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Evaluation of Dopamine D3 Receptor Antagonists PG01037, PG01042, and VK4-116 Against D1R-induced Dyskinesia

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Evaluation of D_3R antagonists PG01037, PG01042, and VK4-116 against D_1R-induced dyskinesia

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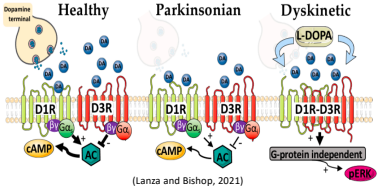
Introduction

- Parkinson's Disease (PD) leads to loss of dopamine (DA) cells in the substantia nigra resulting in motor symptoms. These can be relieved with DA replacement therapy, with L-DOPA but chronic treatment inevitably leads to L-DOPA-induced dyskinesia (LID) characterized by abnormal involuntary movements (AIMs). [1,2]
- The activity of the direct striatonigral pathway in LID is characterized by sensitization of D1R and upregulation of D3R. The D1R and D3R are thought to physically and functionally interact to form the D1R-D3R heteromer.[6]
- This interaction may increase DA affinity to D1R, thereby enhancing D1R signaling and thus LID while also switching DA receptor signaling from G-protein dependent to G-protein independent signaling.[6,8]
- Interestingly, evidence suggests that genetic knockdown of D3R on D1R bearing cells can reduce LID, suggesting D3R may be important in potentiating D1R activity.[7]
- The enhanced D1R signaling via D3R and alterations in intracellular signaling cascades seen in LID models make ideal targets for alleviating dyskinesias.
- The present study was designed to test several D_3R targeting compounds and evaluate their effects on D1R agonist-induced dyskinetic behavior in a 6-OHDA rodent model.

The current study used three D_3R antagonist drugs treatments, hypothesizing that PG01037 should not affect dyskinesia, while PG01042 should reduce dyskinetic behavior and VK4-116 will exacerbate dyskinesia.



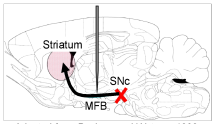
(Lanza and Bishop, 2021)

Methods

Subjects:
Eight adult Sprague Dawley rats (5 male and 3 female).

Surgeries:
All rats had a unilateral injection of 6-hydroxydopamine (6-OHDA) into the medial forebrain bundle (MFB) to deplete striatal DA and mimic human PD.

Unilateral Lesions with 6-OHDA



Adapted from Paxinos and Watson, 1998

Behavioral Testing:

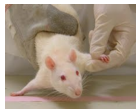
Abnormal Involuntary Movements (AIMs) [3,4]
After DA treatments, dyskinesia severity was quantified using the AIMs scale. The AIMs scale tests for intensity of LID by analyzing 3 behaviors: Axial, Limb and Orolingual (ALO). Behavior was observed for 60 s every 10 min. Each behavior was rated on a scale of 0-4. (0=not present, 1=present for <30 s, 2=present for > 30 s, 3=present for 60 s and interruptible, 4=present for 60 s and uninterruptable).

1. *Axial* – dystonic twisting of the trunk to the side contralateral to lesion
2. *Limb* – up and down, side-to-side movement of the right forelimb
3. *Orolingual* – asymmetric jaw tremors and tongue protrusions

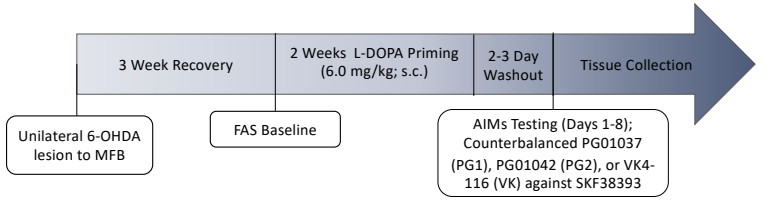
Rat LD Human LD

Drug induced rotations in the contralateral (+) and ipsilateral (-) direction were also counted to determine behavioral sensitivity to D1R agonist effects.

Forepaw Adjusting Steps Test (FAS) [4,5]
The FAS test evaluates motor impairment due to lesion by recording steps taken with both the lesioned paw and the intact paw. Data is represented as total percent intact (lesioned steps/intact steps) * 100. FAS was used to confirm a parkinsonian phenotype.



Experimental Design

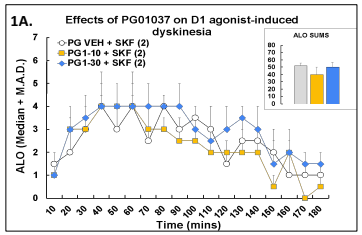


Treatments

SKF38393	2 mg/kg; s.c.
PG01037	10mg/kg; i.p.
PG01037	30mg/kg; i.p.
PG01042	5mg/kg; i.p.
PG01042	10mg/kg; i.p.
VK4-116	10mg/kg; i.p.
VK4-116	20mg/kg; i.p.

Table 1: In a within subjects, counterbalanced design, rats were split into treatment groups that received D3R antagonists: PG01037, PG01042 or VK4-116.

Results



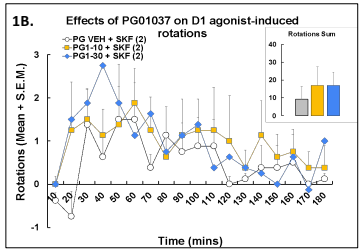
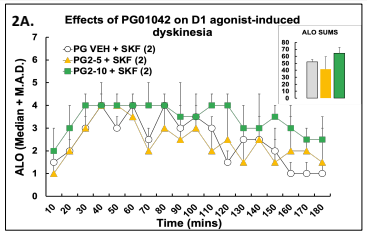


Figure 1: ALO and rotational behavior were no reduced by PG01037 (p>0.05). PG01037 Vehicle scores did not differ significantly.



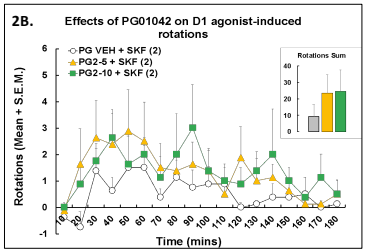
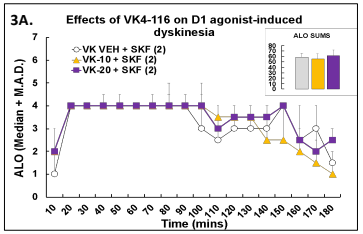


Figure 2: Rotational behavior was not reduced or exacerbated by PG01042, At higher doses PG01042 maintained a higher ALO score compared to the vehicle (p<0.05).



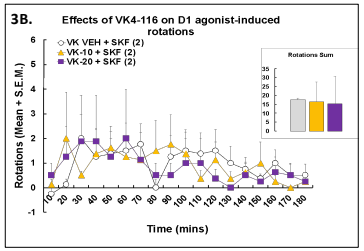


Figure 3: All VK4-116 groups did not demonstrate a significant difference in ALO and rotational behavior.

Conclusions

- PG01037, a mixed G-protein dependent/independent D3R antagonist (**Fig. 1**) and VK4-116, which is a G-protein dependent D3R antagonist (**Fig 3.**) did not alter D1R agonist-induced behaviors.
- Unexpectedly, higher doses of PG01042, a G-protein independent D3R antagonist increased D1R agonist-induced behaviors (**Fig. 2**).
- Although there was no reduction in D1R-mediated dyskinesia, manipulating D3R activity in the presence of endogenous DA as would be seen with L-DOPA, could better evaluate the functional selectivity between D3R antagonists.

Future research will evaluate the dyskinetic behaviors in rats treated with L-DOPA with D_3R antagonists to better understand synergistic effects with L-DOPA induced dyskinesia.

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