MBP-TcPINK1

Cat. # UB401

Background: PTEN-induced putative kinase protein 1 (Serine/Threonine kinase PINK1) is an important protein that saves the dysfunction of mitochondria upon application of cellular stress. When the mitochondria is depolarized, PINK stabilizes and gets accumulated. When E3 Ubiquitin Ligase Parkin (PARK2) is brought to the damaged mitochondria, it is activated by PINK1 by phosphorylation at Ser 65 which in turn interacts with ubiquitin that is also phosphorylated at Ser 65. This process is important for removing the damaged mitochondria by mitophagy (selective autophagy) by mediating activation and translocation of PARK2. Recombinant human PINK1 effectively phosphorylates recombinant Parkin, mono-Ubiquitin, and poly-Ubiquitin chains. It specifically phosphorylates both Parkin and Ubiquitin at serine 65. This recombinant protein contains an MBP tag.

Application:

- For phosphorylating Ubiquitin, Ubiquitin chains and Parkin at Serine 65 residue.
- For activating Parkin E3 ligase in in vitro assays

Product Information

Affinity tag:	MBP
Purity:	≥90% by SDS-PAGE
Molecular Weight:	106 kDa
Physical State:	liquid
Quantity:	50 µg
Species:	*Tribolium castaneum*
Storage:	-80° C. Avoid repeated freeze/thaw cycles

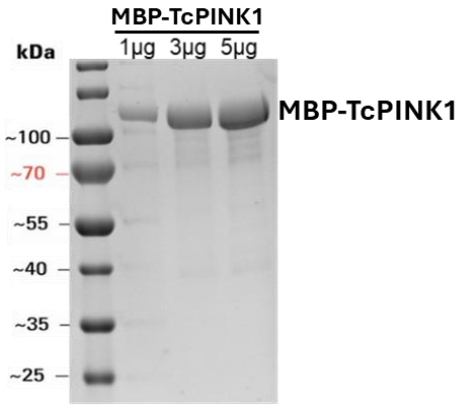

Figure 1. Commassie stained gel of MBP-TcPINK1. Purity is >90%.

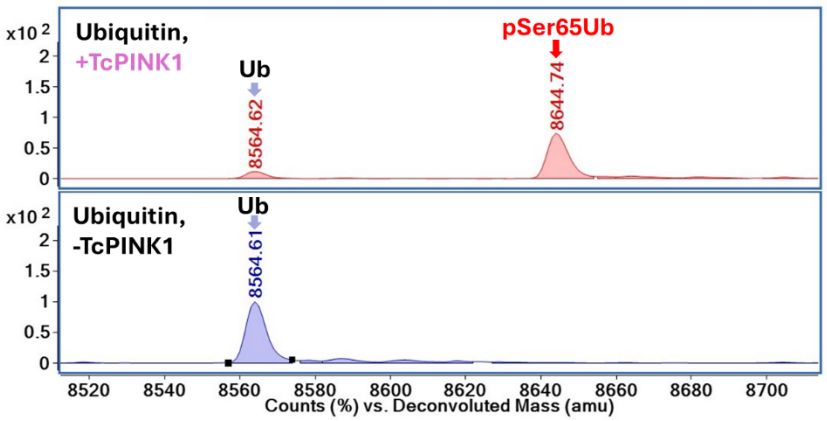

Figure 2. MBP-TcPINK1 efficiently phosphorylate Ser65 on Ubiquitin. Ubiquitin was incubated with 1µM MBP-TcPINK1 (top trace) or without MBP-TcPINK1 (bottom trace) in presence of 5mM ATP and samples were analyzed by LC-MS/MS. pSer65Ub was also confirmed by WB using anti-pSer65 Ub antibody.

References

1. Ge, P., Dawson, V. L., & Dawson, T. M. (2020). PINK1 and Parkin mitochondrial quality control: A source of regional vulnerability in Parkinson's disease. *Molecular neurodegeneration*, *15*(1), 1-18.
2. Kumar, A., Tamjar, J., Waddell, A. D., Woodroof, H. I., Raimi, O. G., Shaw, A. M., ... & van Aalten, D. M. (2017). Structure of PINK1 and mechanisms of Parkinson's disease-associated mutations. *Elife*, *6*, e29985.
3. Quinn, P. M., Moreira, P. I., Ambrósio, A. F., & Alves, C. H. (2020). PINK1/PARKIN signalling in neurodegeneration and neuroinflammation. *Acta Neuropathologica Communications*, *8*(1), 1-20.

Figure 1. Image shows an *in vitro* parkin activation assay with three conditions: parkin + PINK1 + pUb, parkin + PINK1, and parkin. As shown through the lengths and intensities of the ubiquitination smears, the PINK1 and pUb activate parkin and allow it to ubiquitinate at a higher rate.