Differential-Reinforcement-of-Low-rates-of-responding-task performance in a transgenic rat model of Huntington disease

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Huntington's Disease (HD) is an autosomal dominantly inherited progressive disorder which is characterized by psychiatric changes, dementia and motor dysfunction [1]. No effective treatment to influence the onset or the progression of this fatal disease is currently available. The behavioural characterization of a sensitive rodent model for HD is critical, especially in light of the development of therapeutic strategies for HD.

In Tuebingen (Germany), Nguyen's group has generated an HD transgenic rat model using a human bacterial artificial chromosome (BAC), which contains the full-length HTT genomic sequence with 97 CAG/CAA repeats and all regulatory elements. BACHD transgenic rats display a robust, early onset and progressive HD-like phenotype including motor deficits and anxiety-related symptoms [2] rendering this model valuable for further phenotype studies.

One of the aspects our research group is interested in is the evaluation of the impulsivity of the BACHD rats. Impulsivity is not a unitary trait, but can be subdivided in two types: impulsive action (IA) is defined as the inability to withhold a prepotent motor response; on the other hand impulsive choice (IC) is the excessive discounting of delayed reinforcement [3]. It is known that basal ganglia are important for the inhibition of unnecessary or inappropriate neural responses: chorea, perseveration during strategy shifts, behavioural disinhibition and impulsivity are the main features that affect HD patients [4]. For these reasons one of the perspectives is the investigation of impulsivity of the BACHD rats. In particular, the DRL (Differential Reinforcement of Low rates of responding task*)* was used to assess impulsive action (IA). This particular task is broadly utilized to evaluate impulsive action, defined as the withhold of an instrumental response for a specific period of time before that response is reinforced [5]. The DRL schedule is an operant schedule that requires the subject to pause for a specified minimum period between responses to obtain a reward. Therefore, efficient DRL performance depends on various components of cognition such as vigilance, self control, short-term memory, waiting ability, and time estimation.

23 male Sprague-Dawley rats (5-months old) were used in this experiment. The w.t. males were supplied from Harlan Laboratories (Italy). The transgenic males (BACHD) were supplied from the Universitätsklinikum Tübingen (Tuebingen, Germany). A 12-h light/dark cycle was maintained during the experiment (light on at 8:00 A.M.). On arrival in the laboratory, rats were group-housed 2 to 4 per cage with wood shavings, were handled on a daily basis and were given access to food and water *ad libitum* until one week before starting the experiment.

One week before starting the experiment the daily food amount was progressively reduced and rats were fed a daily ration to maintain them at 85% of their normal free-feeding weight. Four operant Skinner boxes (31 × 25 × 31 cm) in sound proof ventilated chambers (background noise 65 dB) were controlled with a Graphic State software (Coulborn Instruments, Harvard Apparatus, USA) which controlled programmed task events and data collection. Each chamber was equipped with two left and right transparent walls with transparent back panel and

front door. A pellet dispenser for delivery of 45 mg grain-based precision pellets, one 4-cm response lever and a red houselight (lit at the beginning of each session) were located on the right panel.

Training phase began with a 30 minutes-session of magazine training, during which a single food pellet was delivered into the food trough every 60 seconds.

On the following day, rats were trained to press a fixed lever in order to receive a single pellet. After achieving a criterion of 50 reinforced lever presses in a 30 min session, rats began the DRL-5.

Rats were initially tested on a DRL-5 s schedule for five sessions, during which a lever press only resulted in a single food pellet delivery if at least 5 seconds had elapsed since the previous press. If the rat performed a premature lever press, the 5 seconds time period was reset; thus, rats were only reinforced if they withheld a response for more than 5 seconds. Each DRL session ended either after 60 minutes or 200 reinforcements were earned, whichever came first. Moreover, the red houselight was lit throughout the session.

Rats were then tested for ten sessions on a DRL-10 s schedule, during which the response had to be withheld for 10 seconds to obtain reinforcement. Also in this case, each session terminated either after 60 minutes or 200 reinforcements were earned.

For each DRL-5 session the frequency of lever pressing during each 1-s time bin (inter-response time; IRT) was recorded. The pattern of responding during DRL-5 was categorized into ‘‘burst,’’ ‘‘premature,’’ and ‘‘timing error responses’’ to include lever presses occurring during 0-1s IRTs, 1-4s IRTs and 4-5s IRTs, respectively.

For each DRL-10 session the frequency of lever pressing during each 2-s time bin (IRT) was recorded. In this case ‘‘burst,’’ ‘‘premature,’’ and ‘‘timing error responses’’ took into account lever presses occurring during 0-2s IRTs, 2-8s IRTs and 8-10s IRTs, respectively.

Thus, in order to assess performance throughout the sessions of DRL 5 and 10s, measures of instrumental responding, efficiency, genotype, delay and session effect were analyzed.

Analysis of response efficiency in each of the five DRL 5s sessions showed no relevant differences between BACHD and control rats . Two-way ANOVA revealed no genotype effect ($F<1$), a significant main effect of session ($F(4,84)=7.13$, $p<.001$) and a significant interaction between genotype and sessions ($F(4,84)= 4.46$, $p<.005$). On the other hand, response efficiency throughout the ten DRL 10s sessions showed no genotype effect ($F(1,21)=1.35$, ns) but significant effect of session ($F(9,189)=26.82$, $p<.001$). Moreover, no interaction between genotype and session was observed ($F(9,189)=1.49$, ns). In order to assess how the temporal characteristics of lever press responding changed between the two different genotypes, we analyzed interresponse times (IRTs) in three different intervals. There were no significant differences between groups in burst responses (0–1 s IRTs), premature responses (1-4 s IRTs) and timing errors (4-5 s IRTs) of the DRL 5s schedule. Interestingly, burst responses (0-2s IRTs) differences were only evident in the DRL 10s, during which BACHD rats were not able to withold lever pressing. Consequently, the number of responses of transgenic rats was significantly greater than the controls. Two-way ANOVA revealed a significant main effect of genotype ($F(1,21)=4.53$, $p<.05$) and session ($F(9,189)=9.44$, $p<.001$); however, no interaction between genotype and session ($F(9,189)=1.43$, ns) was found. Moreover, there were no differences between genotypes in the premature responses (2-8 s IRTs; FIG. 1F) and timing error (8-10 s IRTs; FIG. 1H) of the DRL 10s schedule.

Thus, the main difference between genotypes is evident during the burst (0-2 s IRT) in the DRL 10s schedule, revealing the incapacity of BACHD rats to withold an instrumental response for longer periods of time. Therefore, BACHD rats displayed deficits in behavioral inhibition (i.e. increased responding during non-reinforced periods) that were mainly evident in the DRL 10s schedule.

Taken together, these data provide evidence of BACHD rats deficits in the ability to inhibit lever press responding at increasing IRTs , predicting greater impulsive action of this transgenic rat model of Huntington’s disease.

References

1. Vonsattel JP, DiFiglia M (1998). Huntington disease. *Journal of Neuropathology & Experimental Neurology* **57**:369 –384.

2. Yu-Taeger L, Petrasch-Parwez E, Osmand AP, Redensek A, Metzger S, Clemens LE, Park L, Howland D, Calaminus C, Gu X, Pichler B, Yang XW, Riess O, Nguyen HP (2012). A novel BACHD transgenic rat exhibits characteristic neuropathological features of Huntington disease. *The Journal of Neuroscience* **32(44)**:15426-38.

3. Evenden JL (1999). Varieties of impulsivity. *Psychopharmacology (Berl)* **146**: 348–361.

4. Novak MJ, Tabrizi SJ (2010). Huntington's disease. *BMJ* **340**:c3109.

5. Kramer TJ, Rilling M (1970). Differential reinforcement of low rates: A selective critique. *Psychological Bulletin* **74(4)**: 225-254.