UNDERSTANDING THE ROLE OF LEUCINE-RICH REPEAT KINASE 2, PTEN-INDUCED PUTATIVE KINASE 1, AND PARKIN IN PD

Ryosuke Takahashi, MD, Kyoto University Hospital, Kyoto, Japan, discussed recent publications that have enhanced understanding of the role of leucine-rich repeat kinase 2 (LRRK2), PTEN-induced putative kinase 1 (PINK1), and parkin as genetic risk factors in sporadic PD.

In the first study, Imai and colleagues [*EMBO J* 2008] proposed that mutations in LRRK2 cause dopaminergic degeneration in patients with PD through phosphorylation of eukaryotic initiation factor 4E–binding protein (4E-BP). Chronic inactivation of 4E-BP by LRRK2 with pathogenic mutations deregulates protein translation, eventually resulting in age-dependent loss of dopaminergic neurons.

More recently, Martin and colleagues [*Cell* 2014] suggested that the phosphosubstrate that connects LRRK2 kinase activity to neurodegeneration is ribosomal protein s15. In that study, ribosomal protein s15 phosphorylation by LRRK2 was shown to stimulate both cap-dependent and internal ribosome entry site–mediated protein translation and, through an as yet unknown mechanism, to lead to neurodegeneration. Phosphomutant s15 rescued neurodegeneration in LRRK2 G2019S transgenic *Drosophila*.

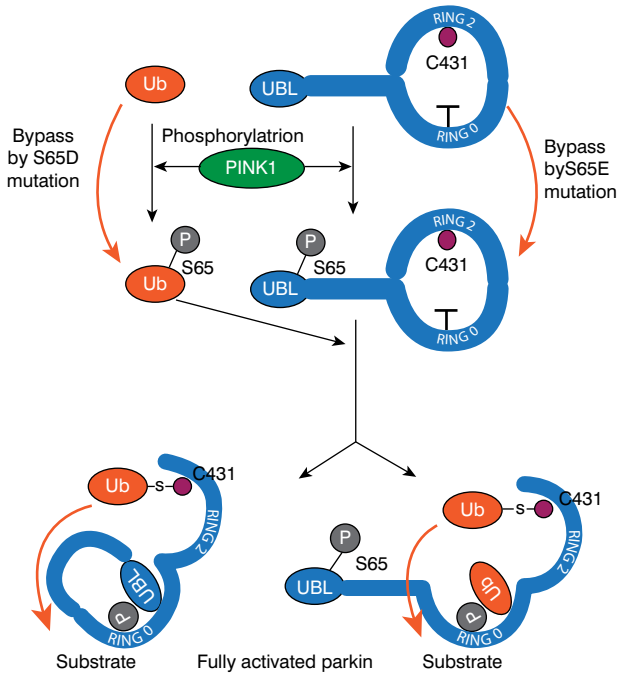
Prof. Takahashi concluded that in a subset of LRRK2 mutation–related parkinsonism, translational dysregulation leading to overproduction of proteins may be causative and that a future therapeutic option for PD may be found in global translational repression.

In the second part of his presentation, Prof. Takahashi turned his attention to PINK1 and parkin, which are known to be responsible for the autosomal-recessive young-onset form of familial PD and essential for the selective elimination of damaged mitochondrion through the autophagy-lysosome pathway.

PINK1-dependent S65 phosphorylation is important for the activation of parkin [Shiba-Fukushima K et al. *Sci Rep* 2012]; however, it can only partially activate it. To fully activate parkin, an additional substrate is necessary. Ubiquitin is also phosphorylated by PINK1, and this phosphorylated ubiquitin is essential for parkin activation [Matsuda N et al. *J Cell Biol* 2010]. Koyano and colleagues [*Nature* 2014] have proposed a model for parkin activation that is based on interaction between ubiquitin and parkin by which PINK1-dependent phosphorylation of both is sufficient for full activation of parkin E3 activity (Figure 2).

In summary, noted Prof. Takahashi, PINK1 phosphorylates both ubiquitin and parkin, thus allowing phosphorylated ubiquitin to activate parkin by binding to and changing the conformation of phosphorylated parkin. Therefore, an excellent biomarker for mitochondrial damage may be phosphorylated ubiquitin.

Figure 2. A Model for Parkin Activation



P=parkin; PINK1=PTEN-induced putative kinase 1; Ub=ubiquitin.

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UPR Activation Evaluated in Lewy Body Dementias and AD

Written by Toni Rizzo

Even though they have distinct clinical, pathologic, and biochemical features, Parkinson's disease (PD), Alzheimer's disease (AD), Huntington's disease, and less commonly prion disease are all characterized by misfolded protein aggregation in the brain and fatal neuronal loss. The unfolded protein response (UPR) is emerging as a common player in all neurodegenerative diseases. Misfolded proteins in the endoplasmic reticulum (ER) trigger the dissociation of the ER chaperone GRP78/BiP from protein kinase ribonucleic acid–like ER kinase and other proteins, activating the UPR stress response. Initially the UPR is neuroprotective, but prolonged activation and failure to restore protein homeostasis can lead to neurodegeneration.

Evidence of UPR activation in both AD and PD suggests that the neurons are subjected to ER stress, but no studies have investigated the UPR in patients who have dementia with PD and AD pathology. The aim of this study, presented by Jean-Ha Baek, PhD, Karolinska Institute, Huddinge, Sweden, was to investigate changes in the UPR pathway in deceased subjects with PD with dementia (PDD) and dementia with Lewy bodies (DLB) in comparison with patients with AD and control subjects.

Postmortem brain tissue from 4 brain regions was provided by Brains for Dementia Research, United Kingdom, for evaluation (Table 1).

Table 1. Number and Type of Brain Tissue Samples

	Number of Cases			
Brain Region	AD	DLB	PDD	Control
Prefrontal cortex (BA 9)	11	44	28	16
Temporal cortex (BA 21)	0	35	33	25
Cingulate gyrus (BA 24)	16	40	10	23
Parietal cortex (BA 40)	15	47	33	23

AD=Alzheimer's disease; BA=Brodmann area; DLB=dementia with Lewy bodies; PDD=Parkinson's disease with dementia.

GRP78/BiP protein levels were quantified with western blot to measure the degree of UPR activation. Binding immunoglobulin protein (GRP78/BiP) expression patterns were assessed with immunohistochemistry.

Brain tissue from the cingulate gyrus of patients with PDD and those with DLB had a significant increase in the level of GRP78/BiP protein compared with tissue from patients with AD and control subjects (p=0.000 for all comparisons). GRP78/BiP protein expression in the parietal cortex was significantly decreased in brain tissue from patients with AD compared with control subjects (p=0.000) and significantly increased in tissue from patients with PDD compared with patients with AD (p=0.002) and in patients with DLB compared with those with AD (p=0.001). No significant differences were observed in GRP78/BiP levels in the prefrontal cortex and temporal cortex between any of the brain tissue sample groups.

Immunohistochemical studies detected GRP78/BiP protein in the cytoplasm of prefrontal cortex neurons in brain tissue from patients with AD, PDD, and DLB and controls. Although the labeling intensity was slightly increased in AD, PDD, and DLB patient samples compared with controls, the intensity was similar across the different disease groups.

This was the first study to investigate changes in the UPR pathway in deceased subjects with PDD and DLB in comparison with patients with AD and control subjects. Pearson correlation analysis found no association between changes in the GRP78/BiP protein level and Mini-Mental State Examination scores before death, rate of decline in Mini-Mental State Examination score, or global and regional pathologic scores for α-synuclein, plaques, and tangles. There was no correlation of Hoehn and Yahr scores with GRP78/BiP levels in a subset of subjects with PDD.

DBS Improves FOG in Patients With PD

Written by Toni Rizzo

High-frequency deep brain stimulation (DBS) is effective for relief of Parkinson's disease (PD) symptoms. However, studies on the effect of subthalamic nucleus DBS (STN-DBS) on freezing of gait (FOG) have reported inconsistent results, with some suggesting that STN-DBS alleviates FOG [Davis JT et al. *Clin Neurol Neurosurg* 2006] and others reporting worsening of gait and balance [van Neunen BFL et al. *Mov Disord* 2008; Krack P et al. *N Engl J Med* 2003].

The objective of the Vercise Implantable Stimulator for Treating Parkinson's Disease trial [VANTAGE; NCT01221948], presented by Michael T. Barbe, MD, University Hospital, Cologne, Germany, was to evaluate motor function improvement in patients with moderate to severe Parkinson's disease following treatment with bilateral STN-DBS. Prospective and nonrandomized, VANTAGE is open-label interventional trial in which a total of 40 patients underwent STN-DBS. DBS was delivered with the new Vercise DBS System, a multiple-source, eight-contact, constant-current system that is implantable and rechargeable.

FOG was assessed before and after surgery with Unified Parkinson's Disease Rating Scale (UPDRS) II Item 14, the Freezing of Gait Questionnaire (FOGQ), and a videotaped walk test known to provoke FOG (with and without dual tasks). For the postsurgery tests, FOGQ was given at 26 weeks and the walking test at 12, 26, and 52 weeks after implantation. The patients were also assessed with the Core Assessment Program for Surgical Interventional Therapies motor tests, Tremor Rating Scale, Dyskinesia Rating Scale, Parkinson's Disease Questionnaire 39, Short Form 36 Health Survey, and the Schwab and England Activities of Daily Living Scale. Resource utilization was assessed and patient motor diaries collected over 3 days at all subsequent visits.

The cohort was 67.5% male and 32.5% female, with a mean age of 60.2 years. At baseline, 26 of the 38 patients