, as they have had a negative experience related to fear. Tiira et al. (2016) observed that 26.2% of the dogs had fear reactions, more than 40% of the time concerning unknown people and new situations. In this context, anxiety can reduce exploration and locomotion; this can occur by retaining impaired memory related to gamma-aminobutyric acid receptors (MASHRA et al., 2021). The homeopathic people might act by increasing the recognition index, which increases exploration and free locomotion and improves the decline of spatial memory in animals due to an antioxidant effect on the brain (MASHRA et al., 2021). This phenomenon may have occurred in the present study, in which there was an increase in interest and sniffing in the homeopathic group.

Thus, rats that treated of a homeopathic medicine based on *Chamomilla* 6CH were able to return to their normal state faster when subjected to stressful conditions (PINTO et al., 2008). This phenomenon may be due to the calming effects of *Chamomilla*, which includes flavonoid and α-bisabolol (UBESSI et al., 2019), the substances responsible for the medicinal properties of this plant. The flavonoid apigenin found in *Chamomilla* decreases plasma cortisol concentrations (YAMADA et al., 1996). Reis et al. (2006) observed that *Chamomilla* in

beef cattle diets reduced stress and that this was due to its sedative, anxiolytic, and muscle relaxant properties. These authors found that *Chamomilla* reduced serum levels of adrenocorticotrophic hormone and cortisol. The increase in these hormones is related to emotional or physical stress. These findings may explain some of the less-expressed behaviors in the group that received the homeopathic in the present study.

Cracknell and Mills (2008) did not observe a significant difference in the behaviors of dogs supplemented with a homeopathic medicine as a response to fireworks. However, the owners observed an improvement in the dogs over time. This same response was observed in the present study, in which there was a difference between D1 and D30. This may be related to the dogs' adaptation mechanism, in which they may become indifferent to frequent noise.

CONCLUSION

We conclude that the homeopathic formulation based on *Natrum muriaticum*, *Chamomilla*, and *Ignatia amara* reduces behaviors such as digging, climbing the fence, and urinating, which are related to anxiety dogs. The consumption of a homeopathic medicine increased the frequency of "interest" behavior, which indicates tranquility on the part of the dog. These results are beneficial for dogs and their guardians because they are related to the welfare and socialization of these animals.

ETHICS COMMITTEE

The animal use research committee approved the State University of Santa Catarina's project, protocol number 6524030419.

DECLARATION OF COMPETING INTEREST

The authors declare no conflict of interest.

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Supplementary Material 1. Characterization of canine behaviors.

INDIFFERENT	When barking, he continues normal behavior before the test takes place
AFRAID	Ears down, tail between legs, looking scared
BRAVE	Barking, attentive ears, walking, tail high
RESTLESS	Running, walking side-to-side, barking or not, staying close to the fence