Mitochondrial intracristal space (ICS) proteome was revealed by mass detection of isotope-coded post-translational modification by APEX

Myeong-Gyun Kang,†[a] Sanghee Shin,†[b,f,g] Song-Yi Lee,[a,h] Ohyeon Kwon,[c] Pratyush Kumar Mishra,[a] Minkyo Jung,[d] Ji Young Mun,[d] Jung-Min Kee,*[c] Jong-Seo Kim,*[b,e] Hyun-Woo Rhee*[a,b]

[a]*Department of Chemistry, Seoul National University, Seoul 08826, Korea*
[b]*School of Biological Sciences, Seoul National University, Seoul 08826, Korea*
[c]*Department of Chemistry, Ulsan National Institute of Science and Technology (UNIST), Ulsan 44919, Korea*
[d]*Neural Circuit Research Group, Korea Brain Research Institute, Daegu 41062, Republic of Korea*
[e]*Center for RNA Research, Institute of Basic Science, Seoul 08826, Korea*
[f]*The Research Institute of Basic Science, Seoul National University, Seoul 08826, Korea*
[g]*Current address: Department of Cancer Biology, Dana-Farber Cancer Institute, Boston, MA, USA*
[h]*Current address: Departments of Biology, Genetics, and Chemistry, Stanford University, Stanford, CA, USA*
†*These authors equally contributed to this work.*
*Corresponding authors.
E-mail: mg132@snu.ac.kr

The intracristal space (ICS) is a distinct sub-domain of the mitochondrial intermembrane space (IMS) where oxidative phosphorylation occurs[1]. This place is expected to control dynamic demands to regulate mitochondrial respiration, however, the molecular components of the ICS have not been identified owing to the lack of an effective detection method. Since there is no known targeting sequence or protein for proximity labeling in this space, we developed isotope-coded desthiobiotin-phenol probes that enable differential mass analysis of post-translocated modifications[2] by IMS-APEX2 and outer mitochondrial membrane (OMM)-APEX2. From this unique proximity labeling approach dubbed iSpot-ID, we identified TMEM177 is exclusively localized in the ICS and TMEM177 also offers a unique ICS targeting modality of various fluorescent sensor proteins for measuring local pH, redox states, and local temperature in the ICS under the uncoupling process. We also obtained an ICS proteome using TMEM177-APEX2 and the results showed that a surprisingly large portion of mitochondrial matrix targeting proteins and several known IMS localized proteases were found in this place. Since most of those matrix proteins (e.g., HSPD1, HSPE1) are self-oligomerized proteins, our data revealed that ICS is an important place to control import and homeostasis of those specific mitochondrial proteins under the dynamic metabolic demands.

References

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