Behavioural Brain Research 279 (2015) 52–61

Contents lists available at ScienceDirect

Behavioural Brain Research

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

Research report

The rotenone-induced rat model of Parkinson's disease: Behavioral and electrophysiological findings

Christof von Wrangel, Kerstin Schwabe, Nadine John, Joachim K. Krauss, Mesbah Alam*

Department of Neurosurgery, Hannover Medical School, Hannover, Germany

HIGHLIGHTS

- Higher subthalamic nucleus firing rate and altered discharge pattern.
- Higher beta oscillatory activity in the STN and the motor cortex region.
- Chronic exposure to rotenone (i.p.) provides a suitable model of Parkinson's disease.
- Rotenone treated rats showed motor impairment and depletion of dopaminergic neurons.

ARTICLE INFO

Article history:
Received 2 July 2014
Received in revised form 29 October 2014
Accepted 4 November 2014
Available online 12 November 2014

Keywords:
Local field potentials
Mitochondrial complex I
Subthalamic nucleus
Single unit activity
Rotenone
Parkinson's disease

ABSTRACT

Exposure to rotenone leads to parkinsonian features, such as loss of dopaminergic neurons in the substantia nigra and motor impairment, however, the validity of this model has recently been questioned. In rodent and monkey models of Parkinson's disease (PD) abnormal neuronal activity in the basal ganglia motor loop has been described, with hyperactivity of the subthalamic nucleus (STN) similar to that found in PD. The present study aims at providing new and more specific evidence for the validity of the rotenone rat model of PD by examining whether neuronal activity in the STN is altered.

Male Sprague Dawley rats were treated with rotenone injections (2.5 mg/kg bodyweight intraperitoneally) for 60 days. Behavioral analysis showed an impairment in the rotarod and hanging wire test in the rotenone group ($p < 0.05$), accompanied by a decline in tyrosine hydroxylase immunoreactive neurons in the nigro-striatal region ($p < 0.001$). Thereafter, single unit (SU) activities and local field potentials were recorded in the STN in urethane anesthetized rats. The SU analysis revealed a higher neuronal discharge rate ($p < 0.001$), more bursts per minute ($p = 0.006$) and a higher oscillatory activity ($p = 0.008$) in the STN of rotenone treated rats. Spectral analysis showed an increase of relative beta power in the STN as well as in the motor cortex.

We found electrophysiological key features of PD pathology and pathophysiology in the STN of rotenone treated rats. Therefore, the rotenone-induced rat model of PD deserves further attention since it covers more aspects than dopamine depletion and implies the reproducibility of PD specific features.

© 2014 Elsevier B.V. All rights reserved.

1. Introduction

While the exact etiology of Parkinson's disease (PD) remains unknown and is likely multifactorial, both clinical and experimental evidence support the concept that deficiency of complex I of the mitochondrial respiratory chain plays a central role in its pathogenesis [1]. Some inherited forms of PD are caused by mutations of DJ-1, PINK 1 and other genes which impair the function of mitochondrial proteins, indicating that a common factor for mitochondrial impairment is an important feature which is also tightly linked with oxidative stress and protein misfolding [2,3].

Rotenone is a potent inhibitor of complex I (NADH: ubiquinone oxidoreductase) of the mitochondrial electron transport chain. There is strong epidemiological evidence that chronic rotenone exposure is associated with PD in humans [4]. In rats, rotenone causes a syndrome that replicates both neuropathological findings and behavioral symptoms of PD. Application of low doses of rotenone in vitro and in vivo have been shown to affect many of the mechanisms involved in the pathogenesis of PD, such as altered

* Corresponding author at: Department of Neurosurgery, Medical School of Hannover, Carl-Neuberg-Str. 1, 30625 Hannover, Germany. Tel.: +49 511 532 3869.
E-mail address: alam.mesbah@mh-hannover.de (M. Alam).

http://dx.doi.org/10.1016/j.bbr.2014.11.002
0166-4328/© 2014 Elsevier B.V. All rights reserved.

calcium signaling, induction of oxidative stress and apoptosis, loss of tyrosine hydroxlase, proteasomal dysfunction, nigral iron accumulation and the formation of fibrillar cytoplasmic inclusions that contain ubiquitin and α-synuclein [5–8]. In addition, studies have shown that exposure to rotenone induced behavioral and motor deficits, which are similar to human PD, including muscular rigidity (catalepsy), bradykinesia, postural instability, unsteady gait and sleep disturbances [9–11].

In PD the loss of dopamine (DA) -producing neurons in the substantia nigra pars compacta (SNpc) and subsequent depletion of striatal DA leads to increased spontaneous firing activity, periodic bursting and synchronized oscillatory activity in the beta (12–30 Hz) range in the subthalamic nucleus (STN) [12,13]. Similar alterations have also been shown in animal models of PD, such as the 6-hydroxydopamine (6-OHDA) rat model and the 1-methyl-4-phenyl-1,2,3,4-tetrahydropyridine (MPTP) monkey model [14–16]. Neuronal activity in the STN is thus regarded critical to assess the construct validity of PD animal models. In the rotenone model, electrophysiological measures have not been obtained, so far.

The rotenone rat model of PD would have certain advantages for experimental studies as compared to other models [17–19]. While most of the toxin models result in acute damage of DA neurons, the rotenone model mimics more closely the chronic progression of PD as observed in patients. Additionally, it provides proof of concept that systemic mitochondrial inhibition and impairment can lead to selective vulnerability for nigrostriatal degeneration. Thus, it may allow to examine various molecular and biochemical processes that converge in the final pathology and clinical manifestations of PD. This study investigates whether the systemic chronic injection of rotenone in rats leads to changes in neuronal firing rates, burstiness and oscillatory activity in the STN, in addition to motor impairment.

2. Material and methods

2.1. Animals

Eighteen adult male Sprague Dawley rats (Charles River Laboratories, Germany) were used in this study. They were housed in groups of three to four animals per cage (Macrolon type IV) and kept under controlled environmental conditions (temperature 22 °C, relative humidity 45–55%, 12 h light/dark cycle). All animals were fed with standard rat chow and water ad libitum. All animal procedures were approved in accordance with the European Council directive (2010/63/EU) and approved by the local animal ethic committee (Bezirksregierung LAVES Hannover, Germany).

2.2. Treatment

Rotenone (Sigma, Taufkirchen, DE) was dissolved in DMSO (Sigma, Taufkirchen, DE) and diluted with sterile natural oil (middle chain triglycerides MCT; Miglyol 812) to a final emulsion containing 2.5 mg rotenone/ml and 20 μl DMSO/ml. Ten rats were randomly assigned to the treatment group and received chronic intra-peritoneal (i.p.) injections with rotenone (2.5 mg/kg bodyweight once daily) for 60 days. Controls were treated with vehicle (20 μl DMSO/ml MCT; $n = 5$) or saline only (equivalent volume; $n = 3$). The weight and wellbeing of all rats were monitored at least once daily.

Motor impairment tests were performed one day before the 60 day injection period (Pre-Injection), one day after the 60 day injection period (Post-Injection Day 0), and after 14 days without any further treatment (Post-Injection Day 14). All tests were performed after a 30–60 min habituation period to the testing room during the day light cycle under artificial light with a fixed intensity and acoustic exposure to a masking noise (playing radio) to reduce adverse effects of impulsive noise.

2.3. Motor impairment

2.3.1. Rotarod testing

To assess the motor coordination of the animals, we used an accelerating Rotarod (IITC Life Science, Woodland Hills, CA). The Rotarod consisted of a suspended rod, accelerating for 60 s from 5 rounds per minute (RPM) to 15 RPM and continuing at that speed for a further 60 s. A trial was stopped when the rat fell off the Rotarod or after the complete 120 s. For each day, the mean of three trials was taken. Prior to the chronic injections, the rats were trained for five days to perform the test and a baseline was established.

2.3.2. Open field

To assess spontaneous locomotion, the animals were placed in a black plastic open field (60 cm × 60 cm × 30 cm). A video of the animals was recorded for 10 min by a camera installed above the box and the total distance traveled was automatically calculated by a video tracking system using the same settings for all rats (TopScan 1.0, Clever Sys. Inc., Reston, VA, USA).

2.3.3. Stepping test

The rats were tested for postural instability as described previously by Cannon et al. [5]. The animal was hold vertically with one forelimb touching the surface of the table, which was covered with medium-grit sandpaper. The rat was moved forward without allowing the second forelimb to touch the table, until the rat performed a "catch-up" step. The displacement distance until this step was recorded. Each trial was repeated three times for each forelimb and the mean was taken.

2.3.4. Hanging-wire

General muscle strength was assessed by using the hanging-wire test, a method previously described by Perez and Palmiter [20]. The rats were placed on the center of a vertical grid. After the rat grasped the grid with all paws, it was turned horizontally so that the animal hung upside down and the time was stopped until the rat released its grip. For each day, the test was performed three times with an intertrial interval of 15 min and the mean was taken.

2.4. Electrophysiological recordings

Electrophysiological recordings were taken in the STN of control and rotenone treated animals after behavioral testing, i.e., at least two weeks after termination of rotenone injection. The rats were anesthetized with urethane (1.3 g/kg, i.p.; ethyl carbamate, Sigma, St. Louis, MO) and placed in a stereotaxic frame. The body temperature was maintained at 37 ± 0.5° C by a heating device (FHC, Bowdoinham, ME). Surface ECG was recorded to monitor and ensure constant physiological conditions during recording. The electrocorticogram (ECG) electrodes were placed in the axilla and pelvic region (Lead II, CED1902 isolated amplifier, Cambridge Electronic Design, Cambridge, UK).

Small craniotomies were made over the target coordinate for the STN in both hemispheres. A single microelectrode for extracellular recordings (quartz coated pulled with a ground platinum-tungsten alloy core (95–5%), diameter 80 μm, impedance 1–2 MΩ) was connected to the Mini Matrix 2 channel version drives headstage (Thomas Recording, Germany) and stereotaxically guided through the skull burr holes to the target coordinates in the STN (A: −3.8 mm; L: ±2.5 mm; V: −7.6 mm) under continuous recording of extracellular neuronal signals using a microdrive (Thomas Recording GmbH, Giessen, Germany). Signals were amplified ×1000. The electrode signal was passed through a headstage with unit gain and then split to separately extract the single unit (SU) and local field potential (LFP) components (Fig. 1A). For the LFPs, the signal was

Download English Version:

https://daneshyari.com/en/article/6257319

Download Persian Version:

https://daneshyari.com/article/6257319

Daneshyari.com