The MAVS and MAV-Nots: PINK1 Clears Prion-like MAVS Aggregates to Extinguish Mitochondrial Inflammatory Signaling

Traditionally considered the "powerhouses of the cell," mitochondria have recently enjoyed a renaissance as hubs for innate immune signaling (1). MAVS (mitochondrial antiviral-signaling protein), a nuclear-encoded adaptor that is anchored to the mitochondrial outer membrane (MOM), interacts with both the RLRs (double-stranded RNA-sensing RIG-I–like receptors) and NLRP3 (NACHT, LRR, and PYD domain-containing protein 3) and is required for viral double-stranded RNA induction of cytokines and for NLRP3 inflammasome-mediated processing of IL-1β (Figure 1) (2). The function of the MAVS "signalosome" is dependent on localization to the MOM, and its activation involves prion-like aggregation of MAVS monomers and a variety of posttranslational modifications (2, 3). Remarkably, prion-like MAVS aggregates released into the extracellular space can be taken up by other cells, dock on mitochondria in a manner dependent on native MAVS, and catalyze further MAVS aggregation and IFN-β production in a self-perpetuating manner (4). Recent findings that MAVS-null mice are protected from bleomycin-induced lung fibrosis (5) have suggested that MAVS activation may also be provoked by host-derived ("damage-associated") signals and that further work is required to identify nonmicrobial MAVS triggers and to better understand how activated MAVS aggregates are cleared.

Reports to date have suggested that MAVS aggregates are degraded by the ubiquitin–proteasome system (2) and autophagy (6, 7). Autophagy receptors and adaptors such as Nix and NDP52, which interact with microtubule-associated protein 1A/1B-LC3 (light chain 3)-positive autophagosomes, keep RLR-induced IFN-β in check by promoting MAVS degradation (2, 6). MAVS itself also interacts directly with LC3, supporting degradative mitophagy (6, 7). The importance of mitophagy in resolving MAVS signaling is underscored by the observation that many viruses exploit mitophagy to dampen antiviral signaling (8). PINK1 (PTEN-induced kinase 1) and the ubiquitin ligase parkin interact at the MOM in perhaps the best-described mitophagy-initiating pathway (9). Although PINK1 has not been studied in relation to MAVS clearance, reports of increased MAVS aggregates in the lungs of bleomycin animal models and patients with idiopathic pulmonary fibrosis (5), taken together with reports that *Pink1*$^{-/-}$ mice have increased inflammation-induced fibrosis in kidney (10) and lungs (11), have indirectly suggested that PINK1 may possibly regulate MAVS.

In this issue of the *Journal*, Kim and colleagues (pp. 592–603) explore nonmicrobial triggers of MAVS aggregation and also confirm an inhibitory effect of PINK1 on MAVS *in vitro* and *in vivo* (12). The authors show that select mitochondrial toxicants (CCCP, valinomycin, and nigericin) induce MAVS aggregation in cultured cells (Figure 1), whereas others (oligomycin A and rotenone) do not. Profiling the effects of these compounds on mitochondrial functions, they show that MAVS aggregation is dissociable from mitochondrial reactive oxygen species generation and mitochondrial membrane depolarization but tracks with agents that reduce mitochondrial pH and thus proton-motive force. They also show that the MAVS-dependent RLR ligand poly(I:C) induces mitochondrial membrane depolarization, thereby raising the interesting possibility that MAVS may "sense" mitochondrial perturbations and that this may be a final common pathway to MAVS aggregation. The authors go on to show that PINK1-null cells have increased MAVS aggregates in response to either nigericin or poly(I:C) and delayed aggregate clearance. In parallel, PINK1-null cells mount increased induction of IFN-β in response to poly(I:C) and increased NLRP3 inflammasome activation and IL-1β processing in response to nigericin. Using co-IP and biomolecular fluorescence complementation, they provide evidence that poly(I:C) and nigericin may induce a direct interaction between PINK1 and MAVS. Finally, demonstrating the *in vivo* relevance of PINK1-dependent suppression of MAVS, they find that influenza A–infected *Pink1*$^{-/-}$ mice have elevated induction of IFN-β and IFN-stimulated genes in their lungs in a manner that is abolished by MAVS deletion. *Pink1*$^{-/-}$ mice are also shown to display increased MAVS aggregates and MAVS-dependent fibrosis in their lungs after bleomycin.

Like all interesting reports, the paper by Kim and colleagues raises several new questions. With regard to the finding that mitotoxicants induce MAVS aggregation, we are left wondering what the precise biochemistry is at the level of the MOM that induces MAVS polymerization and whether this can be intervened on pharmacologically. One also wonders whether mitotoxicants, such as nigericin, that cause MAVS aggregation are sufficient to induce IFN-β, especially in PINK1-null cells. In that regard, it would also be interesting to test whether nigericin-induced MAVS aggregation is RLR dependent, such as might be the case if it induces mitochondrial RNA soiling of the cytosol. A prior report that efficient RLR signaling requires mitochondrial oxidative phosphorylation (13) may suggest that mitotoxicants are unlikely to induce IFN-β, perhaps instead effectively uncoupling MAVS aggregation from IFN-β induction. As a variety of posttranslational modifications of MAVS have already been cataloged, it will be important to define which of these, if any, are induced by agents such as nigericin.

With regard to the PINK1 effect, future studies will be required to define whether MAVS, which is already known to be a substrate

Supported by the U.S. National Institutes of Health, National Institute of Environmental Health Sciences (Z01 ES102005).

Originally Published in Press as DOI: 10.1165/rcmb.2021-0055ED on March 9, 2021

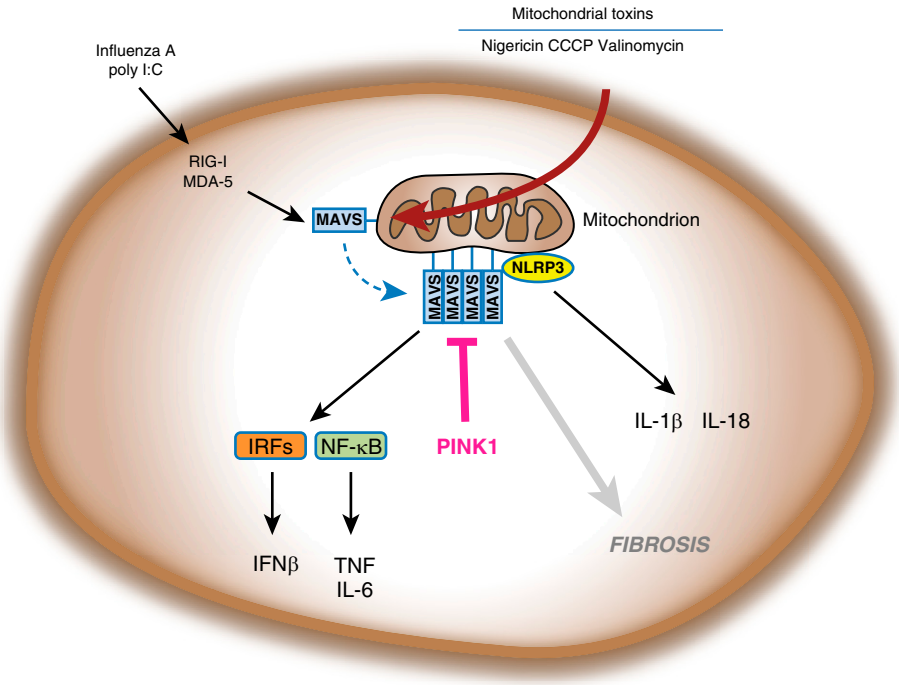

Figure 1. PINK1 (PTEN-induced kinase 1) promotes clearance of proinflammatory MAVS (mitochondrial antiviral-signaling protein) aggregates. MAVS is an adaptor protein localized to the mitochondrial outer membrane. In response to activation of the pathogen recognition receptors RIG-I (retinoic acid-inducible gene I) or MDA-5 (melanoma differentiation-associated protein 5) by viral double-stranded RNA, MAVS forms prion-like aggregates that trigger downstream signaling to induce IFN-β, proinflammatory cytokines (TNF and IL-6), and NLRP3 (NACHT, LRR, and PYD domain-containing protein 3) inflammasome–dependent IL-1β and IL-18. MAVS also reportedly promotes fibrosis in the bleomycin-exposed lung. In this issue of the *Journal*, Kim and colleagues report that mitochondrial toxins such as nigericin also induce MAVS aggregate formation and that PINK1 promotes clearance of MAVS aggregates and suppresses MAVS-dependent cytokine induction and lung fibrosis. CCCP = carbonyl cyanide m-chlorophenyl hydrazone; IRF = IFN regulatory factor; NF = nuclear factor.

for other kinases (3), is a direct or indirect phosphorylation target for PINK1. If so, this may open the possibility for PINK1 inhibitors to manipulate MAVS *in vivo*. It will also be important to confirm whether PINK1-dependent MAVS aggregate clearance is autophagy dependent. As some ostensibly PINK1/parkin-independent pathways to autophagic degradation of MAVS and mitochondria have been identified (14), it seems likely that PINK1 may only regulate MAVS in response to select physiologic challenges. Finally, the *in vivo* lung challenges performed by the authors leave several issues unaddressed, including the effects of PINK1 deletion on viral clearance, lung injury, and animal survival in the influenza A model and the precise mechanism by which MAVS exacerbates fibrosis in the bleomycin model.

Given that mitochondrial dysfunction has been implicated in the pathogenesis of several lung exposures and diseases (15) and aging (11), it will now be important to define whether MAVS supports a broader range of lung disease and aging phenotypes and whether PINK1 mitigates these. PINK1 expression in the lung reportedly declines with aging (11). It will be important to study MAVS aggregation in specific cell types of fibrotic lungs such as alveolar macrophages and alveolar epithelial cells, where mitochondrial abnormalities have been reported (11). Prior reports that MAVS aggregation can itself impair mitochondrial function and trigger mitophagy (7, 14) raise the interesting possibilities that MAVS aggregation may either feed forward to further disable mitochondria, or, alternatively, be adaptive by driving clearance of damaged mitochondria. Either way, one wonders whether MAVS aggregates—in airway cells, peripheral blood leukocytes, or plasma—might prove useful as lung disease biomarkers. Finally, because MAVS localizes at endoplasmic reticulum–mitochondria contact sites (2), it will be interesting to determine whether MAVS aggregation and endoplasmic reticulum stress communicate during fibrosis. Positioned at such a critical crossroads, one MAVS makes for many possibilities. ■

Author disclosures are available with the text of this article at www.atsjournals.org.

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