Journal of Veterinary Behavior 25 (2018) 56–64

Contents lists available at ScienceDirect

Journal of Veterinary Behavior

journal homepage: www.journalvetbehavior.com

Research

The effect of human interaction on guinea pig behavior in animal-assisted therapy

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A R T I C L E   I N F O

Article history:
Received 12 May 2017
Received in revised form
14 February 2018
Accepted 18 February 2018
Available online 26 February 2018

Keywords:
guinea pig
behavior
human-animal interaction
animal-assisted therapy
stress
enrichment

A B S T R A C T

Guinea pigs are included in various animal-assisted interventions (AAIs), but no research has been published to date on behavioral changes in guinea pigs interacting with humans. The goal of this study was to evaluate the behavior in guinea pigs during animal-assisted therapy (AAT) and to identify factors that influence their stress and well-being. Five guinea pigs were studied during 50 observations in a randomized controlled within-subject design with repeated measurement. All guinea pigs were tested under all the following conditions: (1) therapy setting with retreat possibility ($n = 20$), (2) therapy setting without retreat possibility ($n = 10$), and (3) control setting without human interaction ($n = 20$). Behavior was coded according to a specifically designed ethogram using continuous recording and focal animal sampling with The Observer® XT 12.5. The data were analyzed using generalized linear mixed models with SPSS®, version 22.0. Results show that the frequency but not the duration of hiding was significantly increased in the therapy setting with retreat possibility compared to the control condition. During therapy with retreat possibility, the number of comfort behavior episodes stayed constant, while the number of startling and explorative behavior and the duration of locomotion increased significantly in comparison to the control setting. During therapy without retreat possibility, the frequency of freezing was increased significantly in comparison to the therapy setting with retreat possibility and the control setting. Comfort behavior was never observed during therapy without retreat possibility. This study provides evidence that the possibility of retreat is instrumental in reducing stress and should be provided during AAT using guinea pigs. In this form, AAT elicits limited stress and may possibly even provide enrichment. Further research is needed to understand factors influencing guinea pig behavior to ensure animal welfare in AAIs in the future.

Introduction

Guinea pigs are included in various animal-assisted interventions (AAIs). However, there are currently no data investigating the effects of human contact or integration of animals in AAIs on guinea pig behavior.

Animal-assisted therapy (AAT) is a form of AAI defined as a goal-oriented therapeutic intervention with animals as an integral part delivered by health, education, and human service professionals (IAHAIO, 2014). AAI research has documented numerous benefits for humans (Virués-Ortega et al., 2012; Maujean et al., 2015), including a reduction in anxiety, depression, and posttraumatic stress disorder symptoms (O'Haire et al., 2015), and seems to be beneficial in different populations such as people with dementia (Olsen et al., 2016), psychiatric patients (Rossetti and King, 2010), and patients with autism and behavioral difficulties (O'Haire, 2013).

Funding resources: This work was funded by REHAB Basel (Förderverein pro REHAB Basel) and the Forschungsfonds of University of Basel (grant number DZX2120).

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https://doi.org/10.1016/j.jveb.2018.02.004
1558-7878/

Despite these known benefits, further research about the effects of AAIs on both human and animal well-being is needed. Transdisciplinary cooperation between human health care professionals, animal keepers, veterinarians, and ethologists is necessary. "One Health" is an important framework for AAT (Chalmers and Dell, 2015; Hediger and Beetz, 2015; Takashima and Day, 2014; Turner, 2015) due to potential added value in terms of better health for all when compared to separated approaches (Zinsstag et al., 2015). On a practical level, the interdependence of human and animal health in AAT is reflected by guidelines (IAHAIO, 2014) emphasizing the need to consider safety and welfare for humans and animals. However, research focuses on the human health side, whereas little knowledge exists about the impact of AAT on animal welfare. Existing literature investigates only dogs (Glenk, 2017) and horses (e.g., Gehrke et al., 2011).

Guinea pigs (*Cavia aperea f. porcellus*) are common as pets and in research (European Commission, 2013) and also often integrated in AAIs (Matuszek, 2010). There are a few studies investigating the effects of AAI with guinea pigs on the human participants (Marr et al., 2000; O'Haire et al., 2013; 2014; Talarovičová et al., 2010). However, no studies on the effect of human interaction on guinea pig behavior or well-being were found. To address this research gap, the aims of the study were to characterize behavioral changes in guinea pigs caused by human interactions and identify factors linked to guinea pig stress and well-being within the therapeutic setting.

A widely accepted definition of the animal welfare concept refers to an animal's attempts to cope with (Broom, 1986) and the possibility to interact with their environment (Ohl and van der Staay, 2012). Retreat possibility, allowing for free choice of human contact, is a factor which enhances an animal's ability to cope within an environment. Retreat possibility is clearly linked with improved well-being for different species living in captivity such as in zoos, laboratories, or agricultural settings (Morgan and Tromborg, 2007). Guinea pigs, however, are often held and petted by humans, without having retreat possibility. We hypothesized that constant retreat possibility would be a mechanism to reduce stress and improve well-being of guinea pigs during AAT. In the first step, we developed a specific ethogram to observe possible behavioral changes. Then, we investigated the behavior of guinea pigs during AAT with and without retreat possibility, in comparison to a control setting with retreat possibility and no human interaction.

Material and methods

Subjects

We observed 5 guinea pigs (*Cavia aperea f. porcellus*) from a group of six individuals used regularly in AAT at REHAB Basel, a rehabilitation clinic in Switzerland. They were of mixed sources and breeds, and individual identification occurred by natural markings. All animals were housed in a private household and had contact with humans on a daily basis beginning at 8 to 10 weeks. The individuals that were comfortable with human interaction were transferred to the Therapie-Tiergarten at the rehabilitation clinic at the age of at least 1 year and began assisting in the AAT program. All guinea pigs were healthy and housed in accordance with Swiss standards for animal welfare. They were held in 2 groups. Group 1 consisted of 3 5-year-old females, whereas group 2 consisted of 2 2-year-old females and 1 2-year-old castrated male. In this study, the behavior of the male was not further examined with respect to expected behavioral differences compared to the female guinea pigs (Kunkel, 1964; Rood, 1970).

Study design

The study followed a randomized controlled within-subject design with repeated measurement. Each guinea pig was observed in 3 different settings:

(1) Therapy setting with retreat possibility (TWR, table cage with human interaction);
(2) Therapy setting without (no) retreat possibility (TNR, lap of patient);
(3) Control setting (CS, table cage without human interaction).

There were a total of 50 observations, consisting of 4 observations per guinea pig in both the therapy setting with retreat possibility (n = 20) and the control setting (n = 20), as well as 2 observations per guinea pig in the therapy setting without retreat possibility (n = 10).

All 3 settings took place in a room designed for AAT at REHAB Basel, Switzerland. For both the therapy setting with retreat possibility and the control setting, the guinea pigs were placed in a table cage (see Figure 1). The cage had Plexiglas® (polymethyl methacrylate) boards and contained shelters, twigs, wood shavings, hay, straw, and a bowl of water. This structure comprised the "cage part." All elements were arranged in standardized locations (see Appendix). During therapy, the front part of the Plexiglas® board could be removed allowing the guinea pigs to access a wooden board with a pet bed on it. This portion is referred to as the "board part." By choosing either the cage or the board part, the animal could respectively retreat from or approach the patient. For the therapy setting without retreat possibility, 1 guinea pig was placed in a pet bed on the patient's lap (see Figure 2).

Before each session, the guinea pigs were given at least a 5-minute acclimation time in the table cage. During this time, ambient noise and activity were kept to a minimum. The people recording the videos were in regular contact with the guinea pigs and maintained a neutral relationship by minimizing interactions during the standardized filming sessions.

The animals were tested within the groups in which they were housed based on the principal of social buffering (Hennessy et al., 2008). For each observation, 1 animal was randomly defined as focal animal based on simple random numbers generated in Excel

Figure 1. A patient sitting in front of the table cage interacting with 1 of the 3 guinea pigs with assistance from an occupational therapist (therapy setting with retreat possibility). One guinea pig is eating on the board part of the table cage. The scene is posed with 2 members of the study team to ensure anonymity of participating patients and therapists.

Figure 2. In the therapy setting without retreat possibility, a single guinea pig is placed in a pet bed on the lap of a seated patient.

(Microsoft®). Each observation took place on a different day, except for the therapy setting without retreat possibility that was connected to the therapy setting with retreat possibility and followed just afterward. For that, one of the guinea pigs of the group was randomly chosen to be placed on the patient's lap for 5 minutes following the therapy setting with retreat possibility. For each guinea pig, the therapy setting with retreat possibility was defined as the starting condition to ensure that the following observation some days later in the control setting could be matched for length of the session. However, both groups were accustomed to all 3 settings before the study began so that there was no novelty effect and no need to randomize the starting condition. There was no randomization regarding the involved therapist and patient, as it was predetermined which guinea pig group assisted with which patient.

In each session, the following variables were controlled: time of day, time since last feeding, presence of social partners, amount and kind of food, scent of other animals in the litter, position of shelters and water bowl in the table cage, and placement of the observer. Therapy room temperature, outside temperature, the attending therapist and animal keeper, patient identification, and any unexpected events were recorded.

Therapy setting with retreat possibility

In this setting, actual AAT sessions with patients from the clinic were observed. One of the 2 guinea pig groups was placed in the table cage where they had the possibility to retreat or to walk onto the board part of the table cage the entire time. The session duration ranged from 10 to 30 minutes with a frequency of 2 to 5 times per week. The length of the sessions was determined by patient ability and therapeutic goals and was defined by the therapist, who was not involved in the study. During each session, a patient, therapist, animal keeper, and person filming were present. Animal keepers and therapists collaborated closely to oversee interactions between animals and patients and ensure the well-being of all participants. Patients were offered a broad range of activities, including cutting vegetables for the guinea pigs; filling the cut vegetables in wooden pet puzzle toys, branches with holes, or a wooden board with holes; trying to guide the guinea pigs over a wooden seesaw; or brushing them. Patients often encouraged the guinea pigs to approach by feeding them and attempted to pet them.

Therapy setting without retreat possibility

After the therapy setting with retreat possibility, 1 guinea pig was randomly chosen and placed on the lap of the patient. The animal keeper motivated the guinea pig to walk onto a pet bed voluntarily, only handling the animal if necessary. Then the pet bed, with the guinea pig on it, was placed on the lap of the patient. The patient stroked and fed the guinea pig for 1 to 5 minutes. During this time, the guinea pig was observed closely by the keeper for any defined breakup criteria.

Control setting

In this setting, the guinea pigs were placed into the table cage with conditions comparable to the AAT sessions (time of day, room temperature, and duration) except for the human interaction. Only the person filming was present. A control session always followed an AAT session on a different day to ensure that the duration of the sessions was similar and the same kind and amount of food was offered as during the AAT condition. No additional play items were used in the control setting because these are an integral part of the therapy, so the guinea pigs only encounter them when interacting with people.

Data collection

In a first step, an ethogram was developed to observe possible behavioral changes caused by human interaction. Online databases were searched (PubMed, Medline, Science Direct, Web of Science, and Google Scholar; key words: cavy, cavia, caviomorph rodent, ethology, behavior, stress, comfort, vocalization, animal assisted therapy, pet therapy, and animal assisted intervention) for existing guinea pig ethograms, and different guinea pig behavior experts were consulted. Our ethogram was structured similarly to the 2 most frequently referenced ethograms (Kunkel, 1964; Rood, 1970). The following behavior groups were included:

(a) Individual behavior: ingestive, locomotive, and comfort behavior
(b) Interactions with the environment: explorative and non-explorative behavior
(c) Social behavior: sociopositive and general socionegative behavior
(d) Active human-animal interaction (HAI): sociopositive HAI and general socionegative HAI
(e) Passive human animal interaction: stroked
(f) Vocalization
(g) Other groups: visibility, unexpected behavior, and on-going observation

A detailed description of the ethogram is found in the additional material (see Appendix).

All sessions were filmed using a Sony® camcorder. Animal behavior was analyzed using continuous recording and focal sampling (Altmann, 1974) through video coding with Noldus Observer® XT 12.5. Before coding any study videos, coders were trained. Intrarater reliability ranged between 0.85 and 1.0 and interrater reliability was 0.95, as measured by Cohen's kappa (Cohen, 1960).

Data processing and statistical analysis

Frequencies of short countable behaviors were calculated as n/100 s. Longer enduring state behaviors were calculated as percentages of the observed time. For most behaviors, the denominator "visible and on-going" was used. This ensured that the reference time (100%) only counted when the therapy was on-going and the animal was visible in the camera. For "vocalization," "hiding," "on board part," and "in cage unsheltered," only the denominator "on-going" was used because these behaviors also occurred when the animal was not visible.

Countable behavior data were analyzed using a generalized linear mixed model with Poisson distribution. In case of overdispersion, observed via deviance (DF), we fitted a model using the negative binomial distribution. Setting was used as fixed effect, and individual guinea pigs were used as random effect. The logarithmized duration of the session was specified as an offset variable. As effect size, the incidence rate ratio (IRR) was calculated, that is, the relative change in the rates of the observed event.

To analyze the data of state behaviors, arcsine transformed percentage of the analyzed time in total was used. A generalized linear mixed model with setting (TWR, TNR, or CS) as fixed effects and individual guinea pig as random effect was used. As effect size, the linear coefficient (b) was calculated, that is, the difference in the proportions but estimated on arcsine scale. We used IBM SPSS® Statistics, Version 22.0, for all analyses and considered P values ≤ 0.05 as statistically significant.

Results

Therapy with retreat possibility versus control setting

Frequency of hiding was increased in the therapy setting with retreat possibility (IRR = 1.69, CI = 1.28 to 2.23, $P < 0.01$), whereas duration spent hiding remained the same when compared with the control setting (see Figures 3 and 4 and Tables 1 and 2). Time spent in the cage unsheltered decreased significantly during therapy with retreat possibility (b = 0.86, CI = −0.76 to 0.98, $P = 0.02$). There was a trend of increased freezing (IRR = 1.21, CI = 0.97 to 1.50, $P = 0.09$), whereas startling was increased significantly in the therapy setting with retreat possibility (IRR = 1.65, CI = 1.01 to 2.69, $P = 0.05$). Other behaviors such as duration of resting, frequency of general socio-negative, and comfort behaviors did not differ between settings.

During therapy with retreat possibility, duration of locomotion (b = 1.11, CI = 0.08 to 0.14, $P < 0.01$), frequency of explorative behavior (IRR = 7.71, CI = 4.00 to 14.85, $P < 0.01$), and time spent not eating were increased (b = 0.18, CI = 0.10 to 0.26, $P < 0.01$).

Therapy without retreat versus with retreat possibility

During therapy without retreat possibility, the guinea pigs were stroked longer compared to the therapy setting with retreat possibility (b = −0.92, CI = −1.08 to −0.76, $P < 0.01$). Time spent in locomotion decreased in the therapy setting without retreat possibility (b = −0.13, CI = −0.21 to −0.05, $P < 0.01$). Even though total time spent not eating increased in the therapy setting without retreat possibility (b = 0.25, CI = 0.02 to 0.49, $P = 0.04$), time spent eating with human interaction remained the same as in the therapy setting with retreat possibility. Further behavioral changes in the therapy setting without retreat possibility were similar to the changes in the control setting and are described in more detail in the following section.

Therapy without retreat versus therapy with retreat possibility and control setting

During the therapy setting without retreat possibility, there was a strong increase in frequency of freezing compared to both the therapy setting with retreat possibility and the control setting (TWR: IRR = 4.34, CI = 3.33 to 5.88, $P < 0.01$; CS: IRR = 5.32, CI = 3.93 to 7.20, $P < 0.01$). Time spent not eating (TWR: b = 0.25, CI = 0.02 to 0.49, $P = 0.04$; CS: b = 0.44, CI = 0.21 to 0.67, $P < 0.01$) and time spent vocalizing increased significantly compared to both conditions (TWR: b = 0.05, CI = 0.01 to 0.08, $P = 0.01$; CS: b = 0.05, CI = 0.02 to 0.08, $P < 0.01$). In the therapy setting without retreat possibility condition, resting and comfort behaviors were never observed. No other significant changes occurred in the control setting that did not correspond to those in the therapy setting with retreat possibility.

Individual differences and other factors

Five behaviors were considered most important concerning either stress or enrichment and were examined more closely for other influence factors. The 5 key behaviors were locomotion, explorative behavior, comfort behavior, freezing, and hiding. The individual guinea pigs and the different patients, therapists, and keepers were examined as influencing factors.

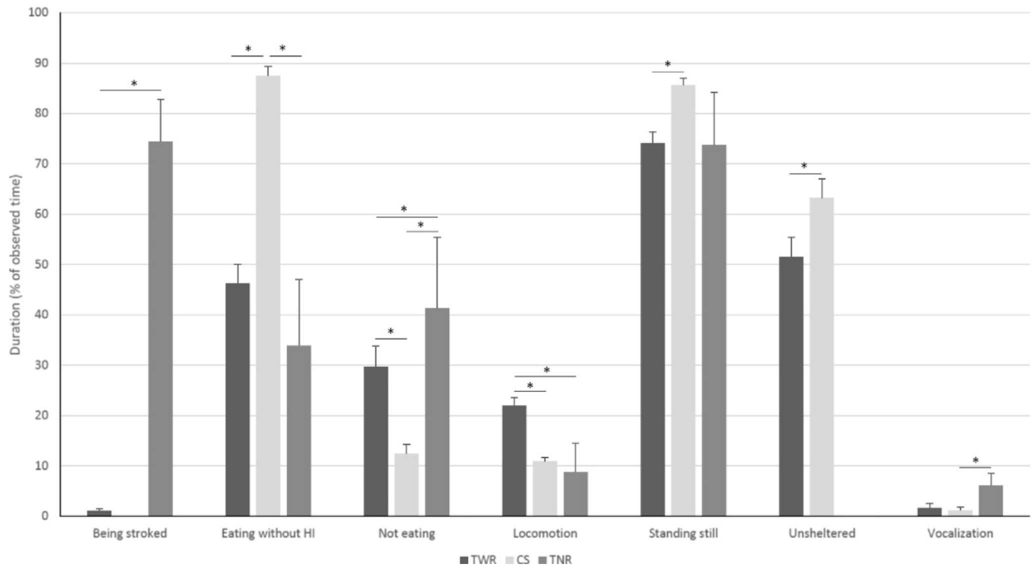

Figure 3. Duration of observed state behaviors which differ significantly between conditions. Error bars denote 1 standard error of the mean, * indicate statistically significant comparisons. HI, human interaction; TWR, therapy setting with retreat possibility; CS, control setting; TNR, therapy setting without (no) retreat possibility.

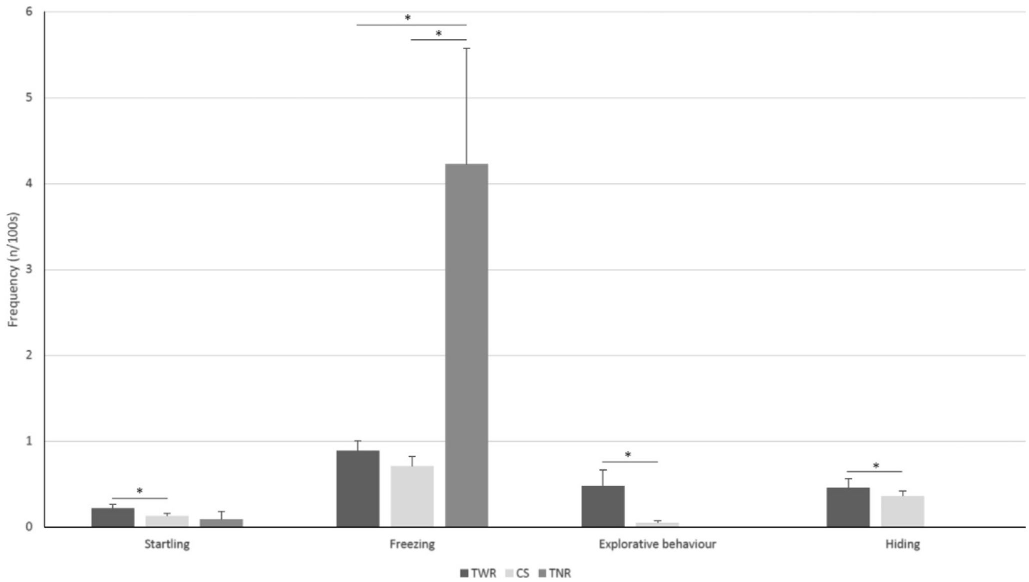

Figure 4. Frequency of count behaviors which differ significantly between conditions. Error bars denote one standard error of the mean, * indicate statistically significant comparisons. TWR, CS, control setting; TNR, therapy setting without (no) retreat possibility.

Locomotion was influenced significantly by the therapist (b = −0.06, CI = −0.10 to −0.02; $P = 0.01$). There was also a tendency that it was influenced by the keeper (b = 0.033, CI = −0.00 to 0.07, $P = 0.06$). Explorative behavior was influenced by the patient (IRR = 0.52, CI = 0.41 to 0.68, $P < 0.01$), therapist (IRR = 0.04, CI = 0.02 to 0.12, $P < 0.01$), and individual guinea pig (IRR = 0.73, CI = 0.64 to 0.85, $P < 0.01$). Comfort behavior was not influenced by these factors.

The keeper (IRR = 1.16, CI = 1.03 to 1.31, $P = 0.01$) and the individual guinea pigs significantly (IRR = 1.22, CI = 1.12 to 1.34, $P < 0.01$) influenced the frequency of freezing, and the patient influenced the frequency of freezing by trend (IRR = 1.13, CI = 0.99 to 1.29, $P = 0.08$). The duration spent hiding was influenced by the individual personality of the guinea pig (b = 0.04, CI = −0.01 to 0.09, $P = 0.05$), whereas the patient present during therapy influenced the frequency of hiding (IRR = 1.79, CI = 1.28 to 2.51, $P < 0.01$).

Table 1
State behaviors

Behavior	Setting	N	Duration	M	SD	Comparison	Coef	95% CI	*P* value
Being stroked	TWR	20	23	1.13	1.61	TNR-TWR	−0.92	−1.08 to −0.76	<0.001[a]
	TNR	10	133	74.45	26.35				
Eating with HI	TWR	20	487	24.01	13.36	TNR-TWR	0.05	−0.13 to 0.24	0.56
	TNR	10	46	24.92	35.17				
Eating without HI	TWR	20	1043	46.33	16.56	TWR-CS	−0.61	−0.72 to −0.50	<0.001[a]
	CS	20	1727	87.54	8.17	TNR-CS	−0.70	−0.92 to −0.48	<0.001[a]
	TNR	10	70	33.92	41.39	TNR-TWR	−0.09	−0.31 to 0.14	0.46
Not eating	TWR	20	688	29.66	18.71	TWR-CS	0.18	0.10 to 0.26	<0.001[a]
	CS	20	257	12.46	8.17	TNR-CS	0.44	0.21 to 0.67	<0.001[a]
	TNR	10	53	41.16	44.54	TNR-TWR	0.25	0.02 to 0.49	0.04[a]
Locomotion	TWR	20	490	22.01	6.92	TWR-CS	1.11	0.08 to 0.14	<0.001[a]
	CS	20	218	10.95	3.36	TNR-CS	−0.02	−0.09 to 0.06	0.64
	TNR	10	9	8.81	17.78	TNR-TWR	−0.13	−0.21 to −0.05	0.001[a]
Resting	TWR	20	26	0.82	3.22	TWR-CS	0.00	−0.01 to 0.02	0.68
	CS	20	10	0.49	2.20	TNR-CS		Perfect prediction	
	TNR	10	0	0.00	0.00	TWR-TNR		Perfect prediction	
Standing still	TWR	20	1627	74.18	9.81	TWR-CS	−0.20	−0.26 to −1.31	<0.001[a]
	CS	20	1695	85.71	5.93	TNR-CS	−0.04	−0.25 to 0.17	0.70
	TNR	10	136	73.79	32.85	TNR-TWR	0.15	−0.46 to 0.35	0.13
Hiding	TWR	20	473	20.08	18.06	TWR-CS	0.022	−0.10 to 0.14	0.71
	CS	20	371	17.72	20.63				
On board part	TWR	20	595	28.35	20.59	TWR-CS	0.10	−0.01 to 0.21	0.07
	CS	20	367	19.03	16.02				
In cage unsheltered	TWR	20	1150	51.57	17.27	TWR-CS	0.86	−0.76 to 0.98	0.02[a]
	CS	20	1247	63.26	16.61				
Vocalization	TWR	20	45	1.67	4.02	TWR-CS	0.01	−0.01 to 0.02	0.51
	CS	20	24	1.15	2.89	TNR-CS	0.05	0.02 to 0.08	0.002[a]
	TNR	10	8	6.13	7.62	TNR-TWR	0.05	0.01 to 0.08	0.01[a]

TWR, therapy with retreat; CS, control setting; TNR, therapy no retreat; HI, human interaction; N, number of sessions; Duration, total observed duration ($\times 10^3$ s); M, mean in % of observed time; SD, standard deviation; Coef, coefficient (effect size); CI, confidence interval.

[a] Statistically significant.

Table 2
Count behaviors

Behavior	Setting	N	Total	M	SD	Comparison	RR	95% CI	*P* value
Socionegative behavior	TWR	20	54	0.25	0.25	TWR-CS	0.96	0.66–1.41	0.85
	CS	20	51	0.26	0.16				
Socionegative active HAI	TWR	20	27	0.13	0.19	TNR-TWR	0.49	0.07–3.70	0.49
	TNR	10	1	0.07	0.23				
Startling	TWR	20	45	0.22	0.18	TWR-CS	1.65	1.01–2.69	0.05[a]
	CS	20	25	0.13	0.14	TNR-CS	0.43	0.059–3.21	0.41
	TNR	10	1	0.09	0.29	TNR -TWR	0.26	0.04–1.92	0.19
Freezing	TWR	20	197	0.89	0.51	TWR-CS	1.21	0.97–1.50	0.09
	CS	20	145	0.71	0.51	TNR-CS	5.32	3.93–7.20	<0.001[a]
	TNR	10	61	4.23	4.26	TNR-TWR	4.34	3.33–5.88	<0.001[a]
Comfort behavior	TWR	20	46	0.22	0.15	TWR-CS	1.77	0.76–1.83	0.47
	CS	20	35	0.18	0.10	TNR-CS		Perfect prediction	
	TNR	10	0	0.00	0.00	TNR-TWR		Perfect prediction	
Explorative behavior	TWR	20	83	0.48	0.84	TWR-CS	7.71	4.00–14.85	<0.001[a]
	CS	20	10	0.05	0.10				
Hiding	TWR	20	143	0.62	0.46	TWR-CS	1.69	1.28–2.23	<0.001[a]
	CS	20	75	0.36	0.28				

TWR, therapy with retreat; CS, control setting; TNR, therapy no retreat; HAI, active human animal interaction; N, number of sessions; Total, total number of events within all observations; M, mean per 100 seconds; SD, standard deviation; RR, rate ratio (effect size); CI, confidence interval.
[a] Statistically significant.

Descriptive statistics

The following behaviors occurred too rarely to compare between conditions and were therefore not described: sociopositive active HAI (n [TWR] = 5, n [TNR] = 0) and jumping (n [TWR] = 4, n [CS] = 0).

Discussion

Guinea pigs showed higher frequency, but not duration, of hiding and an increase in startling, as well as more locomotion, explorative behavior, and time spent not eating during therapy with retreat possibility compared to the control setting without human interaction. Without retreat possibility, guinea pigs showed a strong increase in freezing, not eating and vocalizing. Locomotion and resting decreased without retreat possibility.

In previous studies, stress in guinea pigs was investigated in the context of social buffering (Hennessy et al., 2008; Maken and Hennessy, 2009), isolation (Hennessy et al., 2004), or social conflicts (Haemisch, 1990; Sachser and Lick, 1989; 1991) in combination with novel environments as a stressor. Research showed that stress led to a reduction in comfort behavior, social behavior, explorative behavior, and an increased amount of freezing and withdrawal (Anthony et al., 1959). Stress also led to reduced eating, exploration, and sexual behaviors (Hennessy et al., 2004). Further stress-related behaviors were cataloged: vocalization, freezing, startling, and altered activity, such as hiding, stampeding behavior, fighting, teeth chattering, or stereotypical behaviors like bar biting or barbering (Brandão and Mayer, 2011; Brewer et al., 2014; Hennessy et al., 2004; King, 1956; Sachser and Lick, 1991).

The increase in hiding, startling, freezing, and vocalizing as well as the reduction in comfort behavior and time spent eating due to human interaction which we found in our study can therefore be viewed as stress-related behaviors. The 2 different settings with human interaction differed in the amount of observed behavioral changes related to stress. We, therefore, conclude that the therapy settings with and without retreat possibility can be seen as 2 different levels of stress applied to the guinea pigs, with a higher level of stress during therapy without retreat possibility.

We did not find so-called "stress-induced sickness behaviors" such as crouching, piloerection, or eye-closing (Hennessy et al., 2004), and there was no increase in fighting (socionegative behavior) in either therapy condition. Thus, we conclude that stress was not notably high under either condition.

A main reason the guinea pigs experienced higher stress in the lap condition could be due to the limited retreat possibility. Seeking shelter is a natural behavior of guinea pigs. Ohl and van der Staay (2012) suggested that when this need cannot be met, stress results.

Another factor might be lack of social partners in the lap context, whereas in the therapy setting with retreat possibility, animals were within their group. Social partners play an important role in stress buffering (Hennessy et al., 2008). Also, the guinea pigs were not acclimatized to the lap setting to the same degree, despite being regularly handled, while the table cage was a familiar environment. The occurrence of startling during the therapy setting without retreat possibility likely decreased due to the increased time spent freezing. Variations in time spent in locomotion or eating were possibly based on individual coping tactics or perception of stress.

In addition to the stress-related changes, we also observed changes in enrichment-associated behavior relative to the conditions. Enrichment is designated as change in the frequency of behavior without occurrence of stress-associated behavior (Brewer et al., 2014). It is therefore closely linked to stress. Enrichment enables animals to express the full range of their species-typical behavioral patterns and includes environmental, social, and nutritional enrichment along with foraging (Hutchinson et al., 2005). Enrichment is an important component of animal welfare. It can be integrated into Fraser's concept of "needs" and "opportunities" and provide opportunities for pleasure (Fraser and Duncan, 1998).

In this study, the guinea pigs showed more explorative behavior and more locomotion during therapy with retreat possibility when compared to the control setting. These behaviors are directly enrichment related (Brewer et al., 2014). Therefore, we conclude that even though AAT in the table cage may cause a small amount of stress, it primarily functions as enrichment for the guinea pigs.

Our data confirm that shelter is an important component of environmental enrichment. Shelters were used more frequently in the therapy setting with retreat possibility than in the control condition and may have contributed to stress reduction. Nordlund (2004) found that even the design of the shelter plays a role in decreasing social conflict and stress. Guinea pigs prefer low vegetation in the wild (Cassini and Galante, 1992) and roofed areas under domestic conditions (Büttner, 1992; Lee et al., 2014). Therefore, shelters should be provided to accommodate each individual (King, 1956; Scharmann, 1991).

Social enrichment can be provided via conspecific (other guinea pigs) or contraspecific (other species or humans) interactions. Although conspecific group housing is socially important (Berryman, 1978; Kaiser et al., 2003), interaction with humans can also be a supplement. In our study, the guinea pigs freely chose to interact with the humans in the therapy setting with retreat possibility. Such human interaction can act as social enrichment, whereas the feeding may function as a foraging and nutritional enrichment.

When animal-human interaction was examined more closely in this study, approximately a quarter of the time was spent eating food acquired from the human in both therapy settings (TWR and TNR). There was a trend for a longer duration spent on the board part in the therapy setting with retreat possibility compared to the control setting, which may be in the context of being fed on the board part. This shows that guinea pigs spend time interacting with humans when they can choose freely but only to a certain degree. The guinea pigs very seldom displayed sociopositive behavior directed toward a human. There was no difference in the frequency of socionegative behavior directed toward a human between the therapy settings with and without retreat possibility. In the therapy setting with retreat possibility, the guinea pigs were only stroked for a small fraction of the total time, compared to the therapy setting without retreat possibility, where they were stroked for a majority of the total time. This was probably due to the limited possibility of retreat and may also contribute to the higher degree of stress in the lap setting.

Our data show that in addition to the setting, the individual personality of the guinea pig is an important factor, as also documented by Zipser et al., (2013). Moreover, the relationship with the patient, therapist, and keeper are relevant factors influencing guinea pig behavior.

Limitations

A limitation of the study was the sample size. We investigated 5 female individuals and found distinct individual differences between the guinea pigs. Therefore, our results should be cautiously generalized to other guinea pigs. The total of 50 observations was too small to thoroughly examine rare behaviors like jumping and sociopositive interaction. Moreover, not all guinea pigs were tested with all patients, therapists, and keepers. The exploratory findings regarding these factors must be interpreted with caution and should be seen as a hypothesis-generating outcome.

Another limitation was the fact that the study could not be blinded. However, the video coders were not involved with AAT before the study. To minimize biases, there was a detailed coding scheme, and intrarater and interrater reliability was high. The study could only be randomized to a certain degree, as the combination of a guinea pig group with a patient and their therapist was predetermined.

Finally, observed behavior should always be interpreted with caution. We interpreted our data on the basis of existing literature on stress in guinea pigs. It is unclear how these interpretations correspond with the guinea pigs' perception and physiological or even health or longevity outcomes.

Strengths

This is the first study investigating behavioral effects in guinea pigs assisting in AAT. We compared 2 different types of therapy settings. This gives new insights on how to reduce stress and enhance well-being, hopefully leading to guidelines for AAI with guinea pigs. In addition, we designed the first specific ethogram for observing human-guinea pig interaction, which provides a useful tool for future research.

We carefully controlled for confounding factors to ensure high internal validity. However, we studied AAT sessions in an on-going program with real patients. We observed guinea pigs that were already being used in the program, so we were not able to select for special criteria other than sex. Our approach ensures high external validity and reflects the actual practice.

Implications for research

Our study is a first step toward understanding the effects of human-animal interaction on guinea pig well-being, and specifically the effects of integrating guinea pigs into AAT. Further research is needed to investigate the effects of different types of human-animal interaction and environments more closely on a descriptive level. Further research regarding the meaning and clinical relevance of the observed behavioral changes is needed. It remains unclear what amount of change should be considered as "distress" or "eustress" and what intensity is associated with health problems. It is important to further combine behavioral observations with physiological measures, such as cortisol measurements in previous studies (Maken and Hennessy, 2009; Zipser et al., 2013; Haemisch, 1990; Brewer et al., 2014, Sachser, 1986). Behavior during therapy should be compared with that in a natural habitat. Further research should also include more individuals to investigate whether sex, age, personality, amount of experience in AAT, and contact with humans in general have an effect. We also suggest testing more different setting variables such as the length of a therapy session, the different humans present, and social buffering.

Implications for practice

Guinea pigs are useful in therapeutic settings (Flom, 2005; Talarovičová et al., 2010; O'Haire, 2013; O'Haire et al., 2015; O'Haire et al., 2013; O'Haire et al., 2014); however, the procedure conditions vary markedly. Statements in the literature range from guinea pigs "generally like to be held" (Zawistowski, 2008) to guinea pigs "are very prone to stress" (Hubrecht and Kirkwood, 2010; King, 1956). Our data show that the setting is crucial to reduce stress and ensure well-being. Clearly, domesticated guinea pigs are less susceptible to stress and less aggressive than wild guinea pigs (Sachser, 1998; Künzl and Sachser, 1999; Brust and Guenther, 2015). However, they are still timid flight animals which display subtle defense mechanisms or signs of stress in the face of threats. They rarely bite, preferring to retreat and hide, alternatively freezing when retreat is not possible. They rarely exhibit sociopositive, nonsexual interactions as adults within a group (Kunkel, 1964; Rood, 1972), which was confirmed in our observations. Therefore, it is not natural for guinea pigs to be stroked or touched. We highly recommend that guinea pigs are given the possibility to retreat whenever possible, so they can freely choose to interact with humans.

Our findings have implications for all guinea pigs in human care. Many more guinea pigs are kept as pets in private households. The factors described in this study for guinea pigs assisting in therapy are also relevant for human-guinea pig interactions in general to ensure animal well-being. It is important for pet owners to understand that free interactions and retreat possibilities are relevant for species-appropriate handling and keeping.

Conclusion

Our data show that the setting for human-guinea pig interactions is crucial. A "good practice" for guinea pig-assisted interventions includes retreat possibilities whenever possible to ensure free choice of human interaction. In this way, stress can be reduced, and AAT might even constitute enrichment for the animal.

Acknowledgments

The authors would like to thank Bettina Finger and Christian Weber and the Therapie-Tiergarten team for their cooperation during the study and the involved therapists and patients for participating. They also thank Iris Marti and Paula Ospitia Rodriguez for help in designing the ethogram and collecting data. Special thanks to Dennis Turner, Gottfried Morgenegg, Sabine Gebhardt-Henrich, and Hanno Würbel for discussion and input on human-guinea pig interaction and advice in designing the study and commenting on the ethogram. They also thank Silvia Kaiser for reviewing the ethogram.

Authorship statement: K.H. conceived the idea of the study. The concept of the study was elaborated by K.H., L.C., and J.Z. W.G. designed and conducted the experiments under the supervision of K.H. and J.Z. The statistical approach was elaborated by J.H., and data were analyzed by K.H., J.Z., J.H., and W.G. The paper was written by K.H. and W.G. with input from L.C. and J.Z.

Ethical considerations

This study was approved by the Veterinary Office of the canton Basel-Stadt, Switzerland, and by the Human Ethics Committee EKNZ (Ethics Committee for Northwest and Central Switzerland). The study was conducted in accordance with the Animals (Scientific Procedures) Act 1986, European Directive EU 2010/63, and the Guidelines for the Use of Animals in Research of the Association for the Study of Animal Behaviour and the Animal Behaviour Society. AAT was performed according to the IAHAIO guidelines (IAHAIO, 2014). A therapy session lasted for a maximum of 30 minutes during defined time slots with defined breaks between sessions. Break-off criteria were defined as an excessive display of aggression by the human or animal during AAT. The condition without retreat possibility was limited to 5 minutes, and the additional break-off criterion, an excessive display of stress-associated behavior by the animal (e.g., piloerection, eye-closing, or attempts to flee like jumping out of the pet bed on the patient's lap), was included. No session had to be ended early, and no adverse incidents occurred.

Conflict of interest

The authors state no conflict of interest.

Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.jveb.2018.02.004.

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