Oxidative metabolites of 5-S-cysteinyldopamine inhibit the pyruvate dehydrogenase complex

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Summary. The principal neuropathological feature of Parkinson’s disease is the degeneration of melanized dopamine neurons in the substantia nigra pars compacta (SNc). Characteristic pathobiochemical changes in the parkinsonian SNc include a fall of both dopamine (DA) and glutathione levels (GSH), increased activity of γ-glutamyl transpeptidase, a key enzyme involved in the degradation of GSH to L-cysteine (CySH), together with evidence for elevated intraneuronal superoxide (O$_2^•$), nitric oxide (NO•) and hence peroxynitrite (ONOO$^-$) generation, and accelerated DA oxidation as indicated by a large rise of the 5-S-cysteinyldopamine (5-S-CyS-DA)/DA concentration ratio. The latter effect is consistent with an increased rate of DA oxidation by O$_2^•$ and ONOO$^-$ forming DA-o-quinone which reacts with CySH forming 5-S-CyS-DA. However, 5-S-CyS-DA is readily further oxidized to 7-(2-aminoethyl)-3,4-dihydro-5-hydroxy-2H-1,4-benzothiazine-3-carboxylic acid (DHBT-1). Previous studies have demonstrated that DHBT-1 is rapidly accumulated by isolated intact rat brain mitochondria and selectively inhibits complex I respiration and the α-ketoglutarate dehydrogenase (α-KGDH) complex. In this study it is demonstrated that DHBT-1 also inhibits the pyruvate dehydrogenase complex (PDHC). The mechanism underlying the inhibition of all of these enzyme complexes involves bioactivation of intramitochondrial DHBT-1 by oxidation to highly electrophilic metabolites that covalently bind to active site cysteine residues. Thus, oxidative metabolites of intraneuronal 5-S-CyS-DA may contribute to impaired mitochondrial complex I and α-KGDH activities known to occur in the parkinsonian SNc, and suggest that impaired PDHC evoked by the same metabolites may also occur in PD.

Keywords: Parkinson’s disease, 5-S-cysteinyldopamine, DHBT-1, pyruvate dehydrogenase complex.
Introduction

The degeneration of neuromelanin-containing dopaminergic neurons in the substantia nigra pars compacta (SNc) with resultant decrements of striatal dopamine (DA) is known to account for the motor symptoms of idiopathic Parkinson’s disease (PD) (Hornykiewicz, 1980; Kish et al., 1988). The mechanism that underlies the selective degeneration of dopaminergic SNc cells in PD is unknown. Nevertheless, several characteristic pathobiochemical changes are known to occur in the parkinsonian SNc. These include decreased activity of mitochondrial complex I (Schapira et al., 1990a,b) and the α-ketoglutarate dehydrogenase (α-KGDH) complex (Mizuno et al., 1994). The complex I defect is not accompanied by altered activities of mt complexes II–IV, is specific to the SNc and exclusive to PD (Schapira et al., 1990a,b). The activity of superoxide dismutase (SOD) is also increased in the parkinsonian SNc (Saggu et al., 1989). Since mutations of SOD genes have not been associated with PD (Parboosingh et al., 1995), increased SOD activity implies an adaptive response to elevated superoxide (\(O_2^•−\)) generation. 3-Nitrotyrosine (3-NT) immunoreactivity associated with Lewy bodies in remaining SNc cells in PD, indicative of protein nitration by peroxynitrite (\(\text{ONOO}^−\)) (Good et al., 1998), suggests reaction of elevated cytoplasmic \(O_2^•−\) with nitric oxide (\(\text{NO}^−\)) and therefore activation of neuronal nitric oxide synthase (nNOS) (Hunot et al., 1996). Another characteristic change in the parkinsonian SNc is a large fall of glutathione (GSH) levels compared to those measured in matched control subjects (Sofic et al., 1992; Sian et al., 1994a) that, in part, reflects its loss from remaining pigmented DA neurons (Pearce et al., 1997). A similar fall of GSH also occurs in the SNc of individuals who died with incidental Lewy body disease (ILBD), which is believed to be an early preclinical stage of PD (Dexter et al., 1994). The fall of nigral GSH in ILBD approximately coincides with the initial appearance of a small mt complex I defect (Dexter et al., 1994). Decreased nigral GSH therefore appears to be a very early component of the pathological processes that underlie the death of dopaminergic SNc neurons in PD. Indeed, it has been suggested that low GSH levels and consequent oxidative stress might contribute to the degeneration of SNc neurons in PD (Cohen, 1983, 1986). However, while oxidative damage to lipids, proteins and nucleic acids has been detected in SNc tissue of patients who died at advanced stages of PD (Jenner and Olanow, 1996) such damage is not evident in SNc tissue from ILBD patients (Dexter et al., 1994; Alam et al., 1997). Furthermore, decreased nigral GSH in both ILBD and PD is not accompanied by increased levels of glutathione disulfide (GSSG) (Sofic et al., 1992; Sian et al., 1994a; Dexter et al., 1994). Thus, the fall of nigral GSH in the parkinsonian SNc, is not caused by its reactive oxygen species/reactive nitrogen species-mediated oxidation to GSSG, impaired production of mt ATP needed for its biosynthesis (Seeleg and Meister, 1985), at least at early stages of PD, or to decreased activity of \(\gamma\)-glutamyl cysteine synthetase (Sian et al., 1994b), the rate-limiting enzyme for biosynthesis of the tripeptide. However, the activity of \(\gamma\)-glutamyl transpeptidase (\(\gamma\)-GT) is significantly increased in the parkinsonian SNc (Sian et al., 1994b). This is of interest because \(\gamma\)-GT, an enzyme present on the