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Role of mitophagy regulated by Parkin/DJ-1 in remote ischemic postconditioning-induced mitigation of focal cerebral ischemia-reperfusion injury in rats

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Abstract. – OBJECTIVE: We evaluated the role of mitophagy controlled by Parkin/DJ-1 in remote ischemic post conditioning-induced mitigation of focal cerebral ischemia-reperfusion (I/R) injury in rats.

MATERIALS AND METHODS: Ninety adult male rats were randomly assigned into 5 groups including a sham operation group (S) and ischemia-reperfusion group (I/R). Focal cerebral I/R was induced by right middle cerebral artery occlusion (MCAO). I/R+remote ischemic postconditioning (I/R+RIPoC), I/R+RIPoC+ mitophagy inhibitor Mdivi-1 (I/R+RIPoC+M), and I/R+RIPoC+ normal saline (I/R+RIPoC+NS) groups all received 3 cycles of 10 minutes reperfusion followed by 10 minutes ischemia in bilateral femoral arteries at the beginning of cerebral reperfusion. I/R+RIPoC+M received mitochondrial division inhibitor (Mdivi-1) before ischemia and after 24h of reperfusion, neurological deficit scores (NDSs) were measured and rats were then sacrificed. Brain was removed and size of the infarct was determined. Apoptosis index and LC3-II/I ratio, Parkin/DJ-1 proteins expression, SOD activity, MDA and 15-F2t-Isoprostane content in cerebral ischemic penumbra were studied. Linear correlation between Parkin/DJ-1 proteins expression and LC3-II/I ratio and cerebral infarct size were analyzed.

RESULTS: In experimental groups the NDSs, percentage of cerebral infarct size, apoptosis index, LC3-II/I ratio, MDA and 15-F2t-Isoprostane content significantly increased and Parkin/DJ-1 proteins were up-regulated ($p<0.05$). In I/R+RIPoC and I/R+RIPoC+NS groups, NDSs, percentage of cerebral infarct size, apoptosis index, MDA and 15-F2t-Isoprostane content decreased significantly while LC3-II/I ratio and SOD activity increased compared to I/R group. Parkin/DJ-1 proteins were up-regulated in I/R+RIPoC, I/R+RIPoC+NS and I/R+RIPoC+M groups ($p<0.05$). LC3-II/I ratio and SOD activity significantly decreased ($p<0.05$). Parkin/DJ-1 proteins expression didn't changed in I/R+RIPoC+M group ($p>0.05$). The Parkin/DJ-1 proteins expression were positively correlated with LC3-II/I ratio, and negatively correlated with cerebral infarct size ($p<0.05$).

CONCLUSIONS: Remote Ischemic Post Conditioning (RIPoC) promoted the mitophagy via up-regulation of Parkin/DJ-1 proteins expression and inhibiting the oxidative stress responses, thus mitigating focal cerebral I/R injury in rats.

Key Words:
Parkin, DJ-1, Mitophagy, Reperfusion injury, Brain, Ischemic post conditioning.

Introduction

Cerebral ischemia reperfusion injury may result in severe cerebral dysfunction. Researches[1,2] showed that the limb remote ischemic post conditioning (LRIPoC) can effectively alleviated the focal cerebral ischemia and reperfusion injury in rats. However the mechanism of brain protection is not yet fully understood. Mitochondrial autophagy which is a selective process for the removal of excess or damaged mitochondria[3] plays an important role in regulating the number of mitochondria while maintaining the normal function of the organelle. Prior studies[4] showed that both Parkin and DJ-1 proteins are involved in the regulation of mitochondrial autophagy. Whether the cerebral protection of LRIPoC is related to the Parkin/DJ-1 regulation of mitochondrial autophagy is unknown. Our study intended to evaluate the role of mitophagy controlled by Parkin/DJ-1 in remote ischemic post conditioning-induced mitigation of focal cerebral ischemia-reperfusion (I/R) injury in rats.

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Materials and Methods

Selection and Grouping of the Animals

Ninety adult male Sprague-Dawley rats, weighing 280 to 320 g, were provided by Hubei Center for Disease Control and Prevention. Rats were randomly assigned into 5 groups and each group received 18 rats. Groups were as follows: i- sham operation group (Group S), ii- ischemia-reperfusion group (Group I/R), iii- I/R+remote ischemic post conditioning group (Group I/R+RIPoC), iv- I/R+RIPoC+ mitophagy inhibitor Mdivi-1 group (Group I/R+RIPoC+M), v- I/R+RIPoC+ saline group (group I/R+RIPoC+NS).

Model Preparation and Grouping Process

Focal cerebral ischemia reperfusion model was created by right middle cerebral artery occlusion (MCAO) according to Longa et al[5]. Rats were anaesthetized with 50 mg/kg pentobarbital sodium by intraperitoneal injection. Median incision was performed on the neck. Common carotid artery, internal carotid artery and external carotid artery of right side were isolated, and then the root parts of common carotid artery and external carotid artery were closed with a clamp. The bottom part of the common carotid artery was cut and a nylon wire covered with silicone was inserted at the top into the internal carotid artery. The nylon wire was pushed in about 17-19 mm. A slight resistance signaled that the head of the wire reached in the middle cerebral artery. Then it was ligated at the entrance. During the operation, the rats were kept spontaneous breathing, and the rectal temperature was maintained at 37°C to 37.5°C. After 90 min, the nylon line was extracted and the reperfusion was restored for 24h. Rats in group S were exposed and freed the right carotid artery only. Group I/R+RIPoC, I/R+RIPoC+M and I/R+RIPoC+NS received 3 cycles of 10 min reperfusion followed by 10 min ischemia in bilateral femoral arteries at the beginning of cerebral reperfusion. Mdivi-1 (dissolved in saline solution containing 50% dimethyl sulfoxide – DMSO) was injected to animals in I/R+RIPoC+M group. We injected 3 mg/kg intraperitoneally 5 min before ischemia, and the equal volume of normal saline was injected to the rats in I/R+RIPoC+NS group.

Index Determination

At 24 h of reperfusion, neurological deficit scores (NDSs) were measured[5]. The score zero was given when there was no symptoms for nerve function defect, score one was for those rats unable to extend their left anterior claw, score two for rotation toward the left (for inclining to the left side while walking); and finally four was for the inability to walk accompanied with consciousness disorder.

Cerebral Infarction Volume

After 24 h of reperfusion and neurological function defect score calculation, brains were removed and the size of the infarction was determined in each rat. Intraperitoneal injection of pentobarbital sodium was used and brains were stored at -20°C for 30 min. Then olfactory bulb, cerebellum and lower brainstem were removed. Brain tissue was cut into 6 pieces (2 mm thickness) along the coronal position from the front to the back and then immersed into the TTC solution (Sigma, Saint Louis, MO, USA) for 30 min incubation at 37°C. After color reaction, specimens were fixed in 4% polyformaldehyde for 24h. Photos were taken with digital camera and the percentage of infarct volume was calculated for each sample by Photoshop CS4 image analysis software (the normal brain tissue was pink, and the infarction part was pale blue). For infarct volume correction, we used the method introduced by Swanson et al[6]:

$$\text{the percentage of cerebral infarction volume} = \frac{\text{(contra-lateral normal brain tissue volume-ipsilateral normal brain tissue volume)}}{\text{contralateral normal brain tissue volume}} \times 100\%$$

Detection of Apoptosis by Tunel Assay

After 24h reperfution under anesthesia (by intraperitoneal injection of pentobarbital sodium), we opened the chest and cut the right atrial appendage.

A round needle inserted into the origin aorta via the left ventricle, which was used for perfusion of saline and 4% polyformaldehyde. Samples were fixed with 4% polyformaldehyde for 24h and were dehydrated with 15% sucrose solution for 24h followed by dehydratation in 30% sucrose solution for 2 to 3 times, 24h each. After dehydration, brain tissue samples were embedded with OCT and placed in -80°C for 30 min. TUNEL staining was conducted on frozen sections using the TUNEL Kit protocol (Roche, Mannheim, Germany). The apoptosis of cell nuclei were revealed by brown or dark brown color

(TUNEL positive) while the normal nuclei were blue. Apoptotic cells in the cerebral cortex ischemia half dark area were observed under high magnification (4000X). Each section was selected with five independent positive views and the number of positive cells was recorded. We used the following equation to obtain neurons apoptosis rate.

$$\text{Neurons apoptosis rate} = \frac{\text{apoptotic cells counted in the view}}{\text{all cells counted in the view (average value)}} \times 100\%$$

To obtain the brain tissue of ischemic dark area we employed an improved version of the protocol explained by Ashwal et al[7]. Under anesthesia by intraperitoneal injection of pentobarbital sodium and after 24h of reperfution, brains were removed, placed on ice and sliced for a 6 mm coronal tissue at the position of 3 mm and 9 mm from the frontal lobe. Tissues were then trimmed from the middle structure between the two hemispheres (anterior cerebral artery supply area) approximately 2 mm from both sides along the brain sagittal joint. The remaining left and right sides of the brain tissue were cut at 2 mm to the sagittal section position, which was 30 degrees bevel with the sagittal section. The medial cortex was ischemic penumbra area, which was placed in liquid nitrogen to be measured later. Protein lysis buffer was added and tissues were homogenized. Homogenized tissues were spun and supernatants were collected. Protein concentration was measured using bicinchoninic acid (BCA) method. Protein samples were prepared for SDS-PAGE (they were diluted with loading buffer and boiled for 5 min before being loaded in SDS-PAGE). After SDS-PAGE proteins were transferred to a membrane. Membranes were blocked and incubated with primary antibodies (1:1000) at 4°Covernight. The list of antibodies used in Western blot was as follows: Rabbit anti mouse polyclonal LC3 antibody, Rabbit anti mouse polyclonal Parkin Rabbit anti mouse polyclonal monoclonal DJ-1 antibody and Rabbit anti mouse β-actin antibody (Cell Signaling, Danvers, MA, USA). Membranes were, then, rinsed with phosphate buffered saline and tween (PBST) and incubate with secondary antibody (horseradish peroxidase labeled Goat anti rabbit IgG, 1:5000, Thermo Fisher, Waltham, MA, USA) at room temperature for 1 h. Subsequently developing, fixing, and imaging were conducted. Quantity one software was used to measure the optical density. Protein expression levels were expressed by the ratio of the light density from the target protein band and the density of β-actin. The ratio of LC3-II/I was used for the expression level of the target protein.

Brain ischemia perifocal areas were selected, and 10% tissue homogenate was prepared in low temperature after accurate weighing. The homogenate was spun at 4000 rpm for 10 min at 4°C. The centrifugal radius was 10 cm. The supernatant was tested for superoxide dismutase (SOD) activity, MDA content activity and 15-F_{2t}-Isoprostane content. The list of kits used is as follows: SOD Kit (Nanjing Jiancheng Bioengineering Institute), MDA Kit (Nanjing Jiancheng Bioengineering Institute) and 15-F_{2t}-Isoprostane Kit (Cayman, Ann Arbor, MI, USA).

Statistical Analysis

Data were analyzed using SPSS 17 statistical software (SPSS Inc., Chicago, IL, USA). Measurement data were shown as (±s). Single factor analysis of variance was used in the comparison between the groups. The LC3-II/I ratio and cerebral infarction volume percentage were analyzed for linear correlations with Parkin and DJ-1 proteins expression levels respectively. $p<0.05$ means that the difference was statistically significant.

Results

NDS score, cerebral infarction volume percentage and cell apoptosis index in the cerebral ischemia perifocal areas in experimental groups were higher than group S ($p<0.05$). The NDS score, cerebral infarction volume percentage and cell apoptosis index in the cerebral ischemia perifocal areas of group I/R+RIPoC and group I/R+RIPoC+NS were lower than those in Group I/R ($p<0.05$). NDS score, cerebral infarction volume percentage and cell apoptosis index in the cerebral ischemia perifocal areas in group I/R+RIPoC+M were higher when compared to those in group I/R+RIPoC and group I/R+RIPoC+NS ($p<0.05$). There were no significant differences in those indexes among groups I/R+RIPoC, I/R + RIPoC + NS, I/R and I/R+RIPoC+M (Table I).

Compared with those in Group S, the LC3-II/I ratio in the cerebral ischemia perifocal areas in other groups were higher, and Parkin and DJ-1 proteins expression were up-regulated ($p<0.05$).

Table I. Comparison of NDS score, cerebral infarction volume percentage and cell apoptosis index in the cerebral ischemia perifocal areas of rats in five groups ($\bar{x}$ ±s).

Group	NDS score (point, n=8)	Cerebral infarction volume percentage (%, n=8)	Cell apoptosis index i (%, n=5)
Group S	0	0	2.3 ± 0.8
Group I/R	2.8 ± 0.6[a]	48.0 ± 3.2[a]	54.6 ± 5.2[a]
Group I/R + RIPoC	1.6 ± 0.4[ab]	28.1 ± 3.8[ab]	29.3 ± 3.1[ab]
Group I/R + RIPoC + NS	1.6 ± 0.5[ab]	28.2 ± 3.7[ab]	29.8 ± 3.3[ab]
Group I/R + RIPoC + M	2.5 ± 0.5[acd]	41.2 ± 3.1[acd]	51.2 ± 4.5[acd]

Note: Compared with Group S, [a]$p < 0.05$; Compared with Group I/R, [b]$p < 0.05$; Compared with Group I/R + RIPoC, [c]$p < 0.05$; Compared with Group I/R + RIPoC + NS, [d]$p < 0.05$

Compared to group I/R, the LC3-II/I ratio in the cerebral ischemia perifocal areas in I/R+RIPoC and I/R+RIPoC+NS groups were elevated and levels of Parkin and DJ-1 proteins expression in the cerebral ischemia perifocal areas increased (p<0.05). The LC3-II/I ratio in the cerebral ischemia perifocal areas in group I/R+RIPoC+M decreased more than the I/R+ RIPoC and I/R + RIPoC + NS groups (p<0.05). The differences in Parkin and DJ-1 expression levels were not statistically significant (p>0.05). Differences in LC3-II/I ratio in the cerebral ischemia perifocal areas in I/R and I/R+RIPoC+M groups were not statistically significant (p>0.05). There were no significant differences in the above indexes between I/R+RIPoC and I/R+RIPoC+NS groups (Table II).

MDA and 15-F_{2t}-Isoprostane content in the cerebral ischemia perifocal areas of experimental groups were greater than the group S. SOD activity in the cerebral ischemia perifocal areas decreased in the I/R and I/R+RIPoC+M groups (p<0.05). Compared to I/R group, SOD activity in the cerebral ischemia perifocal areas was higher in I/R+RIPoC and I/R+RIPoC +NS groups, while the MDA and 15-F_{2t}-Isoprostane content in the cerebral ischemia perifocal areas were lower in I/R+RIPoC and I/R+RIPoC+NS groups (p<0.05). The SOD activity in the cerebral ischemia perifocal areas for I/R+RIPoC+M group demonstrated a bigger decline compared to I/R+RIPoC and I/R+RIPoC+NS groups while MDA and 15-F_{2t}-Isoprostane content both increased (p<0.05). There were no significant differences in the above indexes among I/R+ RIPoC, I/R+RIPoC+NS, I/R and I/R+RIPoC+M groups (Table III).

The LC3-II/I ratio was positively correlated with the expression levels of Parkin and DJ-1 proteins in the cerebral ischemia perifocal areas, and the correlation coefficients were 0.7341 and 0.7682 respectively. On the other hand, cerebral infarction volume percentage was negatively correlated with the expression levels of Parkin and DJ-1 proteins in the cerebral ischemia perifocal areas, and the correlation coefficients were -0.8673 and -0.8545 respectively.

Discussion

In this study, the model of focal cerebral ischemia reperfusion injury was prepared using the

Table II. Comparison of LC3-II/I ratio in the cerebral ischemia perifocal areas and expression level of Parkin and of DJ-1 proteins in rats (n=5, $\bar{x}$ ±s).

Group	LC3-II/I	Parkin	DJ-1
Group S	0.13 ± 0.03	0.184 ± 0.016	0.206 ± 0.014
Group I/R	0.32 ± 0.05[a]	0.425 ± 0.013[a]	0.398 ± 0.017[a]
Group I/R + RIPoC	0.53 ± 0.06[a b]	0.723 ± 0.021[a b]	0.629 ± 0.015[a b]
Group I/R + RIPoC + NS	0.48 ± 0.08[a b]	0.731 ± 0.018[a b]	0.634 ± 0.019[a b]
Group I/R + RIPoC + M	0.35 ± 0.06[acd]	0.716 ± 0.015[a b]	0.642 ± 0.023[a b]

Note: Compared with Group S, [a]$p < 0.05$; Compared with Group I/R, [b]$p < 0.05$; Compared with Group I/R + RIPoC, [c]$p < 0.05$; Compared with Group I/R + RIPoC + NS, [d]$p < 0.05$

Table III. Comparison of SOD activity, MDA and 15-F2t-Isoprostane content in the cerebral ischemia perifocal areas of rats in five groups (n=5, $\bar{x}$ ±s).

Group	SOD (U/mg)	MDA (nmol/mg)	15-F_{2t}-isoprostane (mg/g)
Group S	168 ± 19	4.22 ± 0.28	179 ± 86
Group I/R	92 ± 13[a]	8.41 ± 0.42[a]	389 ± 105[a]
Group I/R + RIPoC	162 ± 21[b]	5.14 ± 0.27[ab]	208 ± 89[ab]
Group I/R + RIPoC + NS	165 ± 23[b]	5.26 ± 0.31[ab]	215 ± 85[ab]
Group I/R + RIPoC + M	94 ± 15[acd]	7.93 ± 0.44[acd]	364 ± 103[acd]

Note: Compared with Group S, [a]$p < 0.05$; Compared with Group I/R, [b]$p < 0.05$; Compared with Group I/R + RIPoC, [c]$p < 0.05$; Compared with Group I/R + RIPoC + NS, [d]$p < 0.05$

method described by Ren et al[1], through blocking the right middle cerebral artery for 90 min followed by perfusion for 24h. The results showed that the neurological function deficit score and the volume percentage of cerebral infarction were all higher in I/R group compared to the group S. This indicated that the model was successful. Referring to the method explained by Qi et al[8], remote ischemic postconditioning, namely double femoral artery clamping for 10 min, after reperfusion was implemented and repeated for 3 times. The results indicated that the neurological function deficit score, cerebral infarction volume percentage and apoptosis index of nerve cells in the cerebral ischemia perifocal areas of group I/R+RIPoC were lower compared to I/R group, which meant that remote ischemic postconditioning could induce mitigation of focal cerebral ischemia-reperfusion (I/R) injury in rats.

Oxidative stress is one of the key mechanisms of cerebral ischemia reperfusion injury, and the generation of ROS is related to the dysfunction of the mitochondria. In physiological state, mitochondria can provide energy and substrates for survival of the cells by realizing oxidative phosphorylation. Abnormal accumulation of ROS can cause mitochondrial damage and leads to the release of pro-apoptotic proteins and triggers the apoptosis[9]. In order to maintain the normal state of the cell, damaged or excess mitochondria should be removed via mitochondrial autophagy[10]. In a previously reported study[11] we saw that the inhibition of mitochondrial autophagy *in vivo* and *in vitro* increased the neuronal damage caused by cerebral ischemia and reperfusion. Other authors[12] reported that ischemic preconditioning could selectively increase mitochondrial autophagy to alleviated neuronal damage *in vitro*. The results of our study revealed that SOD activity, MDA and 15-F_{2t}-Isoprostane content, LC3-II/I ratio in the cerebral ischemia perifocal areas in group I/R+RIPoC were higher than group S. In addition, SOD activity in the cerebral ischemia perifocal areas in group I/R+RIPoC+M increased after the administration of Mdivi-1 (a mitochondrial autophagy inhibitor). Also MDA, 15-F_{2t}-Isoprostane content and LC3-II/I ratio decreased, indicating that the remote ischemic postconditioning could inhibit the oxidative stress response by amplifying the level of mitochondrial autophagy, which could then reduce the focal cerebral ischemia reperfusion injury in rats.

Parkin and DJ-1 are two proteins associated with Parkinson's disease, and are commonly used in research in the field of neurodegenerative diseases. The functions of these two proteins were linked to oxidative stress and mitochondrial dysfunction[4]. Previous studies[13] suggested that when the fusion of mitochondria is blocked and the membrane potential is lost, Parkin can selectively be transferred to the damaged mitochondria and mediate the selective removal of the mitochondria. Similar to the Parkin's signaling pathway, DJ-1 regulates the mitochondrial function and the autophagy[14]. DJ-1 deletion can lead to mitochondrial morphology disorder and dysfunction and may cause an increase in ROS produced by the mitochondria[15]. The results of our study showed that the expression levels of Parkin and DJ-1 proteins and the LC3-II/I ratio in group I/R+RIPoC were increased compared to those of I/R group. And the expression levels of Parkin and DJ-1 proteins were positively correlated with LC3-II/I ratio, but negatively correlated with the volume of cerebral infarction. This indicated that the remote ischemic post conditioning reduced the focal cerebral ischemia reperfusion injury by increasing Parkin and DJ-1 proteins expression levels in rats and promoted mitochondrial au-

tophagy. In this paper, due to the lack of Parkin, DJ-1 protein agonists and antagonists, we used the linear correlation analysis. Further research is needed by in order to determine how Parkin and DJ-1 proteins play a neuroprotective role in mitochondrial autophagy. This knowledge will help us in our way to discovery of better methods to treat remote ischemic post conditioning.

Conclusions

We think that remote ischemic post conditioning may increase the level of mitochondrial autophagy and inhibit the oxidative stress reaction in rats, so as to mitigate the focal cerebral ischemia and reperfusion injury damage by up-regulating Parkin and DJ-1 protein expression.

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Conflict of Interest

The Authors declare that they have no conflict of interests.

References

1) Ren C, Yan Z, Wei D, Gao X, Chen X, Zhao H. Limb remote ischemic postconditioning protects against focal ischemia in rats. Brain Res 2009; 1288: 88-94.
2) Liu Q, Zhou S, Wang Y, Qi F, Song Y, Long S. A feasible strategy for focal cerebral ischemia-reperfusion injury: remote ischemic postconditioning. Neural Regen Res 2014; 9: 1460-1463.
3) Fimia GM, Kroemer G, Piacentini M. Molecular mechanisms of selective autophagy. Cell Death Differ 2013; 20: 1-2.
4) Trempe JF, Fon EA. Structure and function of Parkin, PINK1, and DJ-1, the three musketeers of neuroprotection. Front Neurol 2013; 4: 38.
5) Longa EZ, Weinstein PR, Carlson S, Cummins R. Reversible middle cerebral artery occlusion without craniectomy in rats. Stroke 1989; 20: 84-91.
6) Swanson RA, Morton MT, Tsao-Wu G, Savalos RA, Davidson C, Sharp FR. A semiautomated method for measuring brain infarct volume. J Cereb Blood Flow Metab 1990; 10: 290-293.
7) Ashwal S, Tone B, Tian HR, Cole DJ, Pearce WJ. Core and penumbral nitric oxide synthase activity during cerebral ischemia and reperfusion. Stroke 1998; 29: 1037-1046.
8) Qi ZF, Luo YM, Liu XR, Wang RL, Zhao HP, Yan F, Song ZJ, Luo M, Ji XM. AKT/GSK3β-dependent autophagy contributes to the neuroprotection of limb remote ischemic postconditioning in the transient cerebral ischemic rat model. CNS Neurosci Ther 2012; 18: 965-973.
9) Wang CH, Wu SB, Wu YT, Wei YH. Oxidative stress response elicited by mitochondrial dysfunction: implication in the pathophysiology of aging. Exp Biol Med (Maywood) 2013; 238: 450-460.
10) Lemasters JJ. Selective mitochondrial autophagy, or mitophagy, as a targeted defense against oxidative stress, mitochondrial dysfunction, and aging. Rejuvenation Res 2005; 8: 3-5.
11) Zhang X, Yan H, Yuan Y, Gao J, Shen Z, Cheng Y, Shen Y, Wang RR, Wang X, Hu WW, Wang G, Chen Z. Cerebral ischemia-reperfusion-induced autophagy protects against neuronal injury by mitochondrial clearance. Autophagy 2013; 9: 1321-1333.
12) Brown JE, Zeiger SL, Hettinger JC, Brooks JD, Holt B, Morrow JD, Musiek ES, Milne G, McLaughlin B. Essential role of the redox-sensitive kinase p66shc in determining energetic and oxidative status and cell fate in neuronal preconditioning. J Neurosci 2010; 30: 5242-5252.
13) Narendra D, Tanaka A, Suen DF, Youle RJ. Parkin is recruited selectively to impaired mitochondria and promotes their autophagy. J Cell Biol 2008; 183: 795-803.
14) Thomas KJ, McCoy MK, Blackinton J, Beilina A, van der Brug M, Sandebring A, Miller D, Maric D, Cedazo-Minguez A, Cookson MR. DJ-1 acts in parallel to the PINK1/parkin pathway to control mitochondrial function and autophagy. Hum Mol Genet 2011; 20: 40-50.
15) Irrcher I, Aleyasin H, Seifert EL, Hewitt SJ, Chhabra S, Phillips M, Lutz AK, Rousseaux MW, Bevilacqua L, Jahani-Asl A, Callaghan S, MacLaurin JG, Winklhofer KF, Rizzu P, Rippstein P, Kim RH, Chen CX, Fon EA, Slack RS, Harper ME, McBride HM, Mak TW, Park DS. Loss of the Parkinson's disease-linked gene DJ-1 perturbs mitochondrial dynamics. Hum Mol Genet 2010; 19: 3734-3746.