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Pro-social ultrasonic communication in rats: Insights from playback studies

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HIGHLIGHTS

- Rat ultrasonic vocalizations (USV) serve as situation-dependent affective signals with important communicative functions.
- All studies using the 50-kHz USV radial maze playback paradigm indicate that 50-kHz USV serve as social contact calls.
- This playback paradigm allows studying pro-social ultrasonic communication in a reliable and highly standardized manner.

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ABSTRACT

Rodent ultrasonic vocalizations (USV) serve as situation-dependent affective signals and convey important communicative functions. In the rat, three major USV types exist: (I) 40-kHz USV, which are emitted by pups during social isolation; (II) 22-kHz USV, which are produced by juvenile and adult rats in aversive situations, including social defeat; and (III) 50-kHz USV, which are uttered by juvenile and adult rats in appetitive situations, including rough-and-tumble play. Here, evidence for a communicative function of 50-kHz USV is reviewed, focusing on findings obtained in the recently developed 50-kHz USV radial maze playback paradigm. Up to now, the following five acoustic stimuli were tested in this paradigm: (A) natural 50-kHz USV, (B) natural 22-kHz USV, (C) artificial 50-kHz sine wave tones, (D) artificial time- and amplitude-matched white noise, and (E) background noise. All studies using the 50-kHz USV radial maze playback paradigm indicate that 50-kHz USV serve a pro-social affiliative function as social contact calls. While playback of the different kinds of acoustic stimuli used so far elicited distinct behavioral response patterns, 50-kHz USV consistently led to social approach behavior in the recipient, indicating that pro-social ultrasonic communication can be studied in a reliable and highly standardized manner by means of the 50-kHz USV radial maze playback paradigm. This appears to be particularly relevant for rodent models of neurodevelopmental disorders, as there is a tremendous need for reliable behavioral assays with face validity to social communication deficits seen in autism and schizophrenia in order to study underlying genetic and neurobiological alterations.

1. Introduction

Mice and rats produce vocalizations in the ultrasonic range, i.e. in a frequency range clearly above the human hearing threshold and therefore not audible to humans, often referred to as ultrasonic vocalizations (USV). Rodent USV serve as situation-dependent affective signals (for reviews see: Brudzynski, 2013; Wöhr and Schwarting, 2013) and have attracted considerable interest in the behavioral neuroscience research community, particularly because it was suggested that they may serve as a direct measure of affect, i.e. of the animal's subjective emotional state (e.g. Panksepp, 2005; for review see: Burgdorf et al., 2011). Furthermore, since rodent USV convey important communicative information as alarm and social contact calls, measuring USV provides the opportunity to study communication in rodent models for neuropsychiatric disorders characterized by social and communication deficits, most notably autism and schizophrenia (e.g. Won et al., 2012; for review see: Scattoni et al., 2009).

Typically, USV in rodents are categorized depending on the animal's developmental stage and acoustic call features, such as call duration and peak frequency. In the rat, the following three major USV types are most commonly differentiated: (I) 40-kHz USV, which are emitted by pups during social isolation when separated from mother and littermates (e.g. Wöhr and Schwarting, 2008a); (II) 22-kHz USV, which are produced by juvenile and adult rats in aversive situations, including social defeat (e.g. Sales, 1972a), predator exposure (e.g. Blanchard et al., 1991), and fear learning (e.g. Choi and Brown, 2003); and (III) 50-kHz USV, which are uttered

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by juvenile and adult rats in appetitive situations, including rough-and-tumble play (e.g. Knutson et al., 1998), mating (e.g. Sales, 1972b), and when tickled playfully (e.g. Panksepp and Burgdorf, 2000). In the mouse, similar USV types exist, with the exception of 22-kHz USV, which have not been detected in mice yet, despite tremendous effort and numerous USV studies focusing on aversive situations (for review see: Wöhr and Scattoni, 2013).

In support of the notion that rodent USV in juvenile and adult rats may serve as a direct measure of affect, 22-kHz USV and 50-kHz USV emission were repeatedly linked to components of the behavioral repertoire associated with negative and positive affect, respectively. Specifically, in aversive situations, such as fear learning, 22-kHz USV production typically correlates with freezing behavior, the most commonly used measure of fear in rats (Wöhr and Schwarting, 2008a,b), and depends on the individual disposition to display anxiety-related behavior, with rats characterized by high levels of anxiety emitting more 22-kHz USV during fear learning than less anxious rats (Borta et al., 2006). Furthermore, 22-kHz USV production was shown to systematically covary with aversiveness, experimentally manipulated by means of foot shock intensities (Wöhr et al., 2005). In this dose–response study, rats exposed to higher foot shock intensities were found to emit more 22-kHz USV than rats exposed to lower shock intensities during fear learning and testing, i.e. in absence of shock application when exposed to a formerly neutral stimulus that has been repeatedly paired with foot shocks.

In contrast, in appetitive situations, such as rough-and-tumble play, 50-kHz USV production is positively correlated with play behavior in juveniles (Burgdorf et al., 2008). In line with this observation, rats bred for low rates of 50-kHz USV were found to display altered rough-and-tumble play behavior (Webber et al., 2012) and a depression-like behavioral phenotype, along with elevated levels of anxiety (Burgdorf et al., 2009). Furthermore, individual differences in the emission of 50-kHz USV have been linked to response strategies in an ambiguous cue interpretation paradigm (Rygula et al., 2012) and hippocampal cell proliferation (Wöhr et al., 2009), with rats emitting high rates of 50-kHz USV displaying an “optimistic” bias and more newborn cells in the hippocampus. In contrast, rats characterized by a depression-like behavioral phenotype with low rates of 50-kHz USV were found to have markedly lower numbers of newborn cells in the hippocampus. This is in line with findings from preclinical treatment studies showing that hippocampal neurogenesis is required for the efficacy of major antidepressant drugs, such as selective serotonin reuptake inhibitors (Santarelli et al., 2003).

2. Pro-social function of 50-kHz USV

Besides studies linking USV emission and affective state, there is a wealth of evidence supporting the idea that USV serve important communicative functions. It fact, it is well established that isolation-induced 40-kHz USV emitted by pups serve as social contact calls and elicit maternal search and retrieval behavior (e.g. Allin and Banks, 1972; for review see: Hofer and Shair, 1993). It is further widely believed that 22-kHz USV emitted in aversive situations, such as predator exposure, serve as alarm calls (e.g. Blanchard et al., 1991; for review see: Litvin et al., 2007). Finally, there is evidence that 50-kHz USV serve communicative functions as well.

Most early studies focusing on a potential communicative function of 50-kHz USV were conducted in the sexual context (for review see: Barfield and Thomas, 1986). The most commonly used approach to assess the effects of 50-kHz USV was devocalization by cutting the laryngeal nerves. In such devocalization experiments it was shown that females display less darting behavior when exposed to a devocalized male mating partner and are more likely to move away while being mounted by a male unable to vocalize (Thomas et al., 1981, 1982; White and Barfield, 1990; White et al., 1990). By means of combining devocalization and playback of 50-kHz USV, White and Barfield (1990) further showed that female receptive behavior is restored when they are exposed to tape-recorded male 50-kHz USV. Specifically, they reported that playback of male 50-kHz USV leads to an increase in the level of immobility shown by the female exposed to a devocalized mating partner that is sufficient for the male to obtain an intromission. In addition, Thomas et al. (1982) found that females display more darts directed toward devocalized males when male 50-kHz USV are played in their proximity. In line with this, Geyer et al. (1978) found that darting is facilitated in females exposed to male 50-kHz USV before mating and McIntosh et al. (1978) demonstrated that darting can even be enhanced by male 50-kHz USV in females receiving low levels of sexual stimulation while being confronted with a castrated mating partner. Moreover, Barfield et al. (1979) used a T-maze apparatus and demonstrated that females spent more time in the one of the two arms in which male 50-kHz USV were presented. However, in a very recent study, Snoeren and Ågmo (in press) did not detect clear behavioral changes in females exposed to playback of male 50-kHz USV and concluded that male 50-kHz USV have a most limited, if any, incentive value for sexually receptive females. Snoeren and Ågmo (in press) conducted three experiments. In the first two experiments, 50-kHz USV recorded from sexually active males during the precopulatory phase were used as playback stimuli. In each of the two experiments, four different acoustic stimuli were used: background noise, a natural 10 min sequence of male 50-kHz USV, a flat 50-kHz call, and a frequency-modulated 50-kHz call, with the latter two stimuli being presented repeatedly. The results were inconsistent and the overall effects of 50-kHz USV playback were found to be weak. While in the first experiment, a small, but significant preference toward the speaker was seen in response to the flat 50-kHz USV, but not the other acoustic stimuli, significant preference scores were obtained in response to all acoustic stimuli, with exception of background noise, in the second experiment. However, when comparing the time spent in front of the speaker with the time spent on the opposite side, significant approach behavior was seen in response to the natural 10 min sequence of male 50-kHz USV in the second experiment only. In the third experiment, a devocalization study, females did not display a preference for a non-devocalized over a devocalized male. Besides changes in overt behavior, USV emission by the recipient female was studied. White et al. (1993) found that male 50-kHz USV elicit USV in females, but such a response was not seen in another study (White et al., 1991). Finally, female USV emission is also believed to play an important role in coordinating sexual activity (White and Barfield, 1987, 1989; White et al., 1991), yet again, findings by Snoeren and Ågmo (2013) do not support such a communicative function. Overall, however, the main proportion of studies focusing on a potential communicative function of 50-kHz USV indicate that male 50-kHz USV play an important role in establishing proximity between mating partners and that they possibly orchestrate sexual activity. Yet, in contrast to the sexual context, strong evidence for a communicative function of 50-kHz USV in other social situations was almost completely lacking until recently.

The idea that 50-kHz USV serve communicative functions in the social context was born out of studies showing that remarkably high 50-kHz USV emission rates occur during rough-and-tumble play in juveniles (Burgdorf et al., 2008; Knutson et al., 1998; Webber et al., 2012) and that young rats spent more time with adults displaying higher levels of 50-kHz USV than with conspecifics characterized by limited 50-kHz USV emission rates (Panksepp et al., 2002). This view is further supported by the fact that deafening juvenile rats affects rough-and-tumble play (Siviy and Panksepp, 1987). Siviy and Panksepp (1987) showed that deafening