Behavioural Brain Research 350 (2018) 80–86

Contents lists available at ScienceDirect

Behavioural Brain Research

journal homepage: www.elsevier.com/locate/bbr

Research report

Individual behavioural predictors of amphetamine-induced emission of 50 kHz vocalization in rats

Kevin G. Mulvihill*, Stefan M. Brudzynski

Department of Psychology, Brock University, St. Catharines, ON, L2S 3A1, Canada

ARTICLE INFO

Keywords:
Ultrasonic vocalization
Long Evans rats
50 kHz vocalization subtypes
Trill calls
Systemic amphetamine
Latency to approach reward
Prediction
L-maze

ABSTRACT

Measurement of ultrasonic vocalizations (USVs) produced by adult rats represents a highly useful index of emotional arousal. The associations found between 50 kHz USV production and a variety of behavioural and pharmacological protocols increasingly suggests they serve as a marker of positive motivational states. This study used a powerful within-subjects design to investigate the relationships among individual differences in approach to a sweet-food reward, predisposition to emit 50 kHz USVs spontaneously, and 50 kHz USVs emission following acute systemic administration of amphetamine. Both approach motivation and predisposition to call were found to not correlate with each other but did predict 50 kHz USV response to acute amphetamine. These two behavioural phenotypes appear to represent dissociable predictors of acute amphetamine-induced emission of 50 kHz USVs in a non-sensitization paradigm. In contrast to that, a measure of sucrose preference was not found to predict 50 kHz USV emission following amphetamine. Acute amphetamine was also found to increase average sound frequency of emitted USVs and selectively increase the proportion of Trill subtype 50 kHz USVs. Together, these data demonstrate that acute amphetamine-induced 50 kHz USVs in the adult rat represent more than just a univariate motivational state and may represent the product of dissociable subsystems of emotional behavior.

1. Introduction

Adult rats emit vocalizations in the ultrasonic frequency range that serve to convey their motivational and emotional state to other conspecifics [1,2]. This has made the detection and measurement of these ultrasonic vocalizations (USVs) a highly useful index of emotionality in a wide variety of experimental settings [3]. The recorded emission of so-called 50 kHz USVs provides a quantifiable metric of positive emotional states that does not require conditioning [1,2]. This call type is elicited by appetitive, positive behavioural situations [4,5], rewarding brain stimulation [6,7], and administration of psychostimulants, e.g., amphetamines [8,9]. Due to the unconditioned nature and generalizable characteristics of this form of behavior, recording of 50 kHz USVs has been utilized in many models of drug addiction and sensitization [8–13]. The emission of 50 kHz USVs is generally dependent on the activity of the mesolimbic dopamine system [7,14], and associated with increased dopamine activity in the nucleus accumbens (NAc; [15–17]).

50 kHz USVs are categorized and characterized by their respective acoustic parameters, with the degree of sound frequency modulation appearing to be the best index of positive emotional arousal [1,2,8,18,19]. Due to differing degrees of frequency modulation observed across individual calls, 50 kHz USVs can be subdivided into 'flat' (non-modulated) and frequency-modulated (FM) call subtypes. These call subtypes are characterized by different sonographic profiles though they share the same average peak frequency range around 50–55 kHz [1,2,18]. These call subtypes are associated with distinct behaviours and contexts, with FM calls most strongly related to reward-associated contexts such as psychostimulant administration [5,20–22]. It has been suggested that 50 kHz USVs may represent motivational markers whereby they are elicited in association with the expectation/anticipation of a reward [6,23,24]. This is supported by findings of anticipation-related 50 kHz USV emission to psychostimulants which directly stimulate dopamine release in the NAc [11,22,25,26]. Moreover, beyond drugs that directly stimulate the mesolimbic dopamine system, there is evidence of non-DAergic agents (e.g. morphine and sucrose) having the capacity to elicit anticipatory or conditioned 50 kHz USVs [9,11,27].

Two conceptual components related to an animal's response to a given reward are the hedonic value (the 'liking') and the motivation to consummate (the 'wanting') [28,29]. In the present experiment, an L-maze apparatus was used. The L-maze removes much of the aspect of reward learning typically associated with T-maze performance [30].

* Corresponding author.
E-mail address: km07gk@brocku.ca (K.G. Mulvihill).

https://doi.org/10.1016/j.bbr.2018.05.009
Received 29 March 2018; Received in revised form 9 May 2018; Accepted 10 May 2018

Evidence from rodents in the T-maze indicates that differences in dopamine levels result primarily in differences in the motivation to obtain reward [31–33]. Thus, a measure of latency in the L-maze should index an individual rats' *motivation* to approach and consume a food reward and therefore should strongly positively correlate with individual differences in 50 kHz USV production.

The consumption of sucrose has been used widely as a measure of hedonia in rats [34,35] and appears dependent on the function of dopamine in the NAc [36,37]. Sucrose solutions, that are readily self-administered and consumed, induce significant 50 kHz USV production though without the same sensitization effect noted with cocaine self-administration [11]. Positive relationships are found between individual differences in sucrose intake and psychostimulant self-administration (cocaine and Damphetamine; AMPH) [38–40]. Moreover, there have been findings of positive relationships between individual differences in the preference for sucrose and high 50 kHz USV emission behavioural phenotype [35,41,42]. Following selective breeding based on 50 kHz USV emission, the high-line rats (which more readily emit 50 kHz calls) showed evidence of greater sucrose preference compared with random-line rats [42]. Thus, a measure of sucrose preference should index a rats' individual *hedonic response* and should positively correlate with individual differences in 50 kHz USV production, providing some accounting for the 'liking' aspect of reward.

Amphetamine (AMPH) has been extensively used as a psychostimulant inducer of 50 kHz USVs with strong individual differences in response to systemic administration both in regard to call rate ('low callers' versus 'high callers') as well as subtypes emitted ('call profile') [22,43–45]. Additionally, Engelhardt et al. [46] recently found evidence of a positive relationship between spontaneous and AMPH-induced 50 kHz call emission. Moreover, these inter-individual differences in baseline 50 kHz USV production were potentiated by injections of AMPH and appeared related to trait-like differences in approach behaviour to 50 kHz USV playback. It should also be noted that inter- and intra-individual differences in calling extend beyond AMPH-induction and are found when 50 kHz USVs are elicited via non-pharmacological methods [47]. High caller rats that produce more 50 kHz USVs after systemic AMPH administration also show conditioned place preference to AMPH and a greater proportion of FM calls after AMPH when compared to low callers or controls [22,45]. In contrast, it has been reported by several researchers that locomotor activity and emission of 50 kHz USVs appear to be dissociable behavioural phenotypes with only partial overlap [9,12,22,45,46]. High caller rats that show behavioural sensitization to repeated AMPH injections in their production of 50 kHz USVs do not necessarily show locomotor sensitization of a comparable nature [22,45]. A similar dissociation between reward related behaviours is observed between 50 kHz USV emission after AMPH and social play behaviour, whereby the individual behavioural phenotypes for each behaviour are not necessarily positively correlated [48,49]. These results indicate that multiple subsystems likely underlie an individual animal's orientation to rewards and raise the question of what AMPH-induced 50 kHz USV emission represent.

The present study set out to determine if the aspects of reward (liking and wanting) and USV production (individual predisposition to call) could be dissociated in a non-sensitization model of acute AMPH induction of 50 kHz calling. To date there appears to be a strong convergence of evidence indicating 50 kHz USVs as motivational markers indexing the 'wanting' component of reward [6,11,22,25]. This focus on the motivational aspect of 50 kHz USVs is especially the case in regards to research involving drugs of abuse [8,26]. However, there are discrepancies in this narrowed notion of 50 kHz USVs as signals of individual motivation phenotype as they are often dissociable from other reward-related behavioural phenotypes [13,49]. Additionally, most research that has recently found a dissociation of AMPH-induced calling behaviour and reward-related behavioural phenotypes has employed repeated administration protocols of sensitization [9,44,45]. To uncover the nature of what AMPH-induced 50 kHz USVs represent in regards to individual characteristics associated with reward and calling, the present study uses a non-sensitized, non-anticipatory AMPH-induction protocol and a within-subjects design. For each animal measures of hedonic drive (sucrose preference), motivation for reward (latency to approach and consume a food reward in an L-maze), and predisposition to emit 50 kHz USVs (calls after saline) were used to predict call rate after 1.5 mg/kg of systemic AMPH. It was hypothesized that call rate after AMPH would reflect a measure of an individual's motivation to approach reward, their hedonic drive, and also their predisposition to emit 50 kHz USVs. These AMPH-induced 50 kHz USVs thus may represent a general positive emotional state rather than simply motivational drive. Therefore, it was hypothesized that each of these variables would predict USV response after AMPH in a dissociable fashion (see Fig. 1).

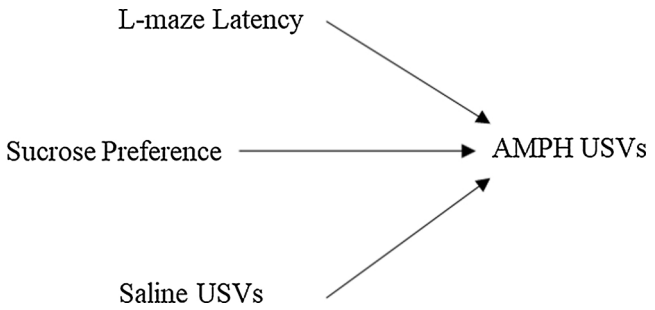

Fig. 1. Diagrammatic representation of predictive model representing L-maze Latency, Sucrose Preference, and Saline USVs as independent predictors of the response to AMPH measured via USV rate.

2. Methods

2.1. Subjects

Forty-six male Long Evans rats (Charles River Laboratories, Saint-Constant, QC, Canada) were used for all behavioural procedures. All animals were approximately 53 (± 1) days old at the beginning of the study with an average body weight of 288 g (*SD* = 26.1 g, min. = 234 g, max. = 344 g). At the end of the study animals had an average body weight of 390 g (*SD* = 42.1 g, min. = 315 g, max. = 474 g) and were approximately 78 (± 1) days old. In accordance with Brock University protocols for laboratory handling, all animals were housed in polycarbonate cages (19" × 10½" × 8") with dust-free beta-chip® bedding (autoclaved wood fibers, Northeastern Products Corp., Warrensburg, NY, USA) and with a plastic tube inside for hiding. The housing room was maintained with controlled room temperature (23 °C ± 2 °C) and humidity (40–60%) conditions. Subjects were housed in pairs with a maintained 12:12 light/dark cycle and *ad libitum* access to food and water. Animals were gently handled once before the initial behavioural procedure. All research protocols were approved by Brock University Animal Care and Use Committee and complied with guidelines and policies set forth by the Canadian Council on Animal Care.

2.2. Procedure overview

All animals underwent behavioural procedures in the same order, with the procedures separated by 3 days between them. The behavioural procedures involved measuring sucrose preference (using 10% w/v solution versus water) and latency to approach a palatable food cue in seconds (s) before receiving subcutaneous injections followed by recording of ultrasonic vocalizations (USVs) on two separate occasions. The first injection and recording was saline (0.2 ml), 3 days later the animals received an injection of amphetamine (1.5 mg/kg in 0.2 ml saline s.c.).