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Oxytocin Enhances the Appropriate Use of Human Social Cues by the Domestic Dog (*Canis familiaris*) in an Object Choice Task

Jessica Lee Oliva[1], Jean-Loup Rault[2], Belinda Appleton[3], Alan Lill[1]

[1]School of Biological Sciences, Monash University, Melbourne, Australia; [2]Animal Welfare Science Centre, University of Melbourne, Australia; [3]Life and Environmental Sciences, Deakin University, Geelong, Australia

Abstract

It has been postulated that the neuropeptide, oxytocin, is involved in human-dog bonding. This may explain why dogs, compared to wolves, are such good performers on object choice tasks which test their ability to attend to, and use, human social cues in order to find hidden food treats. The objective of this study was to investigate the effect of intranasal oxytocin administration, which is known to increase social cognition in humans, on domestic dogs' ability to perform such a task. We hypothesized that dogs would perform better on the task after an intranasal treatment of oxytocin. Sixty-two (31 male; 31 female) pet dogs completed the experiment over two different testing sessions, five to fifteen days apart. Intranasal oxytocin or a saline control was administered forty-five minutes before each session. All dogs received both treatments in a pseudo-randomised, counterbalanced order. Data were collected as scores out of ten for each of the four blocks of trials in each session. Two blocks of trials were conducted using a momentary distal pointing cue and two using a gazing cue, given by the experimenter. Oxytocin enhanced performance using momentary distal pointing cues and this enhanced level of performance was maintained over 5-15 days time in the absence of oxytocin. Oxytocin also decreased aversion to gazing cues, in that performance was below chance levels after saline administration but at chance levels after oxytocin administration.

Keywords: Cognition, Cues, Dog, Oxytocin, Social

Introduction

Domestic dogs seem to have evolved specialised abilities to communicate with humans in a way that their progenitor, the wolf, cannot. Social cognitive intelligence has been postulated to underpin human evolution (Whiten & Erdal, 2012), and in relation to using human social cues, it may also have been important in the domestic dog's evolution from the wolf. The 'Object Choice Task' (OCT) was first applied to dogs by Miklósi et al. (1998) in an attempt to investigate dogs' ability to use human social cues, and has since been utilised in numerous studies of domestic dogs and various other canids. The OCT involves a human experimenter using non-verbal, social cues to indicate the location of a hidden piece of food, located in one of two objects, usually bowls, located to the right and to the left of them. The subject's task is to correctly use these cues in order to obtain the hidden reward. The cues can involve replica cards, marker placement, pointing, tapping, orienting to and/or gazing at the object for various lengths of time and from various distances.

Of all the pointing cues used, the momentary distal point is potentially the most informative with respect to canines' ability to use human communication signals, as it is the most challenging. This is because the distance from the experimenter's finger to the bowl is relatively large and the cue relatively brief. Indeed, the cue is delivered *before* the dog is released and allowed to make its choice and is only given for 1-2 seconds. As such, the dog has to rely not only on the cue itself, but also on its *memory* of the cue. Whilst domesticated dogs (Hegedüs, Bálint, Miklósi, & Pongrácz, 2013; Miklósi, et al., 2005; Schmidjell, Range, Huber, & Virányi, 2012; Soproni, et al., 2002; Virányi, et al., 2008) and socialized dingoes (Smith & Litchfield, 2010) generally perform above chance on the OCT when given the momentary distal point cue, young, hand-reared wolves that have been highly socialised to levels comparable with pet dogs do not (Miklósi, et al., 2003; Virányi, et al., 2008), or at least not without extensive training (Virányi, et al., 2008). An additonal study where domestic dogs were tested in the same outdoor conditions as wolves (as opposed to being tested indoors as in Miklósi, et al. (2003) and Virányi, et al. (2008)) suggests that the fact the wolves in these studies were tested outdoors may have handicaped them (Udell, et al., 2008a). However, this is somewhat contradictory, as in Udell, et al.'s study mature wolves with a high level of socialisation and involvement in public education programs *were* able to demonstrate above-chance performance on this task when tested outdoors. Indeed, the authors claim that the wolves even out-performed dogs tested in the same outdoor conditions, though their methodology has been criticised (for responses see, Hare, et al., 2010; Udell & Wynne, 2010a).

Of all the cues that have been used on domestic dogs, only one has yielded OCT performance *not* above-chance level: gaze (Soproni, Miklósi, Topál, & Csányi, 2001). These authors

demonstrated that domestic dogs did respond to gazing cues, but paradoxically avoided the bowl at which the experimenter gazed, rather than approaching it. This may reflect a behaviour learned from communicating with conspecifics. However, domestic dogs do demonstrate an ability to learn to use this cue correctly to solve the task over time (Miklósi, et al., 1998). With the exception of the gaze cue, most studies on the OCT in domestic dogs reveal no learning (Hare, et al., 2002; Miklósi, et al., 2005; Riedel, et al., 2006; Schmidjell, et al., 2012; Wobber, et al., 2009), even in 6, 8, 16 and 24 week old puppies (Riedel, et al., 2008). Although Riedel, et al's analysis has been criticised and upon independent re-analysis of the data, learning was found in to be present in the very young 6 week old puppies (Wynne, et al., 2008). Nonetheless, taken together, these findings suggest that dogs may have an inherent ability to perform at above chance level on tasks that require understanding of human cues, without training.

The superior ability of domestic dogs (in comparison with wolves) to perform OCTs may be due to the fact that they gaze significantly more at the humans than do wolves. This notion has been supported by the shorter latency to make eye-contact with an experimenter in both pet dog puppies (separated from their mothers at 6-9 weeks to live with a human family) and hand-reared dog puppies (separated from their mothers at 4-10 days and hand raised by humans who either kept them as pets or re-homed them) than in hand-reared wolf pups (separated from their mothers at 4-7 days and hand raised by humans who re-homed them to a wolf farm at 2-4 months) (Virányi, et al., 2008). More gazing has also been observed in pet dogs than in hand-reared wolves performing other problem-solving tasks (Miklósi, et al., 2003). Furthermore, similar findings have been obtained in anthropomorphically-viewed and treated companion dogs that glance more at their owners and perform less well on a problem-solving task than less anthropomorphically-viewed and treated working dogs (Topál, Miklósi, & Csányi, 1997). This suggests that companion dogs not only have the ability to use human cues to solve tasks and find food, but that they have become *dependent* on them. Only with extensive hand-rearing and training do wolves demonstrate an increase in gazing and in task execution that takes their performance to the level of naïve domestic dogs (Virányi, et al., 2008). Interestingly, gazing was also less frequent in domestic cats than in domestic dogs (Miklósi, et al., 2005), supporting the idea that the human-dog bond transcends the effects of domestication. This suggests that there is an inherent ability in dogs to communicate with humans in *humans' own way,* because gazing is a common phenomenon in human communication (Dickstein, et al., 1984; Striano, et al., 2006).

A link has been found between dogs that gaze at their owners for long durations and higher urinary oxytocin concentrations in the owner (Nagasawa, et al., 2009). Given that oxytocin is

implicated in mammalian bonding (Lim & Young, 2006), this suggests that a dog's gaze, imperative for the successful completion of the OCT, may be more prominent in more strongly bonded human-dog dyads. Oxytocin increases have been observed in both humans and dogs after human-dog interactions (Handlin, et al., 2011; Miller, et al., 2009; Odendaal & Meintjes, 2003) and are thought to be associated with human-dog bonding. Indeed, a new or enhanced role of oxytocin and/or its receptors in the domestic dog brain, compared to the wolf brain, may explain why dogs gaze at their owners more than hand-reared wolves do, and, in turn, do better than wolves in tasks involving human communicative signals. In humans, intranasal oxytocin administration: (a) enhances detection of human biological motion (Keri & Benedek, 2009), (b) increases the understanding of social cues and improves social memory (see reviews by, Bartz, et al., 2011; Guastella & MacLeod, 2012), (c) increases trust (Kosfeld, et al., 2005) and (d) increases a subject's gazing towards the eye region of other human faces (Guastella, et al., 2008), a phenomenon also observed in monkeys (Dal Monte, et al., 2014).

If dogs' ability to perform well on OCTs is dependent on their ability to look at humans and use human gestures, which is dependent on their central oxytocin function (as demonstrated in humans), increasing central oxytocin availability should improve their performance on OCTs. The aim of this study was to test the effect of intranasal oxytocin administration on dogs' performance on an OCT, using two different cues, momentary distal pointing and gazing (without head-turn). It was hypothesized that: (1) dogs would perform better on the OCT after an intranasal treatment with oxytocin than after a control saline administration when momentary distal pointing cues were given, and (2) oxytocin would both increase dogs' gazing toward the experimenter's eyes and their trust of the gaze cue, which would therefore improve their performance when gaze cues were offered as well. However, as Bartz, et al. (2011) highlight in their review of the pro-social effects of oxytocin in humans, increases in trust do not occur in situations with prior trust violations, out-groups or clinical populations who are rejection-sensitive. In these groups of people, trust was actually decreased by oxytocin administration. Therefore, whilst we did not expect the dogs in our study to fall into any of the categories mentioned above, we could not rule out the possibility that they would interpret the gazing cue negatively, and that this negative interpretation would be enhanced by oxytocin, thereby decreasing performance after oxytocin administration.

Method

Subjects. Seventy-five pet dogs (33 males, 42 females) were recruited for the study. Owners with healthy dogs over 12 months old were invited to participate, but owner-reported pregnant, lactating or visually-impaired dogs were excluded. Owners were recruited through poster

advertisements at Monash University Caulfield and Clayton campuses, as well as through university e-newsletters and social media websites. Dogs were randomly allocated into two separate groups: those that received oxytocin first and saline second (oxy-sal) and those that received saline first and oxytocin second (sal-oxy). Of the 75 dogs recruited, two males and 11 females did not complete the study; two dogs failed the pre-training, five dogs failed the test of motivation, three dogs passed the test of motivation but refused to continue when the more difficult cues were introduced, one dog was too excitable, and two dogs were withdrawn from the study by their owners. Partial data could, however, be used for two of the female dogs with incomplete records, leaving a total of 31 males and 31-33 females in the analysis. This study was approved by the Monash University School of Biological Sciences Animal Ethics Committee (BSCI/2013/07).

Materials. Twenty-four international units (equivalent to 50µg) of oxytocin (Auspep, Melbourne, AU) diluted in 0.5 ml of 0.09% saline, or 0.5 ml of 0.09% saline only (acting as a control) were administered to the nostrils of each dog, with a half-dose in each nostril. Treatments were delivered using a Mucosal Atomizer Device (MAD 300, Wolfe Tory Medical Inc., Salt Lake City, UT) connected to a 1mL syringe, while the dogs were maintained in a head-up position. When it could not be determined whether a dose was successfully administered, a second administration (half-dose) was delivered in the nostril concerned.

Two identical, opaque spaniel bowls (19cm base diameter, 11cm rim diameter, 12cm high, 8cm deep) were used to conceal the food treats. Spaniel bowls were selected for their height and ability to conceal the treat from the dogs' vision. Two additional and identical spaniel bowls were placed underneath the two testing bowls and treats identical to those used in the experiment were hidden in the space between them. This method was used by Udell, Giglio and Wynne (2008b) to ensure that both bowls smelled of the treats and the dog was consequently not able to rely on olfaction when making its choice between the bowls. Treats were also hidden around the testing room so that the entire room smelled of treats. The treats used were lamb puff cubes: light, low fat cubes of lamb lung, puffed with air. Scores were recorded by the experimenter using a pen and paper and the same experimenter conducted all testing of dogs in the investigation.

Procedure. On the day of the testing session, owners were asked not to feed their dog prior to participation so that motivation to perform the task was high. In cases where testing occurred in the afternoon, some dogs were fed a small snack in the morning at the owner's discretion. Owners and their dogs came to the testing location on two separate occasions, five to fifteen days apart. When they arrived for their first testing session, the dog received one of two intranasal treatments,

oxytocin or saline. When they arrived for their second session, the dog received the other intranasal treatment. Tubes containing the treatments were labelled 'A' or 'B', so that both the experimenter and the owner were 'blind' as to which treatment the dog received on which day. Order of treatment administration was pseudo-randomised and counterbalanced. The dog was restrained by its owner while the experimenter administered the intranasal spray. The owner was then required to fill out a few questionnaires to be used in an associated study while the dog was free to roam the testing room and interact with its owner or the experimenter. The owner and dog could then leave the room to wander outside or remain inside. Forty-five minutes after the treatment was administered, the first pre-training session commenced. A forty-five minute window was selected in accordance with the majority of previous human (MacDonald et al., 2011) and a recent pig (Rault, 2013) study and can be accepted as a sufficient time period in which neuropeptides can reach the brain (Born, et al., 2002; Rault, 2013).

Pre-training. The experimental set-up was similar to that of Virányi, et al. (2008). The two spaniel bowls were placed 1.5 m apart and the experimenter kneeled 30 cm behind the mid-point between the bowls. The dog, restrained by its owner, faced the experimenter at a distance of 2.5 m. The experimenter first got the dog's attention by calling its name or an affirmatory epithet ("good girl/good boy"; no address was used if the dog was already looking and calling the dog's name proved distracting to the dog). The dog was then shown a treat before it was placed in one of the bowls. The experimenter then said the release word "ok" (in some cases a different release word, more familiar to the dog, was used, such as "okay", "free", "take" , "go on", "(go) get it". The owner then released the dog and allowed it to approach one of the food bowls. If the dog approached the bowl containing the treat, it was allowed to eat the treat before both bowls were collected by the experimenter; if the dog approached the empty bowl or the experimenter, both bowls were collected by the experimenter and the dog did not receive a treat. The dog had to select the correct bowl four times in a row to move on to the testing session proper. A 10-minute cut off time was applied to the pre-training; if the dog was unable to pass the pre-training within this time, it was excluded from the study. Most pre-training sessions required only four trials and the maximum number required was 25 trials for one dog in one of its pre-training sessions.

Testing. The experimental set-up was the same as in pre-training. Each testing session contained four blocks of fifteen trials (10 where a cue was provided and five in which no cue to the treat's whereabouts was provided). The control condition was used to verify that the dogs were not relying on scent to find the hidden food. Numerous studies have found that performance is at

chance level when a control condition is employed (Hare, et al., 2002; Riedel, et al., 2008; Soproni, et al., 2002; Udell, et al., 2008b; Wobber, et al., 2009).

The first test block (B1) comprised, in sequence: three control trials, five trials with the momentary distal point cue, two control trials and then another five trials with the momentary distal point cue. The second test block (B2) comprised, in sequence: three control trials, five trials with the gaze cue, two control trials and then another five trials with the gaze cue.
The third test block (B3) was the same as the first (B1), and the fourth block (B4) was the same as the second (B2). The ordering of the blocks was such that the easier point cue was delivered first so as not to discourage the dogs from participating by delivering a difficult gaze cue straight away. Having only 10 trials per block was also strategically designed to keep the dogs motivated. Position of the correct bowl (left or right) was predetermined according to a pseudo-randomised chart that did not allow more than two consecutive trials where food could be obtained on the same side. Each test block was preceded by a pre-training session to maintain motivation to approach the baited bowl. The dog was allowed approximately five minutes of free play with its owner between testing blocks to avoid burnout.

Momentary distal point cue. The experimenter was kneeling, propped up on her toes, with her arms by her side. She got the dog's attention and then raised her ipsilateral arm and pointed (using her index finger) towards the correct bowl for 1-2 seconds, keeping her head straight, before lowering her arm back down to her side and saying "ok" (or an alternative release word). The approximate distance between the experimenter's index finger and the rim of the baited bowl was 42cm and 50cm to the treat inside. The dog was then released and allowed to make a choice between the bowls.

Gaze cue. The experimenter was kneeling with her arms by her side, the tops of her feet flat on the floor to achieve better eye-level with the dog. She got the dog's attention and then gazed towards the correct bowl for 1-2 seconds, keeping her head straight. She then said "ok" (or an alternative release word) and the dog was then released and allowed to make a choice.

Control condition. The kneeling experimenter, propped up on her toes, got the dog's attention, then kept her head straight for 1-2 seconds, then said "ok" (or an alternative release word) before the dog was released by its owner and allowed to make a choice in the absence of any cue.

Scoring. Scores were recorded as correct responses out of 10 trials per block (20 per cue) for each testing session. If the dog did not move within five seconds of being released, the cue was

given again, as in Virányi, et al. (2008), and the dog could be prompted to move by its owner. If no choice was made, the experimenter decided subjectively whether this was due to a distraction. If it was clearly due to a distraction, the trial was repeated. In cases where the experimenter was unsure why the dog did not make a decision, the test of motivation used by Udell, et al. (2008a) (two pre-training trials, one to each side) was conducted. If the dog was found to be unmotivated, the trial was discontinued; if the dog was found to be motivated, the trial continued and the experimenter assumed that the 'no choice' outcome of the previous trial was probably due to the dog not understanding the task, so that the score for that trial was 'incorrect choice'. The vast majority of dogs were found to be motivated (i.e. did not need to be tested for motivation) throughout the entire testing session, or were excluded from the study. Choices were also considered incorrect if the dog approached the incorrect bowl or the experimenter.

Statistical analysis. The raw scores for each testing block of the OCT performed after oxytocin and saline administration were entered into IBM SPSS Statistics version 22 (SPSS IBM, New York, U.S.A, 2013). Blocks one and three were also combined to give a total score for the pointing cues and blocks two and four were combined to give a total score for the gazing cues. One sample t-tests were used to investigate whether performance on the task was different from what would be expected by chance. To test for learning within each session, we compared the mean of the first 10 point and gaze cue trials (B1 and B2, respectively) with the last 10 point and gaze cue trials (B3 and B4, respectively) using paired samples t-tests. To test the effect of treatment, an independent samples t-test was run on session 1 only. The effect sizes of all significant t-tests were measured using Cohen's *d*. The effect of treatment (oxytocin, saline), gender (male, female) and group (oxy-sal, sal-oxy) on difference scores (score after oxytocin – score after saline) was evaluated using mixed model analyses of variance (ANOVA). The effect size of all significant F-tests was measured using partial eta squared. The assumption of homogeneity of covariances was tested using Box's *M* and was not violated for any test. Likewise, the assumption of homogeneity of variances was tested using F_{max} and the Levene's test and was met for all measures. Šidák-corrected pairwise comparisons (Abdi, 2007) were employed *post-hoc* to test for the effect of treatment in the oxy-sal group and sal-oxy group dogs separately, and to test the effect of treatment in male and female dogs separately.

Results

Performance different from chance. Control trials where the dog chose the left bowl, right bowl and correct bowl were scored out of a possible 20 choices per session and the means and standard deviations are given in Table 2.1.

Table 2.1. Mean (± standard deviation) object choices out of 20 and sample size for control trials.

	M	*SD*	*N*
Left after oxytocin	9.32	4.98	62
Right after oxytocin	10.08	5.16	62
Correct after oxytocin	9.35	2.04	62
Left after saline	9.22	5.27	63
Right after saline	10.14	4.99	63
Correct after saline	8.87	1.96	63

The dogs performed significantly below chance levels (score of 10) during both testing sessions, which demonstrates that they were not relying on olfactory cues to find the hidden food treat for the session after oxytocin administration (t_{61} = -2.49, *P*=.016, *d* = -0.32), and for the session after saline administration (t_{62} = -4.58, *P*<.0001, *d* = -0.58). There were no biases for the left bowl after oxytocin administration (t_{61} = -1.07, *P*=.29), the right bowl after oxytocin administration (t_{61} = 0.12, *P*=.90), the left bowl after saline administration (t_{62} = -1.17, *P*=.25) or the right bowl after saline administration (t_{62} = 0.23, *P*=.82).

Mean scores and standard deviations for each block(s) are given in Table 2.2.

Table2. 2. Mean (± standard deviation) correct object choices out of 20 for all dogs combined, according to treatment.

	Oxytocin			Saline		
	M	*SD*	*N*	*M*	*SD*	*N*
Point B1	7.41	1.86	63	7.41	2.29	64
Point B3	8.32	1.64	63	8.05	1.74	64
Point total	15.73	3.01	63	15.45	3.63	64
Gaze B2	4.68	1.67	63	4.59	1.49	64
Gaze B4	4.82	1.49	62	4.92	1.49	63
Gaze total	9.52	2.17	62	9.51	2.09	63

Dogs performed significantly better than chance (score of 5) on point B1 after oxytocin (t_{62} = 10.28, *P*<.0001, *d* = 1.30), point B3 after oxytocin (t_{62} = 16.01, *P*<.0001, *d* = 2.02), point B1 after saline (t_{63} = 8.39, *P*<.0001, *d* = 1.05) and point B3 after saline (t_{63} = 14.00, *P*<.0001, *d* = 1.75). However, dogs performed no differently from chance on gaze B2 after oxytocin (t_{62} = -1.51, *P*=.14), gaze B4 after oxytocin (t_{61} = -0.94, *P*=.35) and gaze B4 after saline (t_{62} = -0.46, *P*=.65), and significantly worse than chance on gaze B2 after saline (t_{62} = -2.19, *P*=.033, *d* = -0.28).

The influence of cue type of the effectiveness of treatment. A repeated measures ANOVA was run using total scores for each cue (point and gaze) to test whether performance using either one was more affected by the treatment than the other. There was a main effect of cue, (F1, 61 = 232.74, P<.0001, partial η^2 = .79). There was no main effect of treatment, (F1,61 = .20, P = .66) and no interaction between cue and treatment (F1, 61 = .30, P = .59).

Learning within sessions. Paired samples t-tests revealed that learning occurred within each session using the point cue, as the subjects performed better on B3 than B1 after oxytocin, (t_{62} = -3.95, *P*<.0001, *d* = -0.52) and after saline, (t_{63} = -2.79, *P*=.007, *d* = -0.32). No learning was demonstrated within each session using the gaze cue, as dogs performed no differently on B4 than B2 after oxytocin (t_{61} = -0.44, *P*=.66) or saline (t_{62} = -1.35, *P*=.18).

As learning appeared to be taking place between the pointing trial blocks, only data relating to the second block using point cues (B3) were used in the following analysis. As no differences were observed between the gaze trial blocks, they were combined in the analysis to follow.

The effect of oxytocin on performance. Mean correct choices for the second point block (B3) of trials and the gazing blocks combined for session 1 are shown in Table 2.3.

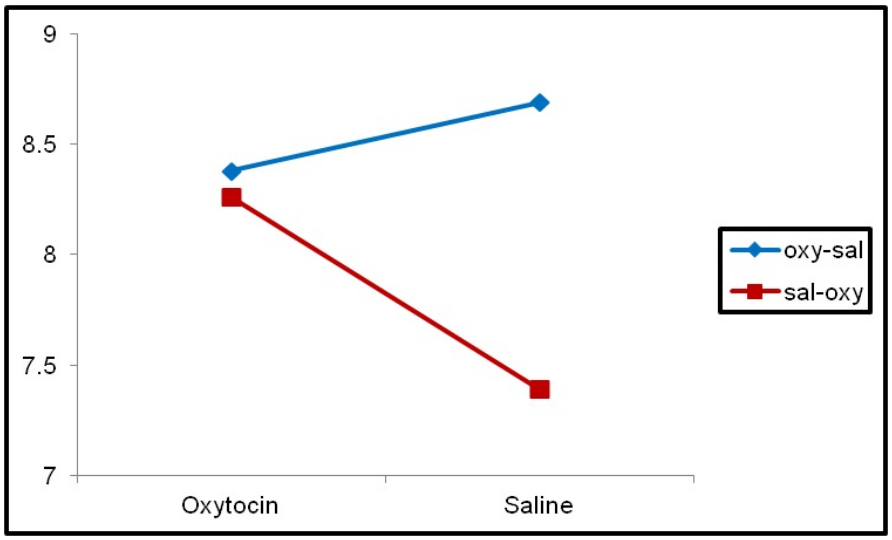
Table 2.3. Means (± standard deviation) correct choices out of 10 (point) and 20 (gaze) for dogs that received oxytocin and dogs that received saline on their first testing session.

	Oxytocin			Saline		
	M	*SD*	*N*	*M*	*SD*	*N*
Point B3	8.38	1.70	32	7.41	1.60	32
Gaze total	9.52	2.19	31	9.19	1.75	32

Independent samples t-tests were used to compare means between the dogs that received oxytocin and dogs that received saline in this session. The t-test revealed that dogs that received oxytocin on their first testing session performed significantly better than dogs that received saline on their first testing session for B3 (t_{62} = 2.35, *P*=.022, d=0.47). No significant difference was observed between the two groups of dogs for the gazing cues (t_{61} = 0.66, *P*=.51).

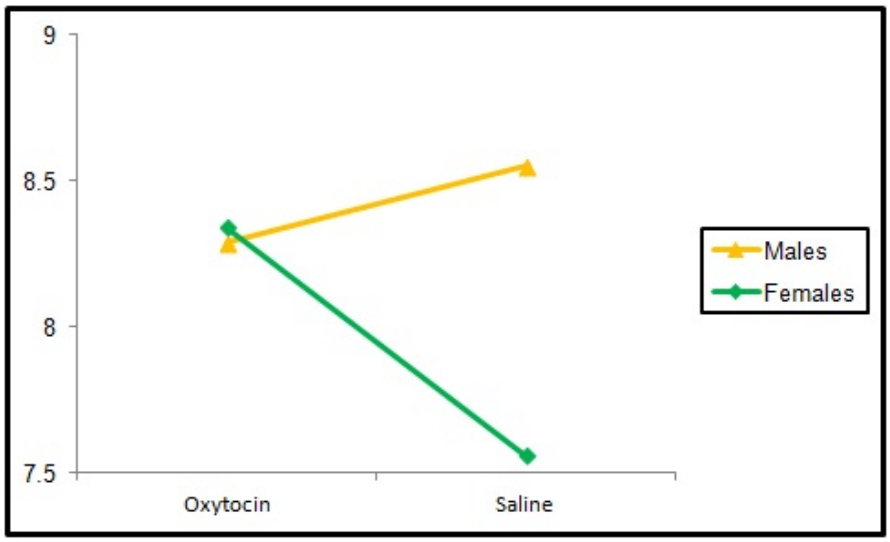
The effect of gender, group and treatment on performance. Examination of Figure 2.1 indicates that dogs in both groups performed similarly with the pointing signal in B3 after oxytocin but differently after saline, in that the oxy-sal dogs' performance improved after saline and the sal-oxy dogs' performance declined.

Figure 2.1. Mean point B3 scores (out of 10) for oxy-sal group and sal-oxy group dogs, according to treatment



There is a similar pattern in Figure 2.2 for males and females, in that males and females perform similarly after oxytocin, but male dogs' performance slightly improves after saline, whereas female dogs' performance after saline declines.

Figure 2.2. Mean point B3 scores (out of 10) for male and female dogs, according to treatment



A mixed model ANOVA revealed a significant interaction between treatment × group ($F_{1, 59}$ = 6.40, P=.014, partial η^2= .10) and treatment × gender ($F_{1, 59}$ = 5.01, P=.029, partial η^2= .08), but not among treatment × group × gender ($F_{1, 59}$ = 0.77, P=.013). There were no significant main effects of treatment ($F_{1, 59}$ = 1.36, P=.25), or gender ($F_{1, 59}$ = 1.75, P=.19), but there was a significant main effect of group ($F_{1, 59}$ = 4.35, P=.041, partial η^2= .07). Dogs that were administered oxytocin first performed more poorly after oxytocin than saline (mean difference = -0.31, SD = 1.87), whilst dogs that were administered saline first performed better after oxytocin than after saline administration (mean difference = 0.87, SD = 1.88). Male dogs performed worse after oxytocin administration (mean difference = -0.26, SD =1.81), but female dogs performed better (mean difference = 0.78, SD =1.98). Four Šidák-corrected pairwise comparisons were conducted using an adjusted alpha of .013 (1-tailed). Difference scores between treatments were significant in sal-oxy group dogs (t_{30} = 2.59, P=.0075, d = 0.54), but not oxy-sal group dogs (t_{31} = -2.40, P=.18), female dogs (t_{31} = 2.23, P=.0165) or male dogs (t_{30} = -0.80, P=.22).

For the gaze total scores, a mixed model ANOVA revealed no significant interaction effects between treatment × group ($F_{1, 58}$ = .50, P=.48), treatment × gender ($F_{1, 58}$ = .61, P=.44) and among treatment × gender × group ($F_{1, 58}$ = .57, P=.45). Nor were there any significant main effects of treatment ($F_{1, 58}$ = .01, P=.96), gender ($F_{1, 58}$ = .001, P=.98) or group ($F_{1, 58}$ = 0.73, P=.40).

Discussion

The ability of dogs to use momentary distal pointing cues, and the effect of oxytocin. Consistent with previous research, this study demonstrated an ability of domestic dogs to use

momentary distal pointing cues to find hidden food in an OCT (Hegedüs, et al., 2013; Miklósi, et al., 2005; Schmidjell, et al., 2012; Soproni, et al., 2002; Virányi, et al., 2008). In addition, consistent with our first hypothesis, a treatment effect was observed in that dogs performed significantly better after oxytocin than saline administration in session 1. This is consistent with findings for humans demonstrating that oxytocin increases perception of biologically relevant human motion (Kéri & Benedek 2009) which is imperative for social cognitive processing and communication, and supports the notion that oxytocin increases social cognition (see reviews by Bartz, et al., 2011; Guastella & MacLeod 2012). In addition, when examining difference scores between testing sessions, we observed performance improvements from session 1 to session 2 for point B3 scores in sal-oxy group dogs. Inspection of Figure 2.1 shows that their performance in session 2 was only bought up to the level of performance demonstrated by the oxy-sal group dogs in session 1, after oxytocin administration. The absence of a significant difference between sessions for the oxy-sal group dogs indicates that this group of dogs was able to maintain their performance at this level 5-15 days later, after saline administration. Thus oxytocin not only enhanced performance on the OCT, but the enhanced level of performance was maintained over time.

The effect of gender on the efficacy of oxytocin. The enhancing effect of oxytocin seems to have been driven by the female subjects in this study who performed better after oxytocin and more poorly after saline administration (see Figure 2.2). The reason why males were possibly not as influenced by oxytocin as females (whose performance was able to be brought up to the level of the males after oxytocin administration) may simply be ceiling effects, as they performed similarly after both treatments and significantly better than females after saline administration. The reason for the superior performance of male dogs compared to females after saline administration is unknown and somewhat surprising; in humans, females have shown greater social cognitive abilities than males, as demonstrated by their better perception of others' emotions (Brabec, Gfeller, & Ross, 2012; Donges, Kersting, & Suslow, 2012). However, the OCT differs in that it tests an ability to solve a task using human communicative cues, not human emotions. Estrogen is known to enhance the production of oxytocin and its receptor (Rissman, 2008) and this may explain why the female dogs in this study did not did not perform as well as human female subjects in other tests of social cognition, as the majority (88%) had been spayed, thereby reducing the volume of estrogen their bodies would be producing. However this does not explain why the male dogs (the majority of whom had also been neutered, 97%) performed so much better than the females dogs following saline.

The ability of dogs to use gazing cues, and the effect of oxytocin. Contrary to our second hypothesis, no treatment effect was observed for gazing cues. We did find some support, however,

for the negative interpretation of the gaze cue being dampened by oxytocin. For example, in gaze B2 after saline administration we obtained the same findings as Soproni, et al. (2001), who reported that dogs interpreted the gaze cue negatively, avoiding the bowl to which the experimenter gazed. Our lack of a similar finding for B2 after oxytocin administration supports our hypothesis that oxytocin increases trust in the dog, as it does in humans (Kosfeld, et al., 2005), despite the fact that the dogs were unable to use the cue, performing no better than chance after oxytocin administration. That this below-chance level performance was lacking in gaze B4 after saline administration may reflect the dogs learning that no aversive consequences would occur when they went to the bowl containing the treat, so they no longer used the gaze cue to complete the task, and just guessed.

The Clever Hans phenomenon. Whilst mean performance on the majority of the gaze cue blocks was at chance level, it is intriguing that mean control trial performance (where no cue was given) was *below* chance levels, as shown in Table 2.1. The so called 'Clever Hans' phenomenon (Pfungst, 1911), involving some form of unintentional or subconscious cueing from the owner, has been independently tested for in dogs subjected to an OCT with momentary distal pointing cues, and yielded negative findings (Hegedüs, et al., 2013; Schmidjell, et al., 2012), but we cannot completely rule this out as the reason for these unexpected results. The above-mentioned studies only tested for possible unintentional, subconscious cueing by the owner, not by the experimenter. In the current study it is conceivable that the experimenter was subconsciously 'hoping' that the dogs were not using scent to find the food in the control trials and may have been unintentionally cueing the dogs to go to the empty bowl in order to validate the experimental design. This highlights the critical importance of blind treatment testing for both the owner and experimenter, which was a strength of the current study. Nonetheless, the effect of the experimenter on the Clever Hans phenomenon warrants further study.

Learning within sessions. Another unexpected finding in the study was that of the learning observed within sessions for the pointing cues. Despite the pre-training that took place before B1, it appears that dogs were still learning to use the point cues to do the task in B1 compared to B3, where they performed better. This finding contrasts with those of previous studies, which did not report performance differences within sessions (Hare, et al., 2002; Miklósi, et al., 2005; Riedel, et al., 2006; Riedel, et al., 2008; Schmidjell, et al., 2012; Wobber, et al., 2009). As learning was observed within both treatment sessions, we do not believe this is a consequence of the oxytocin administration unique to our study. One possibility is that this 'learning' is a reflection of the dogs being less anxious about the novel environment in B3 compared to B1, and therefore less inhibited

in performing the task. However, it is still interesting that this was observed in our study but not in previous studies which employed similar testing methods and less habituation time to the testing environment. This disparity may be due to the fact that testing in the current study was carried out in a room that was not well insulated against distracting external sound disturbances, and therefore may have required more habituation time than the testing locations employed in other studies.

Limitations and future directions. The above-mentioned external sound disturbances may have varied on different testing days, which was an unavoidable limitation of our study. Other limitations of the present investigation included possible variation in the dog's hunger levels among sessions. Although efforts were made to test a particular dog at the same time of day in each session, this was not always possible. Owners were also instructed to keep the dog's day as similar as possible between sessions, but this could not be fully controlled either. Given the gender differences we observed, future studies should consider the effect that spaying and neutering has on oxytocin function, as our findings, compared to those of human studies on social cognition, may suggest this has particular influence in females. Lastly, although efforts were made to be as consistent as possible with the majority of previous studies' dosages and behavioural testing timeframes, it is currently unknown what constitutes the optimal behavioural testing time after administration of oxytocin in dogs, and how long the behavioural effects last. Extrapolating from the findings of a human study investigating the intranasal application of 40IU and 80IU of a very similar peptide, vasopressin (Born, et al., 2002), and a recent pig study investigating the intranasal application of 24IU of oxytocin (Rault 2013), we can reasonably assume that oxytocin is still active in the brain 100-120 minutes after administration, and potentially longer. Therefore the behavioural effects in the current study were likely to have been maintained for the entire testing session, which normally lasted between 90 and 120 min.

Conclusion

Administration of oxytocin was effective in aiding dogs' performance on the OCT using momentary distal pointing cues. Moreover, this enhancing effect persisted at least 5-15 days later, in the absence of further oxytocin administration. Oxytocin also appeared to decrease dogs' aversion to gazing cues, with performance being at chance level after oxytocin administration but below chance level after saline administration.

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