Gastrointestinal dysfunctions in experimental Parkinson's disease: alterations of excitatory and inhibitory neuromuscular pathways regulating colonic motility

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Parkinson's disease (PD) is characterized by degeneration of nigrostriatal dopaminergic neurons (Blandini et al., 2000) and the presence, in surviving neurons, of eosinophilic cytoplasmic inclusions containing aggregated α-synuclein (Lewy bodies). Patients with PD can develop gastrointestinal motor dysfunctions and alterations of their enteric nervous system (Labouvier et al., 2009); furthermore, Lewy bodies have also been found in enteric neurons (Braak et al., 2006). The aim of the present study was to examine the patterns of colonic neuromuscular excitatory (cholinergic and tachykininergic) and inhibitory (nitrergic) pathways in a rat model of PD.

PD-like nigrosriatal degeneration was induced in rats by injecting 6-hydroxydopamine into the medial forebrain bundle (Blandini et al., 2009). Animals were sacrificed at 28 or 56 days after surgery. Colonic longitudinal muscle preparations were set up in organ baths containing Krebs solution, and connected to isometric transducers to record contractile responses (g/g tissue) elicited by electrical stimulation (1-10 Hz), in the presence of guanethidine and N^{ω}-nitro-L-arginine methylester (L-NAME). L-732,138 (NK_1 receptor antagonist) or atropine were then used to record contractions driven primarily by acetylcholine or tachykinins, respectively. In addition, electrically evoked contractile responses (5 Hz) in the presence L-NAME were recorded and expressed as percentage of contractions obtained in the absence of L-NAME. Contractions elicited by exogenous substance P (SP, 0.01-10 μM) or carbachol (0.01-10 μM) in the presence of tetrodotoxin (1 μM) were also recorded. Sodium nitroprusside (SNP, 0.001-100 μM) was used to assess relaxations elicited by exogenous NO in colonic preparations pre-contracted with potassium chloride (KCl, 50 mM). The NO-dependent effect was confirmed by means of the guanylyl cyclase inhibitor (ODQ, 10 μM). Each value represents the mean ± S.E.M obtained from 6 experiments.

In control preparations incubated with L-732,138, electrical stimulation evoked atropine-sensitive cholinergic contractions (49.6±5.3 g/g tissue), which were reduced in rats with PD at 28 and 56 days (31.3±4.6 and 37.7±2.3 g/g tissue, respectively). On the other hand, carbachol-evoked contractions were enhanced in rats with PD both at 28 and 56 days (92.6±3.3 and 91.7±2.4 g/g tissue, respectively), as compared with controls (51.7±4.4 g/g tissue). In the presence of atropine, electrically evoked L-732,138-sensitive tachykininergic contractions were enhanced in PD rats at 56 days, as compared with controls (49.1±4.1 vs 28.7±3.8 g/g tissue). Likewise, the contractions elicited by exogenous SP were also enhanced in tissues from PD rats both at 28 (45.0±5.3 g/g tissue) and 56 days (77.2±4.2 g/g tissue), as compared with controls (27.0±1.9 g/g tissue). In control preparations maintained in standard Krebs solution, L-NAME enhanced electrically evoked contractile responses (+35.2±5.6%), while no significant effects were recorded in tissues form rats at day 28 and 56 after 6-OHDA injection (+19.7±8.7% and +7.6±5%, respectively). The NO-dependent relaxant responses elicited by SNP did not differ in controls and rats with PD at 28 and 56 days (-40.7±2.1% vs -45.9±2.1% and -37.1±2.0%, respectively).

The present results suggest that experimental PD is associated with changes in neurotransmitter pathways driving colonic excitatory and inhibitory motor functions: an impairment of nitrergic and cholinergic transmission occurs in concomitance with an enhancement of tachykininergic control. Such a shift takes place along with an up-regulation of the contractile responses mediated by muscarinic receptors on smooth muscle, which might be compensatory in nature.

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Lebouvier et al. (2009). *Eur J Neuroscie* 30, 735–41

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