Original Article

Voltage-dependent anion channel involved in the α-synuclein-induced dopaminergic neuron toxicity in rats

Lingling Lu, Chunyan Zhang, Qing Cai, Qiang Lu, Chunli Duan, Yuangang Zhu, and Hui Yang*

Beijing Center of Neural Regeneration and Repair, Key Laboratory for Neurodegenerative Diseases of the Ministry of Education, Beijing Institute of Brain Disorders, Capital Medical University, Beijing 100069, China
*Correspondence address. Tel: +86-10-83950070; Fax: +86-10-83950070; E-mail: huiyang@sina.com

Inclusion bodies containing the neural protein α-synuclein (α-syn) are observed in several neurodegenerative diseases, including Parkinson’s disease (PD). Furthermore, over-expression of α-syn in rat brain partly mimics the neuropathological and behavioral features of PD by triggering the degeneration of dopaminergic neurons in the substantia nigra (SN). Mitochondrial dysfunction is also central to PD pathogenesis, and α-syn is found in the mitochondria. However, the precise mechanisms of α-syn-induced neurotoxicity remain elusive. To examine the potential mechanisms of α-syn-induced neurodegeneration, we over-expressed α-syn in the SN of rats using a recombinant adeno-associated viral vector (rAAV-syn). Immunohistochemical and immunogold labeling results indicated that α-syn was successfully over-expressed in the SN and striatum after vector injection. The number of tyrosine hydroxylase-positive (dopaminergic) neurons was significantly reduced in rats injected with rAAV-syn when compared with control rats. Compared with control rats, the density of α-syn-conjugated gold particles was greater in the axons, cytoplasm, nuclei, and notably also in the mitochondria of SN neurons in rAAV-syn-injected rats. In addition, SN neurons transfected with rAAV-syn exhibited swollen mitochondria with discontinuous outer membranes and internal vacuole-like structures, strongly suggesting α-syn-induced mitochondrial dysfunction. Mitochondria in rAAV-syn-injected rats were also observed in autophagosomes. α-Syn co-immunoprecipitated with voltage-dependent anion channel 1 (VDAC1), a component of the mitochondrial permeability transition pore (mPTP) that induces mitochondrial uncoupling and apoptosis. Over-expression of α-syn may cause the degeneration of dopaminergic neurons through an interaction with mitochondrial VDAC1, which leads to mPTP activation, mitochondrial uncoupling, and cell death.

Keywords Parkinson’s disease; α-synuclein; electron microscope; subcellular localization; mitochondrial dysfunction

Introduction

α-Synuclein (α-syn), a small soluble acidic protein enriched in the brain, has been implicated in the pathogenesis of both familial and sporadic Parkinson’s disease (PD) [1–4]. Aggregates of α-syn are a major component of Lewy bodies (LBs), a hallmark of dopaminergic neurons in PD patients [5]. Furthermore, over-expression of the α-syn gene in rodents and primates can mimic the neuropathology and motor symptoms of PD. However, it remains unknown how α-syn contributes to the pathophysiological processes underlying PD. α-Syn was first identified as a nuclear and presynaptic protein in the mammalian brain. Our recent study and those of several other groups have demonstrated α-syn expression in axons and cytoplasm, as well as high expression in mitochondria [6,7], suggesting that α-syn may cause neurodegeneration by disrupting mitochondrial function.

Neurons are particularly sensitive to mitochondrial dysfunction, as the high energy demands and unique morphology of neurons require a precise subcellular distribution of mitochondria. Other aspects of mitochondrial function, including cytosolic calcium buffering and regulation of apoptosis, are also critical for neuronal survival. Mitochondrial dysfunction has been linked to several neuropathologies and neurodegenerative diseases, but evidence for a critical role of mitochondrial dysfunction in disease progression is particularly strong for PD [8–13]. The mitochondrial permeability transition pore (mPTP) is a multi-protein complex including the outer membrane protein voltage-dependent anion channel (VDAC) [14], the mitochondrial matrix protein cyclophilin D [15], and the inner membrane protein adenylate translocator [16]. When the mPTP opens, the transmembrane potential that couples electron transport to oxidative phosphorylation is lost (uncoupling), and the mitochondrial inner membrane becomes freely permeable to solutes <1.5 kDa, causing organelle
swelling, loss of calcium buffering capacity, and the release of factors that initiate apoptosis [17]. Although several studies have demonstrated α-syn in mitochondria, the functional implications of mitochondrial α-syn remain unclear. For instance, it is not known whether α-syn interacts with the mPTP to regulate the permeability transition.

In this study, we over-expressed the α-syn gene in the substantia nigra (SN) of rats. The subcellular distribution of α-syn in SN and striatum was determined by immunohistochemistry and immunogold labeling. The effects of α-syn over-expression on dopaminergic cell survival was assessed by counting tyrosine hydroxylase (TH)-positive neurons in the SN and striatum, whereas effects on SN mitochondrial function were evaluated by ultrastructural analysis under electron microscopy. Finally, co-immunoprecipitation (Co-IP) and dual immunohistochemical labeling were used to demonstrate an interaction between α-syn and VDAC. Over-expression of α-syn reduced the number of dopaminergic neurons in the SN and mitochondria exhibited the morphological and physiological signs of dysfunction.

Materials and Methods

Reagents and animals

The 3D5 anti-α-syn monoclonal antibody was produced and characterized as described previously [18]. A BCA protein assay kit and an enhanced chemiluminescence kit were purchased from Pierce (Rockford, USA). Biotinylated goat-anti-mouse immunoglobulin G (IgG), horseradish peroxidase (HRP)-conjugated streptavidin, and HRP-conjugated goat-anti-mouse IgG were obtained from Vector Laboratories (Burlingame, USA). R-Gent selenium-enhanced electron microscopy (SE-EM) silver enhancement reagent was from Aurion (Wageningen, The Netherlands). The monoclonal antibodies anti-VDAC, anti-β-actin, and anti-TH were purchased from Cell Signaling Technology (Danvers, USA). Protease and phosphatase inhibitor cocktails were from Sigma (St Louis, USA).

To assess degeneration of dopaminergic neurons under normal α-syn expression and over-expression, 64 specific pathogen-free (SPF) Sprague Dawley rats (180–220 g) were divided into two groups: one group of 32 was injected with recombinant adeno-associated viral vector containing α-syn (rAAV-syn) into the SN, whereas the other (control) group was injected with rAAV-LacZ. Each group was further divided into four groups of eight that were sacrificed sequentially with 3D5 anti-α-syn or anti-TH monoclonal antibody (1 : 10,000), biotinylated goat-anti-mouse IgG (1 : 1000), and HRP-conjugated streptavidin. The bound peroxidase was subsequently revealed using a solution containing diaminobenzidine, hydrogen peroxide, and nickel ammonium sulfate. To control for non-specific staining, some of the sections were incubated in a pre-absorbed 3D5 antibody solution. Briefly, 20-μm-thick sections were blocked with 5% normal goat serum, and then incubated sequentially with 3D5 anti-α-syn or anti-TH monoclonal antibody (1 : 10,000), biotinylated goat-anti-mouse IgG (1 : 1000), and HRP-conjugated streptavidin. The bound peroxidase was subsequently revealed using a solution containing diaminobenzidine, hydrogen peroxide, and nickel ammonium sulfate. To control for non-specific staining, some of the sections were incubated in a pre-absorbed 3D5 antibody solution. Briefly, different amounts of recombinant α-syn (0.8, 1.6, and 3.2 ng/ml) were added to the diluted antibody solution (1 : 10,000, 0.4 ng/ml of 3D5 antibody) and incubated at 4°C overnight. The solutions were then incubated with protein A, followed by centrifugation at 12,000 × g for 5 min at 4°C. The procedures for the pre-absorption of antibodies used for immunogold labeling were the same as above except that the antigen concentrations were increased to maintain the same antigen: antibody ratios (2, 4, and 8). The stained sections were mounted on glass slides, dehydrated, cleared, and sealed under cover slips.

Immunogold staining and electron microscopy

Sections were fixed, labeled, and imaged under a transmission electron microscope as described [18]. Briefly, sections of 50 μm thickness were treated with 0.1% NaBH₄ and blocked with a solution containing 5% bovine serum albumin (BSA), 5% normal serum, and 0.1% gelatin. The sections were then incubated with 3D5 monoclonal antibody [1 : 500 in 0.1 M phosphate buffer (PB), pH 7.4] overnight at 4°C and then with a gold-conjugated goat-anti-mouse IgG (1 : 50) for 6 h at room temperature. Afterfixing in PB with 2% glutaraldehyde for 10 min, the sections were silver-enhanced, fixed with 1% osmium tetroxide, dehydrated in an ascending ethanol series, and flat-embedded in Epoxy resin. Ultrathin sections were examined under an electron microscope (Nihon Kohden 1230; Nihon Kohden, Tokyo, Japan) after counterstaining with uranyl acetate and lead citrate.
The density of silver-enhanced gold particles in different subcellular structures was measured by using Leica Qwin standard V2.8 software (Leica, Cambridge, UK) as described previously [6]. Briefly, electron micrographs were captured from five serial EM sections and the boundaries of different subcellular structures drawn. The enclosed areas within the boundaries were automatically calculated by the software, and the gold particles within an area were marked and counted automatically. The density of gold particles in each structure was calculated as gold particle number per μm².

The cell ultrastructure was analyzed by electron microscopy. Cells were fixed in 4% paraformaldehyde and 1% glutaraldehyde in 0.1 M PB, pH 7.4, for 1 h at 4°C. For conventional osmium–uranyl lead staining, sections were stained overnight with 1% aqueous uranyl acetate, dehydrated in an ascending series of ethanol solutions (up to 100%), and processed through a progressively increasing ratio of resin-VCD4260 (Sigma) to ethanol with a final step of 100% VCD4260. After overnight infiltration in pure resin, sections were embedded between strips of ACLAR plastic film (Electron Microscopy Sciences, Hatfield, USA) and the resin polymerized at 45°C for 12 h followed by an additional 24 h at 60°C. Ultrathin sections were then cut on an ultramicrotome (Leica) and mounted on mesh nickel grids (Electron Microscopy Sciences), counterstained with lead citrate, and then examined under a transmission electron microscope (JEOL-1230; JOEL Ltd., Tokyo, Japan). For immunogold electron microscopy, the sections were heated in citrate buffer, blocked in 5% BSA, and then incubated overnight at 4°C with mouse monoclonal anti-α-syn antibody (BD Biosciences, San Jose, USA) followed by incubation to the secondary antibody (biotin-conjugated goat-anti-mouse antibody (BD Biosciences, San Jose, USA) and immunoprecipitated with a rabbit polyclonal antibody (Pierce) and immunoprecipitated with a rabbit polyclonal α-syn antibody (3D5, 3 μg) or VDAC1 monoclonal antibody (3 μg), followed by western blot with VDAC1 or α-syn antibody.

Co-immunoprecipitation
For Co-IP analysis, the proteins from the SN of rats were lysed in non-denaturing lysis buffer with protease inhibitors. The lysates were pre-cleared with Protein-G agarose (Pierce) and immunoprecipitated with a rabbit polyclonal α-syn antibody (3D5, 3 μg) or VDAC1 monoclonal antibody (3 μg), followed by western blot with VDAC1 or α-syn antibody.

Statistical analysis
All data were analyzed by Student’s t-test or one-way analysis of variance post hoc using SPSS V13.0 software (IBM, Armonk, USA). All the measurements were performed independently by two researchers.

Results
Degeneration of SN dopaminergic neurons over-expressing α-syn
The human α-syn gene was introduced into the SN of rat brain by injection of the rAAV-syn. The left SN was injected with rAAV-syn or rAAV-LacZ (control group) and sacrificed 4, 8, 12, or 16 weeks post-injection to assess the effects of rAAV-syn on α-syn expression and the viability of SN dopaminergic neurons. Immunohistochemical staining revealed increased α-syn expression in the left SN (injected side) compared with the right uninjected SN (control side) [Fig. 1(A,B)], while double staining demonstrated that α-syn co-localized with the dopamine synthesis enzyme TH (Supplementary Fig. S1). We did not observe obvious LB-like inclusions, but we observed α-syn-positive aggregates in SN neurons (Supplementary Fig. S1).

Immunohistochemical staining for TH revealed a significant loss of dopaminergic neurons in the SN [Fig. 1(C)] and a loss of dopaminergic fibers in striatum [Fig. 1(D,E)] at 12 weeks after rAAV-syn injection. The SN TH-positive neurons number exhibited a progressively decreasing manner from 8 to 16 weeks post-rAAV-syn injection [Fig. 1(F,G), Supplementary Fig. S2].

Subcellular distribution of α-syn-immunoreactive gold particles
The full spectrum of α-syn functions is still unknown. Subcellular location provides a strong clue to protein function, so we investigated the subcellular localization of α-syn by immunogold labeling and electron microscopy in both control and rAAV-syn-injected rats. Four weeks after SN injection, the total density of gold particles was greater in SN and striatum of rAAV-syn-injected rats than in control rats. In control rats, gold particles were localized mainly in presynaptic terminals [Fig. 2(A)] and mitochondria [Fig. 2(B)]. In contrast, no gold particles were found in the presynaptic terminals of rAAV-syn-injected rats [Fig. 2(D)], but gold particles were found in mitochondria [Fig. 2(E)] and axons [Fig. 2(G)] of rAAV-syn-injected rats. Nonetheless, α-syn-immunoreactive gold particles are also observed in neurofilaments in both control and rAAV-syn-injected rats [Fig. 2(H,I)]. Statistically, rAAV-syn-injected rats showed significantly higher α-syn-immunoreactive gold particle densities in axons, nuclei, cytoplasm, and mitochondria [Fig. 2(J)].

In the striatum of control rats, most gold particles were detected in the mitochondria [Fig. 3(C)] and presynaptic terminals [Fig. 3(B)], with relatively a few in axons [Fig. 3(A)], nuclei, and cytoplasm [Fig. 3(C)]. In the striatum of rAAV-syn-injected rats, gold particles are also
observed in axons [Fig. 3(D)], presynaptic terminals [Fig. 3(E)], nuclei [Fig. 3(F)], mitochondria [Fig. 3(G)], cytoplasm [Fig. 3(H)], and myelin [Fig. 3(I)]. Statistically in stark contrast to SN neurons following rAAV-syn injection, most gold particles in the striatum of rAAV-syn-injected rats were located in presynaptic terminals, whereas nuclei and mitochondria exhibited substantially fewer particles than SN [Fig. 3(J)]. In total, however, striatal neurons of rAAV-syn-injected rats still exhibited significantly more gold particles than control rats.

In summary, immunogold labeling of brain slices from rAAV-syn-injected rats indicated that α-syn was highly over-expressed in the nuclei of neurons, and was also significantly higher in SN neuron, cytoplasm, axons, and mitochondria compared with control rats. In contrast, virtually no α-syn was expressed in SN presynaptic terminals of rAAV-syn-injected rats (Fig. 2). Over-expression of human α-syn in the SN also led to a dramatic increase in α-syn expression in striatal presynaptic terminals, nuclei, and cytoplasm (Fig. 3).

**Ultrastructural damage to mitochondria in SN of rats over-expressing α-syn**

Both mitochondrial dysfunction and α-syn over-expression have been implicated in the pathogenesis of PD. To investigate a possible link between these two, we evaluated the impact of α-syn over-expression on mitochondrial ultrastructure of SN neurons 16 weeks after rAAV-α-syn injection, a time when α-syn over-expression exhibited significant neurodegeneration of dopaminergic neurons.
Electron microscopy revealed a typical mitochondrial morphology in control rats [Fig. 4(A)], while swollen mitochondria, mitochondria with discontinuous outer membranes [black arrow, Fig. 4(B)], mitochondria with vacuole [black pointer, Fig. 4(B)], and mitochondria within electron-dense inclusions [box, Fig. 4(B)] in the SN neurons of rAAV-syn-injected rats.

**Discussion**

Over-expression of α-syn in rat brain partially mimics the pathology of PD [19]. Furthermore, α-syn aggregates are observed in the dopaminergic neurons of PD patients, so induced α-syn over-expression in animals is a potential model for PD. However, the role of α-syn in the pathogenesis of PD remains an enigma. We over-expressed α-syn in the rat SN and examined the ensuing pathological responses, specifically degeneration of dopaminergic neurons, mitochondrial dysfunction, and the possible role of α-syn in activation of mPTP that triggers neuronal cell death. Our results clearly demonstrated that over-expression of α-syn in the rat SN led to loss of SN dopaminergic neurons, consistent with the previous studies [20–22], but we further showed that α-syn-induced neurodegeneration was associated with mitochondrial dysfunction. As mitochondrial dysfunction has been implicated in PD, rats with targeted over-expression of α-syn may represent a valuable animal model to investigate this aspect of PD pathogenesis.

To determine the pathogenic mechanisms of α-syn over-expression in SN dopaminergic neurons, we first investigated the subcellular distribution of α-syn using immunogold labeling under transmission electron microscopy (TEM). The inset (top right) is a magnification of the area demarcated by the central black box in each figure. α-Syn-positive gold particles mainly localized in presynaptic terminals (PSTs) (A), mitochondria (B), nucleus and cytoplasm (C) of SN neurons in uninjected control rats. In contrast, rAAV-syn-injected rats exhibited a greater number of α-syn-conjugated gold particles in mitochondria (E), nuclei and cytoplasm (F), and axons (G), while no gold particles were observed in PSTs (D). α-Syn-conjugated gold particles were also observed associated with neurofilaments (H, I). (J) Quantification of α-syn-conjugated gold particles in the axons, PSTs, nuclei, cytoplasm, and mitochondria of SN neurons. *P < 0.05 and †P < 0.01, n = 3. Bar = 0.4 μm (A, I), 2 μm (B, E, G, H), 1 μm (C, D, F).

**α-Syn interacts with VDAC, a major component of the mPTP complex**

To investigate possible pathogenic mechanisms of mitochondrial α-syn, we measured the interaction between α-syn and VDAC proteins by Co-IP and co-localization by double immunofluorescent staining. Co-IP indicated an interaction between α-syn and VDAC ([Fig. 5]) as both α-syn and VDAC antibodies precipitated the other non-target protein. Confocal imaging of dopaminergic MN9D cells double-labeled with anti-α-syn and anti-VDAC antibodies confirmed the co-localization of α-syn and VDAC1 ([Fig. 6]). Moreover, the fluorescence overlap (yellow emission) was stronger in cells over-expressing α-syn ([Fig. 6(B)]) than in cells of control transfected with empty vector ([Fig. 6(A)]).
labeling and electron microscopy 4 weeks after injection of rAAV-syn viral expression vector. The average density of gold particles was greater in both SN and striatum of rAAV-syn-injected rats compared with control rats, and increased immunostaining was observed in axons, nuclei, cytoplasm, and mitochondria, whereas expression was

---

**Figure 3** Subcellular distribution of α-syn in striatal neurons as revealed by immunogold labeling under TEM Subcellular distribution of α-syn in striatal neurons of control (A–C) and rAAV-syn-injected rats (D–I). The box in the upper right corner of each figure (except C) is an enlargement of the area demarcated by the black box, while the lower right box in (C) shows gold-conjugated α-syn in the mitochondria of a striatal neuron from a control rat. In control rats, α-syn-conjugated gold particles localized mainly in axons (A), PSTs (B), mitochondria (inset of C), and nuclei and cytoplasm (C) of control rats. In rAAV-syn-injected rats, α-syn-conjugated gold particles were localized to axons (D), PSTs (E), nuclei (F), cytoplasm (H), and myelin (I), but fewer were observed in striatal mitochondria (G). (J) Quantification and statistical analysis of gold particles in axons, PSTs, nuclei, cytoplasm, and mitochondria. *P < 0.05 and **P < 0.01, n = 3. Bar = 2 μm (A, D, G, H, I), 1 μm (B, E), 0.4 μm (C, F).

---

**Figure 4** Over-expression of α-syn in rat SN was accompanied by ultrastructural anomalies including signs of mitochondrial dysfunction (A) In control rats, SN neurons had typical morphological features. (B) In rAAV-α-syn-injected rats, many mitochondria in SN neurons exhibited discontinuous outer membranes (black arrow), swelling, and vacuole-like structures (black pointers). In addition, SN neurons from rAAV-α-syn-treated rats (B) exhibited electron-dense inclusions (black box) and electron-dense inclusions containing membranous structures and mitochondria that appeared similar to autophagosomes. Bar = 1 μm.

---

**Figure 5** α-Syn co-immunoprecipitated with VDAC1 Sixteen weeks after rAAV-syn injection, proteins were extracted from the rat SN and immunoprecipitated with an anti-α-syn antibody or an anti-VDAC1 antibody (or control IgG). Immunoprecipitation indicated an interaction between α-syn and the mPTP protein VDAC.
almost zero in presynaptic terminals of SN. In contrast, rAAV-syn injection resulted in higher α-syn expression (as indicated by gold particle density) in presynaptic terminals of striatum. Similar to the SN, α-syn expression was also enhanced in striatal nuclei, cytoplasm, and axons, but expression in mitochondria was slightly lower in rAAV-syn-injected rats than in controls.

Since α-syn was over-expressed in dopaminergic neurons at SN, its distribution on striatal presynaptic terminals, where the dopaminergic neurons project to, was increased accordingly. However, its location on striatal mitochondria was decreased. The striatal mitochondria may belong to presynaptic terminals of dopaminergic neurons from SN, or to presynaptic terminals of GABAergic neurons from cortex, or to the interneurons or glia cells which reside in striatum. Given the complex ownership of mitochondria, it is difficult to tell why α-syn on striatal mitochondria decreased. Further investigations to discriminate the origin of striatal mitochondria are needed to explain why α-syn location on striatal mitochondria was decreased.

Mitochondrial dysfunction has been implicated to some degree in most neurodegenerative diseases, including PD [8–10]. Ultrastructural analysis of mitochondria in SN neurons over-expressing α-syn revealed significant swelling, loss of outer membrane integrity, and formation of electron-dense inclusions containing mitochondria, suggesting that α-syn over-expression damaged dopaminergic (TH-positive) neurons by disrupting mitochondrial function which is consistent with the previous studies [6,23].

Pioneering studies by Haworth and Hunter [24,25] and later by Crompton et al. [26] showed that this mitochondrial swelling might result from enhanced opening of a non-specific ionic channel known as the mPTP. Recently, α-syn was found in the mitochondrial membrane of mouse dopaminergic neurons [7,27], and we also observed specific α-syn immunoreactivity in both the mitochondrial cristae and matrix in rat SN neurons [6]. Here, we expanded on these findings by showing that α-syn interacts with VDAC, a mitochondrial channel critical for the permeability transition [28,29]. It is thus possible that α-syn regulates mPTP opening by interacting with VDAC. In the present study, the interaction between α-syn and VDAC was observed in both control dopaminergic MN9D cells transfected with empty vector and syn cells over-expressing α-syn, as indicated by dual immunohistochemical staining, it was stronger in cells over-expressing α-syn. In light of our EM data showing loss of mitochondrial membrane integrity, these data suggest that the interaction of α-syn with VDAC may promote opening of the mPTP. One major consequence of this enhanced mPTP opening is that the mitochondrial membrane can no longer maintain a barrier to protons, which leads to dissipation of the proton motive force. The resulting uncoupling of oxidative phosphorylation inhibits mitochondrial production of ATP, leading to bioenergetic failure [28]. A second consequence of mPTP opening is that molecules < ∼1500 Da can diffuse out of the mitochondria, including Ca^{2+} and the apoptotic trigger cytochrome c. Moreover, mitochondrial swelling [28], as shown in **Fig. 4**, may permanently damage the membrane. Enhanced interaction between α-syn and VDAC concomitant with α-syn over-expression and subsequent mPTP activation may explain the neuropathology observed in a familial form of PD with α-syn triplication.

Ultrastructural analysis revealed electron-dense inclusions in some SN neurons from α-syn over-expressing rats.
These electron-dense inclusions, which were bounded by a continuous membrane and often contained mitochondria, resembled autophagosomes and suggested the presence of mitophagy, a selective autophagy of mitochondria, which is an important mitochondrial quality control mechanism that eliminates damaged mitochondria. Previous studies reported an increase of mitophagy in α-syn/A53T (A53T mutant of α-syn) or α-syn/WT (wild-type α-syn) over-expressing mid-brain dopaminergic neurons [30–32]. The role of mitophagy in the pathogenesis of α-syn is still controversial. Some studies showed that it is likely a compensatory attempt to remove defective mitochondria, whereas others reported that it is a culprit responsible for α-syn toxicity. Sampao-Marques et al. [33] reported that the expression of human wild-type α-syn gene and the clinical A53T mutant in yeast results in a reduced chronological life span (CLS) together with an increase of mitophagy activities. Impairment of mitophagy by deletion of ATG11 or ATG32 resulted in a CLS extension, further implicating mitophagy in the α-syn toxicity. Some molecules, e.g. Atg8, Atg32, Atg11, Uth1p, and Aup1p, are identified to be responsible for mitophagy regulation. However, mammalian mitophagy presents some unique features that may also be specific to mammals. What most interested us is that mPTP has been proposed to be responsible for the mitophagy of depolarized mitochondria in mammalian cells [34]. Here, we suggest that mitophagy may be an adaptive response to eliminate the depolarized mitochondria, which resulted from the mPTP opening induced by α-syn VDAC interaction.

Supplementary Data

Supplementary data are available at ABBS online.

Funding

This work was supported by grants from the National Basic Research Program of China (2011CB504102, 2012CB722407), the National Natural Science Foundation of China (81200995, 30970940), the Natural Science Foundation of Beijing Municipal Education Foundation of Beijing (5102007, 5102012), and the National Natural Science Basic Research Program of China (2011CB504102, 2012CB722407), the National Natural Science Foundation of China (81200995, 30970940), the Natural Science Foundation of Beijing Municipal Education Foundation of Beijing (5102007, 5102012), and the Natural Science Foundation of Beijing Municipal Education Commission (KZ201010025022).

References