Oxytocin and self-reported anxiety during interaction between humans and farm animals – an exploratory pilot study.

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Abstact

The aim of this pilot study was to explore the possible change in serum Oxytocin and state anxiety as respons to an interaction between healthy female nursing students (n=18) and domesticated farm animals (cows) in a natural setting. The students performed stroking and brushing of a well socialized cow for 15 minutes. Blood samples were drawn at baseline (T1) after 5 minutes intervention (T2) and immediately after (T3). At T1 and T3 the students also filled is a self evaluation instrument measuring their state anxiety (Spielberger Stait Trait Anxiety Inventory, State subscale- STAI-SS).
No significant change in serum Oxytocin were detected, but there was a significant decline in stait anxiety score (p=0.014) between T1 and T3. There was also a significant positive correlation between the decline in STAI-SS score and the change in serum Oxytocin level between T2 and T3. This indicate a later onset of the anxiolytic effect of Oxytocin when interacting with a big unfamiliar cow, than with a friendly dog as previous studies report.

Background

Interactions between humans and animals, described as *Animal-assisted Interventions* (AAI), has once again become a topic within schools and healthcare settings. One of the earliest documented use of animals in therapeutic purpose is dated back to year 1790. At the York Retreat in England patients been treated for different mental illnesses, were urged to interact with small domestic animals in the hospitals garden (Burch 1996 sited in Palley et al. 2010). In Norway the first mental hospitals were founded around 1850. They were located in the country side and built around an already excising farm. One regarded the peaceful environment and doing practical work at the farm, to a part of the treatment. Care for the animals at the farm was considered being health promoting, and are still in use at many psychiatric institutions around Europe (Berget & Braastad 2008). The child psychiatrist Boris Levinson (1965) argued for the therapeutic value of using companion animals in the treatment of children. The last decades the use of companion animals has become more and more popular in therapeutic and occupational settings (Sempik et al. 2010). The scientific studies published so far, has mainly focused on AAI with companion animals, mainly dogs.. Farm animals all over Europe serves as a re-instated "co-therapists" for rehabilitation, therapy and health promotion, in the concept of Green care (Haubenhofer et al. 2010; Sempik et al. 2010). The concept of Green Care covers a broad spectre of activities within the health maintaining, and health promoting measures for different groups of users in different settings, including

animals or other nature elements (Haubenhofer et al. 2010). There are until now, few evidence-based studies of AAI with farm animals within the concept of Green Care. Two Norwegian studies have documented effects of AAI with farm animals for persons with psychiatric disorders (Berget et al. 2008; 2011; Pedersen 2011). Both studies fill the criteria of randomized controlled trial (RCT). Other studies have described children's interaction with cows and other farm animals at the Green Chimneys Educational Farm (Mallon 1994). The participants were not randomized, and there was no control group.

There is a need for more research to establish a paradigme regarding the effectiveness of AAI. When arguing for the effect interactions with farm animals may have upon humans, no research has been done regarding the *physiological parameters*.

The aim of our study was to investigate the possible influence interaction with well socialized, but unfamiliar cows, may have upon the level of human Oxytocin (OT) and self reported level of anxiety before and after a single intervention.

Oxytocin is a neuro peptide released from Hypothalamus and is hypothesized to both influence, and gain effects from social interactions between individuals within spices. Previous studies have documented that OT also plays a part in the interaction between e domesticated animals and humans. The studies report an increase in OT in both human and animal serum (Handlin et al. 2011; Odendaal 2000; Odendaal & Lehmann 2000) , in human only (Miller et al. 2009) and human urine (Nagasawa et al. 2008) after interacting with a friendly dog. .

The relationship between humans and animals is complicated and not yet fully understood. The purpose of this study was to explore the possible change in OT from baseline in concentrations of OT in human blood, during and after one single interaction with an unfamiliar cow.

Methods

Design

The method chosen to study the possible effect from human – farm animal interaction was a single-subject experimental approach. The data were sampled using validated self report questionnaires and blood sampling on site. Approval to perform the study was given by the Regional Ethics Committee for Medical Research Etichs – Norway (REK).

Participants

Healthy female students (n=18) between 20 and 30 years of age who were physically able to participate in the intervention, were recruited. The exclusion criteria were pregnancy, breastfeeding, anxiety connected with blood sampling and/or animals, allergies, psychiatric diagnoses or other severe disease, and medication (other than oral contraceptives). The participant signed an informed written consent form.

Setting

The study was conducted three Saturdays in a row at an ecological farm with well socialized cows. Due to the protection of animal-welfare the same cow could not participate more than one day.

Instruments

Level of anxiety was measured using the Norwegian version of Spielberger State-Trait Anxiety Inventory- State Subscale (STAI-SS) (Spielberger et al. 1970) adapted by Haaseth et al. (1990). STAI –SS consist of 20 items to which the participant indicate how they feel *at the moment* on a 4-point scale ranging from 1-"almost never" to 4-"almost always". Ten items is formed to record the presence of anxiety symptoms, the other 10 to record the absence of anxiety. The items that scored the absence of anxiety symptoms were inverted before calculating the sum score (Spielberger 1983). The total score, ranging from 20 to 80 points, reflect the level of trait anxiety. High score reflect greater level of trait anxiety than lower score. The internal consistency has a median alpha coefficient of 0.92 (Spielberger 1983).
The Measuring points were before (T1l) and after the intervention (T3).

Intervention and Procedures

The participants were at arrival placed in a waiting room were they could change into suited clothing for working with the animals. Proper safety boots was also provided. When ready, the participant was relocated to another room were she was asked to fill in the STAI-SS. She was then left alone in the room for about 10 minutes reading a book. After 10 minutes the bio engineer entered and collected the first blood sample (T1), baseline sample. She was then brought to the barn were the farmer instructed her in what to do with the cow. The farmer stayed in the barn at a distance, and not talking to the participant during the intervention. This was for safety precautions. Intervention time started when the participant climbed into the cows bin.

After 5 minutes the participant was asked to climb out of the bin, and the second blood sample was drawn immediately (T2). To avoid delays, this relocation of the participant took only a few seconds and the distance was approximately only 1,5 – 2 meters.
The sampling took only 30 seconds. The participant immediately returned to the cow.
The intervention was stopped after 15 minutes, and the participant returned to the room for the final blood sampling (T3). Immediately after sampling the participant filled in the instrument (STAI) again.

Blood sampling

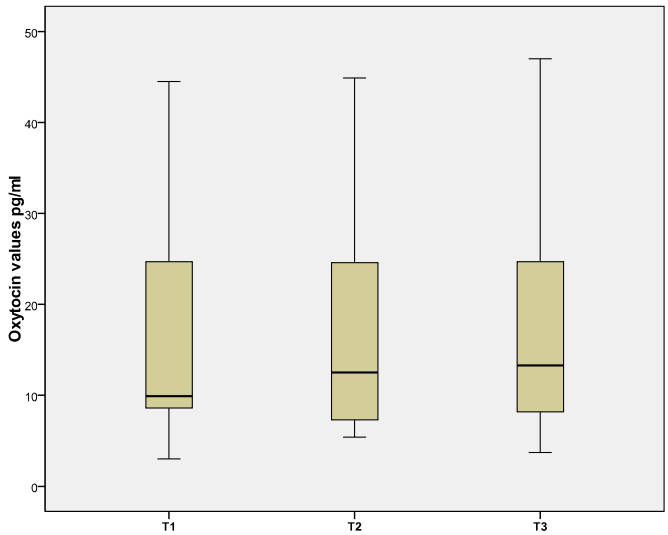
The blood samples were drawn into chilled vacuum tubes using vacutainer. The tubes contained EDTA (1mg/ml blood), Aprotonin (500 KIU/ml blood). The blood was sentrifuged at 5,600 x rpm i 5 minutter ved 4°C. The serum was withdrawn and placed using a pipette in 1.75 ml Eppendorf tubes. The tubes were stored at dry ice at – 70 °C for transport to the main laboratory and further storage until analyzes.

Assays

The analyzes were performed at Oslo University Hospital. Oxytocin levels were determined in humans using Assay Designs' Oxytocin Immunoassay (EIA) kit according to the manufacturers instruction (Enzo Life Sciences, Inc., Farmingdale, NY, USA) (sensitivity 11.7 pg/ml, Intra-assay CV%: <10 for values < 80 pg/ml). Before assay the samples were purified using phenyl silica minicolumns (Bond Elute) according to Sernia et al. (1991). Recovery was checked by adding standards to samples, and was close to 100 %.

Results

The level of Oxytocin for all participants summarized are shown in Table 1, and the distribution displayed in Fig 1, showing significant skewness.



Figur 1 Distribution of Oxytocin values (pg/ml) at T1 (baseline), T2 and T3

Descriptive statistics performed on the results from OT analyzes showed a skewness > 1 for all measuring point.

Table 1. Oxytocin concentration (pg/ml) at (T1) (T2), and (T3) with max. and min. values for each measuring point.

		Oxytocin Levels (pg/ml)					
		Statistics		Mean		Skewness	
	N	Min.	Max.	Stat.	SE	Stat.	SE
T1	18	3.00	44.50	17.82	3.32	1.057	0.536
T2	18	5.40	44.90	17.31	2.88	1.092	0.536
T3	17	3.70	47.00	18.02	3.42	1.223	0.550

Nonparametric tests; related-samples Wilcoxon Signed Ranks tests, were performed to evaluate the change between sampling points. We found no significant change in mean values of serum Oxytocin between T1 and T2 (p=0.65) or between T2 and T3 (p=0.42), or between T1 and T3 (p=0.88).

Self reporting level of anxiety – STAI-SS

All participants (n=18) compleeted STAI-SS at baseline (T1) and after intervention (T3). The mean sum score was calculated using SPSS version 18 (Table 2).

Table 2. Spielberger State Trait Anxiety Inventory State Subscale (STAI-SS), showing N, Min., Max., Mean and SE

	STAI-SS				
	N	Min.	Max	Mean	SE
T1	18	21.00	40.00	27.50	1.13
T3	18	20.00	36.00	25.39	1.05

With the purpose of analyzing the change in self-reported anxiety between T1 and T3, a Paired sample T-test was performed.

Table 3. Paired sample T-test indicating a significant change in self reported state of anxiety between T1 and T3, STAI-SS.

							Sig. (2-tailed)
N=18	Mean	SE	95% CI		t	df	(p-verdi)
STAI T1-T2	- 2.11	0.77	-3.73	-0.49	-2.75	17	0.014*

sig.level p<0.05

There was a significant reduction, p = 0.014 (Sig.2-tailed), from T1 to T3. Cohens d (effect size) = mean (T1-T3) / SD = 0.65, indicating a medium effect size. Cohens d = 0.4 – 0.8 indicates medium effect (Cohen 1977; Cohen 1992). The results from both T1 and T2 were evenly distributed (Fig.2).

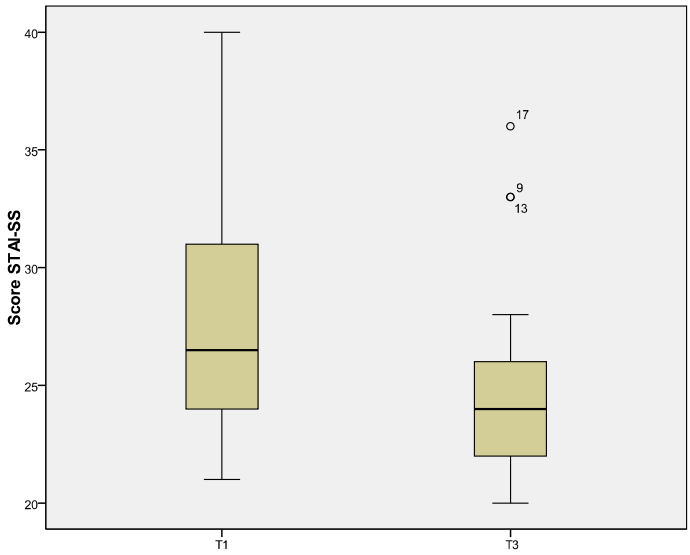


Fig. 2 Distribution of score STAI-SS at T1 and T3.

A correlation analyze between the change in OT values and reduction in self-reported anxiety (STAI-SS) was performed to identify the possible correlation.

Table 3. Correlations between change in STAI-SS (T1 and T3) and OT concentrations (T1-T2, T2-T3 and T1-T3)

	T1-T2	T2-T3	T1-T3
Pearson Correlation	0.00	0.49	0.28
Sig. (2-tailed)	0.989	0.045*	0.277
N	18	17	17

* sig.level $p < 0.05$

Though no significant changes in OT was detected between the measuring points, there was a significant correlation between the *changes* in STAI-SS scores (T1-T3) and OT (T2-T3), p=0.045.

Discussion

There was no significant change in mean OT level between the measuring points. The skewness within each measuring point was significant and surprising. Some studies have suggested a connection between OT and higher levels of estrogens, and therefore hypothesize that the effect is stronger in fertile women, than in men (Petersson et al. 1999). Miller et al (2009) report a gender difference in human OT levels before and after intervention with a friendly dog. Women having a mean 58.% increase in OT, while men having 21.5% decrease in OT. Odendaal (2000) used both men and women, but did not differ between gender in the analyzes. Handlin et al (2011) used only female participants. Due to these findings we would expect an increase in OT because we only recruited women. The skewness might be the result of conditions not possible to control for in such a small sample. The use of oral contraceptives might influence on the OT level (Ludwig & Leng 2006), as will previous pregnancies (Young & Wang 2004) and social contact (Uvnäs-Moberg 1998) with friends while waiting.

The mean STAI-SS score between T1 and T3 showed a significant reduction, and the difference correlated with the difference in OT level between T2 and T3 . These findings might indicate a somewhat later inset of OT secretion in humans interacting with unfamiliar farm animals, compared to previous findings in studies describing interactions with family dogs (Handlin et al. 2011; Miller et al. 2009; Odendaal 2000; Odendaal & Meintjes 2003). Dogs, being our most domesticated animals, and familiar to most people are, well adjusted to human behaviour. The cows used in our study were well socialized, but still unfamiliar and not primarily bred to interacting with humans.

This pilot study aimed to investigate the effects interacting with farm animals might have on the participant's serum OT level, the self reported state of anxiety, and possible correlation between these measurements. No similar experiment, to our knowledge, has earlier been conducted to do compare self-reported measure on anxiety with levels of OT. Other studies have however described a stress reducing effect in humans from interacting with friendly dogs. Decrease in blood pressure after short time interaction with a pet is described in several studies (Barker et al. 2010; Friedmann et al. 1983; Odendaal J.S.J. & Lehmann 2000; Wilson 1987). Wilson (1991) used STAI in her study and reported an positive effect from interacting with dogs for the state, but not trait, anxiety. Barker et al.(2005) reported a decline in both stress hormone levels, and measured stress and anxiety on a VAS (visual analog scale), from baseline, during and following intervention. A study performed by Handlin (2011) measured, beside OT, the level of Cortisol and Insulin at 7 points during interaction with dogs. Both Cortisol and Insulin levels decreased in both owners and controls over time. The increase in OT was only seen in owners, not in controls. Only dog owners displayed a decrease in hart rate. Altogether the study indicates a stress lowering effect from interaction with dogs.
All the above mentioned studies, and other conducted on the human-animal relationship, have mainly focused on dogs. The studies vary considerably in selection, settings, methodology, interventions and outcome. This causes challenges related to comparison between studies. The studies which have been published tend strongly towards a positive effect upon human health. What still remains uninvestigated, also after our study, is how the individual trait anxiety influences the recorded human stress response to the intervention with animals. This is especially important to explore when conducting research on interventions with farm animals.

The strengths of this study are the detailed protocol and procedures developed for the experiment. The principal researcher on site was a registered nurse and master student, and all staff involved were thoroughly briefed and trained in the procedures. Bio engineers sampled and handled the blood samples on site according to procedures. No deviations from the procedures were recorded at any point during the experiment. All samples were transported on dry ice to the laboratory and storage until analyses, which were performed in a well renowned laboratory at Oslo University Hospital, Norway.

Limitations

The study had few (n=18) participants and no control group. Possible biases are unknown personal information (oral contraceptives etc.) and selection. The experiment was conducted with live animals in a real setting, at the farm with the farmer and other study staff present. This makes it difficult to control all possible confounders as is the case for many studies conducted in live settings. Even though a significant effort was made to keep the level of scientific noise to a minimum, we cannot exclude this possible confounding effect. All data collection occurred within max thirty minutes, which the intervention it self took fifteen minutes. This is a very short time, considering that one of the possible OT sources is close contact with another living spices (human or animal) and bonding between them. One assume that dog and man have a inter-art bonding due to evolution (Wilson 1984), what may no be the case for cows and modern humans. The exposure to the cow for fifteen minutes might be too short, and it remains to be explored whether repeated exposure or a longer intervention will give a significant effect on the level of serum OT.

Conclusion

This study does not intend to fully explain the relationship between humans and farm animals. The lack of significant change in OT level merely suggests the complexity of human- animal relationship. The significant decline in state anxiety (STAI-SS) confirmed the results from previous studies on dog-human interactions. The positive correlation between the OT (T2-T3) score and STAI-SS (T1-T3) suggests that the effect from human – cow interaction has a later onset than human-dog. Farm animals are common in farms included in the concept of Green Care. Our study supports the assumption regarding the anxiolytic effect also from big, unfamiliar animals in a farm setting.

Further research

Further studies, with different target groups (participants), a control group, mixed approaches and a longitudinal design, are necessary to continue the journey towards an understanding of the human-animal companionship.

Future researchers might look further into the possibilities of sampling blood and measure other physical parameters in a less invasive way. A longitudinal design might also serve the purpose of mapping behaviour changes. Theories regarding Social support and Attatchment also need to be investigated to evaluate their impact on the special inter-art relationship between man and animal. Unrevealing the mystery of Oxytocin is only a part of it.

Litterature

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