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Estrogens and Parkinson Disease

Novel Approach for Neuroprotection

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Epidemiologic studies revealed that the prevalence of Parkinson disease is higher in males than in females and that the progression of the disease might be rapid in males compared with females. The reason for the gender difference is unknown; however, estrogens may be involved. Many studies have revealed that estrogens provide neuroprotective effects and that the protective mechanisms include antioxidant property and upregulation of Bcl-2, brain-derived neurotrophic factor, and glial cell–derived neurotrophic factor (GDNF). Upregulation of Bcl-2 or GDNF is mediated by nonnuclear estrogen receptor (ER) in addition to transcription regulation by ER. To avoid undesirable effect of estrogens, several selective ER modulators, raloxifene and genistein are considered.

Key Words: Dopaminergic neuron; oxidative stress; estrogen receptor; selective estrogen receptor modulator.

Introduction

Parkinson disease is one of the major neurodegenerative disorders, and it is characterized by chronic progression of mesencephalic dopaminergic neuronal degeneration. Although the pathogenetic mechanism is still unresolved, the disease is thought to be a multietiologic disorder related to genetic disorders, environmental toxicity inhibiting the mitochondrial complex I activity, and neuronal damage associated with aging. Clinical features of the disease include muscular rigidity, resting tremor, and bradykinesis, and the disability is much improved by pharmacologic replacement of deficient dopamine (DA) with levodopa. However, within 5–10 yr from the clinical onset, the benefit of replacement therapy is deteriorated because of progression of dopaminergic neuronal loss. A recent study using [^{18}F]-labeled carbomethoxyfluorophenyl-tropane as a marker of DA transporter revealed that striatal terminals of dopaminergic neurons are reduced to 42% in the putamen and 76% in the caudate in patients with Parkinson disease compared with age-matched control subjects. In patients with Parkinson disease, annual decline of the dopaminergic neuronal terminals is estimated to be 13.1% (putamen) and 12.5% (caudate), whereas it is 2.1 and 2.9% in control patients, respectively *(1)*. Another study using [^{123}I]-carbomethoxy-iodophenyl-tropane showed that the annual decline is estimated to be 7.1% in the striatum *(2)*. In this context, it is important to find a therapeutic approach inhibiting progression of dopaminergic neuronal degeneration.

Epidemiologic studies indicate male predominance in Parkinson disease, and the relative proportion of male to female sufferers is 1.36–3.7 *(3,4)*. Furthermore, a prospective study in which 297 consecutive patients (181 men and 116 women) with Parkinson disease were evaluated according to the Unified Parkinson Disease Rating Scale (UPDRS) showed that clinical progression of the disease might be significantly rapid in males rather than females, especially on UPDRS subscores I (mental state) and II (activity of daily living) *(5)*. Although the reason for the gender difference in prevalence and progression of the disease is still unknown, the level of estrogens or progesterone, or expression of their receptors may be involved in the pathologic process of dopaminergic neuronal degeneration. Intriguingly, Leranth et al. *(6)* investigated the relationship between ovariectomy and dopaminergic neurons in African green monkeys and revealed that the density of mesencephalic dopaminergic neurons is higher in females than in males. Furthermore, they demonstrated that ovariectomy reduces the number of dopaminergic neurons to 64% of the intact female monkeys and the reduction is reversed by estrogen replacement therapy (ERT) *(6)*. Therefore, estrogens are the key in the gender difference of the disease and ERT may be applied to protect mesencephalic dopaminergic neurons from neurodegeneration.

Molecular Mechanisms of Neuroprotection by Estrogens

Antioxidant Property

Estrogens have antioxidant property in a variety of in vitro and in vivo models *(7–10)*, which is determined by the presence of the hydroxyl group in the C3 position on the A ring of the steroid structure *(11)*. Oxidative stress plays an important role in dopaminergic neuronal degeneration in Parkinson disease, and, therefore, we investigated neuropro-

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tection by estradiol using rat primary mesencephalic neuronal culture. Although estradiol provides neuroprotective effect against experimental oxidative stress, much higher (10^{-6}–10^{-4} *M*) than physiologic concentration is needed for the antioxidant effects in vitro *(12)*. The antioxidant property is strongly reinforced by the presence of the reduced form of glutathione *(13)*. Because mesencephalic nigral neurons are abundant in glutathione compared with other regions of the brain *(14)*, estradiol might have an antioxidant effect at physiologic concentrations.

Bcl-2 Upregulation

The estrogen receptor (ER) is among ligand-dependent transcription factors. Bound with ligands, the ER binds to several DNA consensus elements including the estrogen response element (ERE) and the activating protein-1 (AP-1) site. When bound to the ERE, ER acts as an enhancer of ER-regulated genes, which include several apoptosis-related genes, and Bcl-2, an antiapoptotic protein, is upregulated by estrogens through transcription mediated via two EREs within the coding sequencing *(15)*. Although the upregulation of Bcl-2 is thought to be mainly mediated by transcription regulation through the EREs, several other mechanisms are also involved. Recently, we demonstrated that estrogens upregulate Bcl-2 by CREB phosphorylation downstream from Akt phosphorylation by phosphatidylinositol 3-kinase (PI3-K) *(16,17)*. Akt phosphorylation begins within 15 min after exposure to estradiol and the phosporylation is not completely blocked by the ER antagonist ICI 182,780. Therefore, activation of PI3-K/Akt signal by estrogens is independent of transcription regulation by the ERs. As well as Akt phosphorylation, estrogen stimulates rapidly a second-messenger system such as adenylate cyclase, protein kinase A (PKA), PKB, PKC, and mitogen-activating protein kinase *(18)*. These rapid responses of second messengers by estrogens are thought to be mediated by nonnuclear ERs.

Neurotrophin Upregulation

In addition to Bcl-2, estrogen elevates the expression of neurotrophins, brain-derived neurotrophic factor *(19)*, and glial cell–derived neurotrophic factor (GDNF) *(20)*. Upregulation of GDNF by estradiol is not blocked by ICI 182,780, which acts as a pure antagonist without partial agonistic effects, but blocked by inhibition of cyclic adenosine monophosphate–dependent PKA signals and, therefore, is thought to be independent of nuclear ERs and might be associated with signal transduction mediated by nonnuclear ERs *(20)*.

Inhibition of Catechol-o-methyltransferase

Catechol-*o*-methyltransferase (COMT) is an enzyme that is crucial for dopamine (DA) metabolism in the brain. Several COMT inhibitors are applied for clinical treatment for Parkinson disease. It has been reported that physiologic concentrations of estradiol reduce gene expression of COMT and the reduction is mediated by the ER *(21)* and that low-dose, conjugated estrogen (i.e., 0.625 mg of Premarin) improves motor disability in postmenopausal patients with Parkinson disease *(22)*.

Neuroprotection from 1-Methyl-4-phenyl-pyridine or 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine Neurotoxicity

Although the pathogenesis has not been clarified in sporadic cases, exogenous or endogenous neurotoxins, such as 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) *(23, 24)* or MPTP analogs *(25,26)*, may cause dopaminergic neuronal degeneration. MPTP is converted to 1-methyl-4-phenylpyridinium (MPP^+) by monoamine oxidase and is taken up into dopaminergic neurons through the DA transporter. It inhibits the activity of mitochondrial complex I and causes selective dopaminergic neuronal death *(27,28)*. MPP^+-induced neuronal death displays features characteristic of the disease; the dopaminergic neuronal death is accompanied by the specific inhibition of mitochondrial complex I activity. Estrogens protect dopaminergic neurons from MPP^+ *(29–31)* or MPTP *(32)*. The included molecular mechanisms are antioxidant effects *(29)*, restoration of DA transport *(29)*, upregulation of Bcl-2, and suppression of c-Jun terminal kinase *(30)*.

Transcription Regulation through ERβ

In the central nervous system (CNS), the distribution of the ER was believed to be restricted to the cerebral cortex, hippocampus, amygdala, and preoptic tegmentum, until identification of the novel type of ER, ERβ. ERs, at the present time, are classified into two forms—ERα and ERβ *(33)*—and the dominant type of ER in the CNS has been shown to be ERβ. In the adult midbrain, the subtype of ER expressed is exclusively ERβ *(34,35)*. Because of homology of amino acid residues in the ligand-binding domain of ERβ and ERα, many ligands such as 17β-estradiol, tamoxifen, and raloxifene have affinity to both subtypes *(36)*. However, the manner of transcription regulation by these ligands is different between ERα and ERβ *(37)*. Transcription mediated via the ERE is enhanced by binding of estrogens such as 17β-estradiol to either ERα or ERβ, and the enhancement is blocked by tamoxifen and raloxifene. However, in application of antiestrogens alone, tamoxifen and raloxifene enhance ERE transcription when bound to ERα. By contrast, when bound to ERβ, antiestrogens provide no agonistic effects *(36)* (Table 1).

Several investigations demonstrate that the ERβ, but not ERα, is associated with regulation of apoptosis, especially of cancer cells *(38,39)*. In this context, mesencephalic dopaminergic neuronal apoptosis may be affected by ERβ. Using primary culture of the rat embryonic mesencephalon in which ERβ is expressed but ERα is not detected, we demonstrated that oxidative stress–induced apoptosis of dopaminergic neurons is suppressed by estrogens, but strongly enhanced by antiestrogen, tamoxifen *(40)*. In this context, oxidative stress-induced apoptosis could be regulated by the ERβ in mesencephalic dopaminergic neurons.

Table 1
Gene Transcription Regulation through ERα and ERβ

	ERE	AP-1
ERα		
17β-estradiol	Agonistic	Agonistic
Tamoxifen	Weakly agonistic	Agonistic
Raloxifene	Weakly agonistic	Agonistic
ERβ		
17β-estradiol	Agonistic	Antagonistic
Tamoxifen	Not agonistic	Agonistic
Raloxifene	Not agonistic	Agonistic

Issues in Estrogen Therapy for Parkinson Disease and Selective ER Modulators

Epidemiologic data demonstrate that estrogens can provide protective effects against dopaminergic neuronal degeneration in Parkinson disease, and a series of studies investigating neuroprotective mechanisms has revealed complex mechanisms including antioxidant property, transcription regulation of genes associated with apoptosis, and protective signal transduction mediated by nonnuclear ERs. In addition to these complex molecular mechanisms, adverse effects of estrogen therapy such as increased risk of uterine and breast cancer, enhanced coagulation, undesirable feminizing effects in male patients have to be resolved. In this context, selective ER modulators (SERMs), raloxifene and genistein, are used as novel neuroprotective agents in several studies. Actually, raloxifene has been revealed to prevent in vivo DA depletion by MPTP *(32)*, and genistein provides neuroprotection against neuronal apoptosis induced by endoplasmic reticulum stress *(41)*. Neuronal apoptosis induced by endoplasmic reticulum stress is taken as an important model of not only Parkinson disease but also neurodegenerative diseases. It is important that clinical trials continue to be conducted in order to determine whether these SERMs work as neuroprotective agents in the disease.

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