1 | INTRODUCTION

Parkinson’s disease (PD) is characterized by degeneration of the nigrostriatal pathway, resulting in motor impairments such as bradykinesia as well as tremor. Treatments for motor symptoms of PD include dopamine (DA) replacement therapies, such as administration of the DA precursor L-3,4-dihydroxyphenylalanine (L-DOPA) and/or DA agonists. As the disease progresses, however, DA replacement...
therapies become less effective (Cotzias, Papavasiliou, & Gellene, 1969; Melamed, 1979; Muenter, Sharpless, Tyce, & Darley, 1977). Given this, novel alternatives and/or adjuncts to DA replacement that can improve therapeutic outcomes are still needed.

Inhibitory neurotransmission in the basal ganglia (BG) is primarily mediated by ionotropic GABA A receptors, which can be distinguished pharmacologically according to α-subunit expression. Within the BG, the α2-subunit is exclusively expressed on spiny projection neurons in the striatum. The α1-subunit is expressed postsynaptically within the external globus pallidus (GPe), subthalamic nucleus (STN), internal globus pallidus (GPi), and substantia nigra pars reticulata (SNr; Arbilla, Allen, Wick, & Langer, 1986; Waldvogel, Kubota, Fritschy, Mohler, & Faull, 1999; Boyes & Bolam, 2007; Figure 1).

Zolpidem (trade name Ambien®) is an imidazopyridine that binds to GABA A receptors at the benzodiazepine site, acting as a positive allosteric modulator with selective affinity for receptors expressing the α1-subunit. In vitro studies have shown that application of zolpidem results in an increase in decay time constant of Cl− channel opening and potentiation of iPSCs in zolpidem-sensitive nuclei within the BG (Chen, Savio Chan, & Yung, 2004; Chen, Xie, Fung, & Yung, 2007; Zhang, Chen, Xu, & Yung, 2008). Similarly, zolpidem potentiates GABA-induced decreases in the firing rate of zolpidem-sensitive cells (Duncan et al., 1995; Mereu, Carcangiu, Concas, Passino, & Biggio, 1990). Furthermore, zolpidem-sensitive GABA A receptors have been hypothesized to mediate the ataxic and myorelaxant properties of benzodiazepines, due to high expression levels of the α1-subunit within extrastriatal BG nuclei (Milic et al., 2012).

Interestingly, the BG nuclei expressing the α1-subunit have also been shown to exhibit oscillatory entrainment to cortical activity in a DA-depleted state (Avila et al., 2010; Deffains et al., 2016; Magill, Bolam, & Bevan, 2001; Mallet et al., 2008). This oscillatory entrainment, and associated alterations in firing pattern, have been observed in PD models (Sanderson, Movoungou, & Albe-Fessard, 1986; MacLeod, Ryman, & Arbuthnott, 1990; Soares et al., 2004; Wichmann & Soares, 2006; Walters, Hu, Itoga, Parr-Bwornie, & Bergstrom, 2007; Zold, Ballion, Riquelme, Gonon, & Murer, 2007; Lobb & Jaeger, 2015; reviewed in Lobb, 2014), and are associated with severity of motor symptoms in PD patients (Sharrott et al., 2014). Combined with the observed efficacy of the GABA analog progabide in the treatment of PD motor symptoms, available evidence indicates that a compound with selectivity for extrastriatal GABA A receptors—such as zolpidem—could yield promising results (Ziegler, Fournier, Bathien, Morselli, & Rondot, 1987). Investigations of this concept utilizing translational models, however, have not been reported.

We examined whether zolpidem-sensitive GABA A receptors constitute a potential therapeutic target in the treatment of PD motor symptoms using a translational model. The presented experiments tested whether zolpidem ameliorates motor deficits in unilaterally 6-hydroxydopamine (6-OHDA)-lesioned rats, utilizing assays sensitive to DA depletion (Iancu, Mohapel, Brundin, & Paul, 2005). First, we performed a dose-response investigation in intact rats to determine the threshold dose at which zolpidem impairs motor coordination using rotarod. Second, we investigated whether an acute subthreshold dose of zolpidem, as determined in the dose-response experiment, improved motor performance and forelimb use symmetry in unilaterally 6-OHDA-lesioned rats using both rotarod and the paw preference/cylinder test.

2 | METHODS

2.1 | Subjects

Adult male Sprague-Dawley rats weighing 250–350 g (Charles River) were housed individually under conditions of constant temperature (21°C) and humidity (40%)...
and maintained on a 12/12 light/dark cycle (0700 on, 1900 off). Food and water were available ad libitum. A total of 54 animals were used in the present experiments. Animal procedures were conducted in accordance with the Guide for the Care and Use of Laboratory Animals (NIH Guide, rev. 1996). All protocols were approved by the Rutgers University Institutional Animal Care and Use Committee.

2.2 | Unilateral 6-OHDA treatment

DA lesion was induced by intracerebral infusion of 6-OHDA. Animals received desipramine (25 mg/kg; i.p.) 20 min prior to surgery to prevent destruction of noradrenergic nerve terminals (Waddington, 1980), and were anesthetized with sodium pentobarbital (40 mg/kg; i.p.). The animal was placed into the stereotaxic frame (Kopf), and the scalp was infiltrated with bupivacaine HCl (0.5% w/v; Hospira Inc.) to provide local anesthesia. Body temperature was maintained with a heating pad (Gaymar Industries) throughout the procedure. 6-OHDA (3 μg/μl, freebase wt.) in vehicle (0.2 mg/ml ascorbic acid in ACSF) was infused into the right medial forebrain bundle at a rate of 2 μl/4 min via a 30 g cannula at the following coordinates (in mm): AP: −3.2 (bregma), ML: −1.5, and DV: -7.2 from dura (Paxinos & Watson, 1982). Postoperative analgesia was provided using meloxicam (2 mg/kg, s.c.; Henry Schein).

Animals recovered for 14 days and were assessed for apomorphine-induced rotational behavior (0.05 mg/kg, s.c.). Behavioral testing commenced 4–10 days post-apomorphine testing.

2.3 | Rotarod balance beam testing

Animals were tested for motor coordination using a rotarod apparatus (Economex, Columbia Instruments), following procedures outlined by Carter, Morton, and Dunnett (2001). Briefly, animals habituated to the testing room for ≥ 60 min. Training consisted of 4 trials at a fixed speed of 10 rpm, repeated over 3 consecutive days. Latency to fall was recorded and a 60 s trial cutoff was used. The inter-trial interval was 5 min. 5 days after the initiation of training, animals were tested at a fixed speed of 20 rpm, which served as the pre-lesion/undrugged measure. Subjects were given 2 trials at 20 rpm, and fall latencies for both trials were averaged together for statistical analysis.

2.3.1 | Rotarod performance in intact rats

To empirically determine non-sedative doses of zolpidem for further examination of effects on motor deficits in DA-lesioned animals, intact rats (n = 14) received zolpidem (0.5, 1, 2.5, 5, 10 mg/kg; i.p.) or vehicle 15 min. before being placed onto the rotarod apparatus. This delay was chosen to ensure testing occurred coincident with the reported t_max for zolpidem in the rat (Durand, Thénot, Bianchetti, & Morselli, 1992). To allow for drug washout, ≥48 hr separated testing sessions. Differences between doses/conditions were investigated using the Wilcoxon Rank Sum test. Error correction was not applied, as the purpose of this analysis was to find the lowest possible dose that may cause significant motor impairment. These data served to provide normative values for subsequent study of the DA-depleted state.

2.3.2 | Rotarod performance in unilaterally DA-depleted rats

6-OHDA lesion procedures were conducted in some animals (n = 26) following rotarod training/testing (see above). Approximately 1 month post-lesion, animals were tested for rotarod performance. Approximately 1 week later, animals were tested 15 min following i.p. injection of zolpidem (0.1, 0.25, 0.5 mg/kg) or vehicle. All animals received each dose following the Latin Square method, with ≥ 48 hr between testing sessions. In order to ensure that animals showed a significant impairment, the criteria for inclusion in statistical analysis for this experiment were as follows: (a) post-lesion rotarod fall latency ≤ 45 s, (b) verification of DA depletion ≥80% compared to the intact hemisphere. Statistical analysis was performed using Friedman’s nonparametric ANOVA for Repeated Measures. A significant main effect was deconstructed using the Wilcoxon Sign Rank test using the Bonferroni correction method for family-wise error rate for the following comparisons: intact vs. lesion, lesion vs. vehicle/0.1/0.25/0.5 Zol, vehicle vs. 0.1/0.25/0.5 Zol (8 comparisons, α_adj = 0.00625).

2.4 | Paw preference behavior in a novel cylinder

Forelimb use symmetry was assessed using a modified version of the cylinder test (Schallert & Tillerson, 2000). Briefly, animals (n = 13) were placed in a plastic cylinder (diameter = 30 cm) lined with fresh bedding (BETA chips) in a dimly lit room for 25 min. No habituation to the cylinder was permitted. The experimenter left the room and exploratory activity was digitally recorded from above. For each session, 15 min of video were analyzed beginning at the onset of first paw contact with the side of the cylinder. Number of weight-bearing contacts made by each forepaw was recorded, and the ratio of contralateral to total forepaw contacts was calculated as follows:

Contralateral/(ipsilateral + contralateral).

Animals were tested pre-lesion (32 days pre-surgery), post-lesion (undrugged; 20 days post-surgery), and 15 min following i.p. injection of zolpidem (0.1 mg/kg) or vehicle (51–58 days post-surgery). Animals that did not execute ≥10
total forepaw contacts in each experimental condition were excluded from analysis. Similarly, animals that were shown to have <80% striatal DA depletion in the lesioned hemisphere were excluded from analysis. Differences between treatment conditions were explored using One-Way Repeated Measures ANOVA followed by multiple comparison post hoc investigation using the Bonferroni correction for all possible interactions (6 comparisons, $\alpha_{adj} = 0.0083$).

### 2.5 Analysis of tissue DA content

Analysis of striatal DA content was performed in 6-OHDA-treated animals. Rats were given chloral hydrate (400 mg/kg; i.p.), rapidly decapitated, and the striata were extracted bilaterally then frozen on dry ice. The dissected striata were stored at −80°C until homogenization and centrifugation. Samples (25 μl) of resulting supernatant were assayed for DA content by high pressure liquid chromatography with electrochemical detection (HPLC-ED; Zackheim & Abercrombie, 2005). Striatal DA content was quantified in ng/g tissue for each hemisphere, and percent DA loss was calculated as the percent decrease in the lesioned versus intact hemisphere. Since 80% striatal DA depletion is required for manifestation of robust behavioral deficits (Abercrombie, Bonat, & Zigmond, 1990; Hornykiewicz, 1993; Kirik, Rosenblad, & Björklund, 1998), this served as the cutoff for inclusion in statistical analysis of experiments.

### 2.6 Drugs & materials

Chloral hydrate (trichloroacetaldehyde hydrate), desipramine hydrochloride, 6-hydroxydopamine (2,4,5-trihydroxy-phenet hylamine HBr), apomorphine, and zolpidem were purchased from Sigma Chemicals (St. Louis, MO). Sodium pentobarbital (Pentasol®) was purchased from Virbac Animal Health (Ft. Worth, TX). All other reagents were of the highest purity commercially available (Fisher Scientific; Springfield, NJ).

Chloral hydrate, sodium pentobarbital, desipramine, and apomorphine were each dissolved in sterile 0.9% NaCl and made fresh before use. Chloral hydrate, sodium pentobarbital, and desipramine were mixed according to pre-defined concentrations (80 mg/ml, 50 mg/ml, and 12.5 mg/ml, respectively), and doses were administered according to weight (mg/kg).

Zolpidem was dissolved in 0.1 M glacial acetic acid, and brought up to volume with ddH2O according to the doses being administered (final concentration = 1% acetic acid). Drug concentrations for each of the reported doses were normalized so that the volume of solution injected was proportional to weight of the animal (1 ml/kg). Vehicle contained 1% acetic acid dissolved in ddH2O (v/v), and was injected at a volume proportional to weight of the animal (1 ml/kg). Both vehicle and zolpidem solutions were pH-adjusted using 10 N NaOH (final pH = 5.0).

### 2.7 Statistics

Statistical analysis was performed using MATLAB (2014b, Mathworks), with the exception of One-Way Repeated Measures ANOVA (GraphPad Prism). Differences were considered statistically significant if $p < 0.05$, unless otherwise stated.

### 3 RESULTS

#### 3.1 Zolpidem induces a dose-dependent impairment on rotarod in intact rats

In order to empirically ascertain a dose of zolpidem that would not interfere with motor coordination, a dose-response experiment was conducted assessing rotarod performance in intact rats ($n = 14$). Doses of 0.5, 1.0, 2.5, 5.0, and 10.0 mg/kg zolpidem (i.p.) were tested, as well as vehicle. Mean ($\pm$SEM) latency to fall (in s) and number of animals given each dose were as follows (Figure 2a): 59.21 ± 0.96 (undrugged, $n = 14$), 57.57 ± 2.09 (vehicle, $n = 14$), 57.8 ± 1.72 (0.5 mg/kg, $n = 10$), 45.86 ± 4.68 (1 mg/kg, $n = 14$), 36.33 ± 9.22 (2.5 mg/kg, $n = 6$), 7.75 ± 4.17 (5 mg/kg, $n = 4$), 3.69 ± 0.96 (10 mg/kg, $n = 8$).

Dose interactions pertinent to subsequent experiments were as follows (Figure 2b): Doses $\geq$ 1.0 mg/kg produced a statistically significant impairment compared to the undrugged condition (1.0 mg/kg: $p = 0.0045$; 2.5 mg/kg: $p = 0.0012$; 5.0 mg/kg: $p = 5.05 \times 10^{-5}$; 10.0 mg/kg: $p = 3.48 \times 10^{-5}$). A dose of 0.5 mg/kg did not produce a significant impairment compared to the undrugged ($p = 0.3534$), nor vehicle conditions ($p = 0.9709$). Doses $\geq$ 1.0 mg/kg produced a significant impairment compared to vehicle (1.0 mg/kg: $p = 0.0298$; 2.5 mg/kg: $p = 0.0067$; 5 mg/kg: $p = 0.0032$, 10 mg/kg: $p = 6.88 \times 10^{-5}$). As a result, the maximal dose utilized in subsequent experiments was 0.5 mg/kg.

#### 3.2 Behavioral and neurochemical effects of unilateral 6-OHDA lesion

The effect of unilateral 6-OHDA lesion was quantified on two measures of motor behavior. We compared performance in the intact to the 6-OHDA condition in both rotarod and cylinder tests (see Methods). A significant effect of lesion was found in both. 6-OHDA lesion significantly impaired animals ($n = 10$) that met inclusion criteria (see Methods) in the rotarod experiment ($p_{adj} = 0.0156$; Figure 3b), and induced a significant forelimb use bias in the cylinder test ($n = 10$, $p_{adj} = 8.78 \times 10^{-7}$; Figure 4b).
The extent of 6-OHDA-induced lesion was assessed via HPLC-ED (see Methods). In animals that met the inclusion criteria for the rotarod experiment (see Methods), mean (±SEM) tissue DA depletion was 96.72 ± 1.84%. In animals tested in the cylinder/paw preference experiment, mean (±SEM) DA depletion was 96.99 ± 0.93% (Table 1).

3.3 | Zolpidem improves rotarod performance in unilaterally 6-OHDA-lesioned rats

The efficacy of zolpidem in the treatment of 6-OHDA-induced motor impairment was assessed using rotarod. Of the animals tested (n = 26), only 10 met the inclusion criteria (see Methods). The predominant reason for exclusion from statistical analysis was a lack of adequate impairment on rotarod performance under the present protocol (20 rpm, 45 s cutoff) despite >80% DA depletion in the lesioned hemisphere (n = 13, mean latency to fall = 56.62 s, mean DA depletion = 98.15%). In rare cases, animals that showed sufficient impairment in rotarod did not meet the DA depletion criteria (n = 3, mean latency to fall = 28.67 s, mean DA depletion = 14.37%).

Animals that met inclusion criteria (n = 10) were tested 15 min following i.p. injection of zolpidem (0.1, 0.25, 0.5 mg/kg) or vehicle. Mean (±SEM) latency to fall (in s) for each condition was as follows (Figure 3): 59.85 ± 0.15 (intact), 23.55 ± 4.21 (6-OHDA), 25.25 ± 6.52 (6-OHDA + Veh), 41.55 ± 3.55 (6-OHDA + 0.1 Zol), 25.10 ± 6.57 (6-OHDA + 0.25 Zol), 17.20 ± 3.37 (6-OHDA + 0.5 Zol). Analysis using Friedman’s nonparametric ANOVA for Repeated Measures yielded a significant main effect (χ² = 22.37, p = 3.08 × 10⁻⁵). Post hoc investigations of effects between conditions revealed that zolpidem improved rotarod performance at the lowest dose tested (0.1 mg/kg; Figure 3a). Injection of 0.1 mg/kg zolpidem significantly improved rotarod performance compared to post-lesion (p adj = 0.0312, Figure 3b). Of note, 0.5 mg/kg zolpidem did not significantly alter rotarod performance, but in most cases (8/10) resulted in shorter fall latency than the post-lesion condition (p adj = 0.1250, Figure 3e). All other tested interactions were non-significant. These data suggest that 0.1 mg/kg zolpidem improved motor performance, and that a dose of 0.5 mg/kg potentiated impairments in unilaterally 6-OHDA-lesioned rats as measured by rotarod.

To directly compare each dose/condition to the intact/undrugged state, a “% intact performance” value was calculated by dividing the latency to fall (in s) of each animal (in each condition) by its individual performance in the intact/pre-lesion condition. Mean (±SEM) values obtained for each condition were as follows: 39.27 ± 7.01% (6-OHDA), 42.10 ± 10.86% (6-OHDA + Veh), 41.88 ± 10.93% (6-OHDA + 0.1 Zol), 28.69 ± 5.61% (6-OHDA + 0.25 Zol). Analysis using Friedman’s nonparametric ANOVA for Repeated Measures yielded a significant main effect (χ² = 12.2, p = 0.0159; Figure 3f). Post hoc investigations were conducted using Wilcoxon Rank tests using the Bonferroni correction method for the following comparisons: lesion vs. vehicle/0.1/0.25/0.5 Zol (4 comparisons, α adj = 0.0125). We found that 0.1 mg/kg zolpidem resulted in a significantly higher value when compared to the lesion condition (p adj = 0.0156). All other comparisons were non-significant (Figure 3f).

3.4 | Zolpidem improves forelimb use symmetry in unilaterally 6-OHDA-lesioned rats

Unilateral 6-OHDA lesion induces asymmetry in forelimb use, such that utilization of the forepaw contralateral to the lesion is significantly reduced compared to the ipsilateral forepaw. We tested whether zolpidem decreased forelimb use asymmetry in 6-OHDA-lesioned rats using the cylinder test.
Animals that met the inclusion criteria \((n = 10)\) were tested 15 min following i.p. injection of zolpidem or vehicle. Following analysis using Friedman’s nonparametric ANOVA for Repeated Measures and Wilcoxon Sign Rank tests, it was found that 0.1 mg/kg zolpidem significantly improved rotarod performance compared to the lesion condition. (a) Performance of each animal for each experimental condition. Data are expressed as mean ± SEM, dots represent each animal. (b–e) Comparisons of experimental conditions: (b) intact (pre-lesion) vs. 6-OHDA (post-lesion), (c) 6-OHDA vs. 6-OHDA + 0.1 Zol, (d) 6-OHDA vs. 6-OHDA + 0.25 Zol, (e) 6-OHDA vs. 6-OHDA + 0.5 Zol. (f) Rotarod performance of each animal (in each condition) was compared to its performance in the intact condition, and a % intact performance value was calculated. It was found that 0.1 Zol significantly improved this measure compared to the lesion condition. Data are expressed as mean ± SEM, dots represent each animal. Top lines indicate significant interactions. * denotes significance (\(p < 0.05\))
FIGURE 4  Zolpidem reduces forelimb use asymmetry in unilaterally 6-OHDA-lesioned rats in the cylinder/paw preference test. Animals (n = 10) were tested for volitional forelimb use pre-lesion (intact), post-lesion (6-OHDA), and 15 min following i.p. injection of vehicle (6-OHDA + Veh) or 0.1 mg/kg zolpidem (6-OHDA + 0.1 Zol). Analysis using a One-Way Repeated Measures ANOVA and multiple comparisons post hoc testing indicated that zolpidem significantly improved use of the forelimb contralateral to the lesion. (a) Box plot illustrating results of the cylinder/paw preference test for each experimental condition, dots represent each animal (b) Unilateral 6-OHDA lesion induced a significant forelimb use bias. (c) A dose of 0.1 mg/kg zolpidem significantly reduced forelimb use bias. (d) Box plot illustrating total forepaw contacts in each experimental condition. (e) Unilateral 6-OHDA lesion induced a significant reduction in total forepaw contacts. (f) There was no significant difference in total forepaw contacts between vehicle and 0.1 Zol conditions. Dots represent each animal. Top lines indicate significant interactions. * denotes p < 0.05, ** denotes p < 0.01, *** denotes p < 0.00001

TABLE 1  Striatal DA depletion as a result of unilateral 6-OHDA infusion. Tissue DA content by hemisphere, expressed in ng DA per g tissue weight, as assessed by HPLC-ED. Percent DA loss was calculated as the percent decrease in the lesioned versus intact hemisphere. Data are expressed as mean ± SEM

<table>
<thead>
<tr>
<th>Group/Test</th>
<th>n</th>
<th>Hemisphere</th>
<th>Intact (ng/g)</th>
<th>Lesion (ng/g)</th>
<th>Percent DA Loss (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rotarod</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Included</td>
<td>10</td>
<td>Intact</td>
<td>6576.22 ± 1192.65</td>
<td>74.45 ± 8.92</td>
<td>96.72 ± 1.84</td>
</tr>
<tr>
<td>Excluded</td>
<td>13</td>
<td>(low deficit)</td>
<td>9000.01 ± 1385.06</td>
<td>126.70 ± 36.93</td>
<td>98.17 ± 0.43</td>
</tr>
<tr>
<td>Excluded</td>
<td>3</td>
<td>(low lesion)</td>
<td>11055.65 ± 1339.75</td>
<td>9511.64 ± 1390.04</td>
<td>14.38 ± 5.38</td>
</tr>
<tr>
<td>Cylinder</td>
<td>13</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Included</td>
<td>10</td>
<td>Intact</td>
<td>6456.26 ± 1320.85</td>
<td>95.04 ± 14.76</td>
<td>96.99 ± 0.93</td>
</tr>
<tr>
<td>Excluded</td>
<td>3</td>
<td>Intact</td>
<td>7605.5 ± 5058.67</td>
<td>258.79 ± 137.53</td>
<td>95.65 ± 0.78</td>
</tr>
</tbody>
</table>
Similarly, total contacts in the intact condition differed significantly compared to zolpidem ($p_{adj} = 3.04 \times 10^{-4}$), as well as vehicle ($p_{adj} = 1.42 \times 10^{-4}$). Zolpidem did not significantly alter total forepaw contacts compared to post-lesion ($p_{adj} = 0.1734$), nor vehicle ($p_{adj} = 4.2847$; Figure 4f) conditions. There was also no significant difference between lesion and vehicle conditions ($p_{adj} = 0.2545$).

4 | DISCUSSION

The presented experiments investigated whether zolpidem-sensitive GABA_A receptors may serve as a potential novel target in the treatment of motor symptoms of PD. First, we explored the dose-response relationship between zolpidem and motor coordination using rotarod in order to determine the threshold dose at which zolpidem encumbered motor behavior in intact rats. Next, we tested the hypothesis that zolpidem may improve motor deficits in unilaterally 6-OHDA lesioned rats using rotarod and cylinder/paw preference tests. Our data indicate that zolpidem induced a dose-dependent impairment on rotarod performance in intact rats, and that a low dose of zolpidem (0.1 mg/kg) selectively improved rotarod performance as well as forelimb use symmetry. Notably, the overall number of forepaw contacts were similar in both zolpidem and vehicle conditions, indicating that the observed improvement was not due to increased “weight” of each individual contact in the zolpidem condition.

The data presented here indicate that zolpidem improved motor deficits in a translational model of PD. However, there are some potential caveats in the rotarod component of this study. First, there was no significant difference between vehicle and 0.1 mg/kg zolpidem conditions. This can potentially be explained by practice effects, as an analysis of the relationship between the number of trials prior to the vehicle condition and rotarod performance indicated a positive relationship ($r = 0.2805$; data not shown). A similar analysis of the 0.1 mg/kg zolpidem condition showed the opposite ($r = -0.5535$; data not shown), indicating that prior trials did not affect performance in this condition. Second, nearly half of the animals tested using rotarod did not meet the inclusion criteria such that they did not exhibit sufficient deficit in the post-lesion condition despite >80% DA depletion in the lesioned hemisphere. Interestingly, an analysis of apomorphine-induced rotational behavior using a Student’s t-test suggests that there was no difference between the included/excluded animals (120.10 ± 18.31 vs. 140.31 ± 23.51; $p = 0.7240$). This can likely be attributed to the stringent nature of the rotarod inclusion criteria, as well as the 60 s trial duration used in this study, which was intended to ensure that the included animals were significantly impaired following 6-OHDA treatment.

To date, the face validity of zolpidem-sensitive GABA_A receptors as a target in the treatment of PD has not been thoroughly investigated. In PD patients, single-patient and small-cohort case studies have shown that subhypnotic doses of zolpidem reduce Unified Parkinson’s Disease Rating Scale (UPDRS) scores and reduce dyskinetic side effects associated with DA replacement (Daniele, Albanese, Gainotti, Gregori, & Bartolomeo, 1997; Ruzicka, Roth, Jech, & Busek, 2000; Chen & Sy, 2008; reviewed in Daniele, Panza, Greco, Logroscino, & Seripa, 2016). More recently, Hall et al. (2014) reported that zolpidem improved UPDRS scores and modified cortical beta oscillations in early-stage PD patients. Combined with the results presented here, the existing literature supports the hypothesis that zolpidem may indeed be a valid pharmacological intervention for PD, particularly when unwanted side effects of DA replacement therapies become problematic.

In summary, we present the first translational evidence of the efficacy of extrastriatal GABA_A receptors in the treatment of motor impairments in a valid PD model. Our data suggest that a low (0.1 mg/kg) dose of zolpidem improves both motor coordination and volitional forelimb use in unilaterally 6-OHDA-lesioned rats, with doses ≥0.5 mg/kg inducing a potentiation of motor impairment. Future research should investigate the impact of zolpidem on the neurophysiological hallmarks of PD, as studies investigating the effect of DA replacement therapies on these hallmarks have shown mixed results. Similarly, due to the overlap between the neural correlates of PD motor symptoms and L-DOPA-induced dyskinesia, our data suggest that zolpidem should be investigated as an intervention in mitigating the side effects of DA replacement therapies.

ACKNOWLEDGMENTS

This work was funded by NS059921 (to EDA) and NS034865 (to James M. Tepper). We thank Fulva Shah, Nupur Jain, and Naiya Butayla for technical assistance, Dr. Samar K. Alsalehedar for her input, and Dr. James M. Tepper for insightful comments on the manuscript. RA is supported by the Behavioral and Neural Sciences (BNS) Ph.D. Program at Rutgers-Newark.

CONFLICT OF INTEREST

The authors declare no competing financial interests.

DATA ACCESSIBILITY

Raw data can be accessed upon request by contacting the corresponding author.
AUTHOR CONTRIBUTIONS

RA designed/performed experiments, devised statistical analysis, analyzed data, wrote the manuscript, and designed figures. EDA designed the experiments, assisted in designing statistical analysis and wrote the manuscript.

ORCID

Robert Assini http://orcid.org/0000-0002-4241-4653

REFERENCES


