Autophagy

Autophagy

ISSN: 1554-8627 (Print) 1554-8635 (Online) Journal homepage: http://www.tandfonline.com/loi/kaup20

Autophagy inhibition and mitochondrial remodeling join forces to amplify apoptosis in activation-induced cell death

Mauro Corrado & Silvia Campello

To cite this article: Mauro Corrado & Silvia Campello (2016) Autophagy inhibition and mitochondrial remodeling join forces to amplify apoptosis in activation-induced cell death, Autophagy, 12:12, 2496-2497, DOI: 10.1080/15548627.2016.1226738

To link to this article: http://dx.doi.org/10.1080/15548627.2016.1226738

Accepted author version posted online: 14 Sep 2016.
Published online: 14 Sep 2016.

AUTOPHAGY
2016, VOL. 12, NO. 12, 2496–2497
http://dx.doi.org/10.1080/15548627.2016.1226738

Taylor & Francis
Taylor & Francis Group

AUTOPHAGIC PUNCTUM

Autophagy inhibition and mitochondrial remodeling join forces to amplify apoptosis in activation-induced cell death

Mauro Corrado[a,b] and Silvia Campello [b,c]

[a]Dulbecco-Telethon Institute, Venetian Institute of Molecular Medicine, Padova, Italy; [b]IRCCS Fondazione Santa Lucia, Rome, Italy; [c]Department of Biology, University of Rome Tor Vergata, Rome, Italy

ABSTRACT
Mitochondrial structural and functional changes and the autophagy pathway crosstalk under several stress conditions. However, their interplay under physiological cell death stimulation has been unclear. In our recent report, we show that during activation-induced cell death (AICD), the T-cell receptor (TCR)-dependent pathway that controls immune tolerance, autophagy is inhibited at an early stage. Further, we found that this inhibition is coupled with mitochondria fragmentation and cristae remodeling to unleash the apoptotic program. Last, we dissected the role of macroautophagy/autophagy versus mitophagy in the context of this physiological cell death, and bulk autophagy turned out to be able to remove dysfunctional and depolarized mitochondria. Our data suggest new possible approaches to modulate the immune function in the context of autoimmunity or immunotherapy.

ARTICLE HISTORY
Received 8 August 2016
Revised 16 August 2016
Accepted 17 August 2016

KEYWORDS
AICD; apoptosis; autophagy; mitochondria; mitochondrial dynamics; T cells

A peculiar balance exists between life and death of T lymphocytes by which an effective immune response takes the stage, while at the same time autoimmunity must be avoided. Activation-induced cell death is active in every stage of a T cell's life: within the thymus, to establish immune tolerance by negative selection of autoreactive lymphocytes during development, as well as after the resolution of an inflammation, to remove expanded T cells and promote peripheral tolerance. Mechanistically, a second stimulation of the TCR, in the absence of the appropriate prosurvival costimulation (this meaning that the inflammation has been resolved), represents the physiological signal for AICD, to kill the now unnecessary T cells and shutdown the immune response.

The function of mitochondria in T lymphocytes is highly regulated and linked to their activity. First, during thymocyte development, the mitochondrial content in the cell is progressively reduced until the T cell is mature and leaves the thymus. Then, when T cells are activated, mitochondria relocate at the immunological synapse to buffer calcium entry and to avoid calcium-mediated T cell inhibition. Third, T lymphocyte chemotaxis is also regulated by mitochondrial shape changes. However, little attention has been paid so far to the influence of mitochondrial morphology on T cell AICD. Indeed, in our report, we observed that mitochondria fragment after TCR re-engagement and their cristae remodel before the onset of cell death, to allow CYCS/cytochrome *c* release from mitochondria. At the same time, mitochondria function also becomes impaired, with a clear loss of their membrane potential that triggers PARK2/PARKIN recruitment. Of note, mitochondrial fragmentation and dysfunction usually mark the organelles for degradation through a selective mechanism termed mitophagy, this mainly depending on PINK1 stabilization on the mitochondrial outer membrane, and the consequent recruitment of the ubiquitin ligase PARK2. Indeed, several lines of evidence exist on the interplay between apoptosis and autophagy, and removal of damaged mitochondria has been proven to be cytoprotective in many pathological conditions. Despite this observation, it has been controversial whether mitochondria are actually removed in the context of a cell committed to apoptosis.

Interestingly, mitochondrial morphology behaves differently when mitophagy, or autophagy, are induced. Dysfunctional mitochondria fragment until they are sterically small enough to be engulfed into autophagosomes in order to be removed from cells. By contrast, when cells undergo a phase of limited nutrient availability and autophagy is induced, mitochondria elongate to resist autophagosome engulfment and sustain cell viability by increased ATP production.

In our work, we showed that modulation of autophagy is, indeed, crucially involved in AICD. In more detail, we found that during AICD autophagy is inhibited through a mechanism impinging on the upstream kinase AMPK. We observed, indeed, that both AMPK and LC3 activity is reduced upon AICD induction in a PRKA/PKA-dependent manner and, consequently, other autophagic proteins downstream of AMPK, such as ULK1 and ATG13, are inhibited. We can thus explain, by this way, the apparent paradox by which dysfunctional PARK2-decorated mitochondria are not cleared from the cell.

CONTACT Silvia Campello silvia.campello@uniroma2.it Department of Biology, University of Rome Tor Vergata Via della Ricerca Scientifica 00133 Rome, Italy.
Punctum to: M. Corrado, F.R. Mariotti, L. Trapani, L. Taraborrelli, F. Nazio, V. Cianfanelli, M.E. Soriano, E. Schrepfer, F. Cecconi, L. Scorrano, S. Campello (2016) Macroautophagy inhibition maintains fragmented mitochondria to foster T cell receptor-dependent apoptosis. The EMBO Journal 2016; 35:1793-1809; http://dx.doi.org/10.15252/embj.201593727

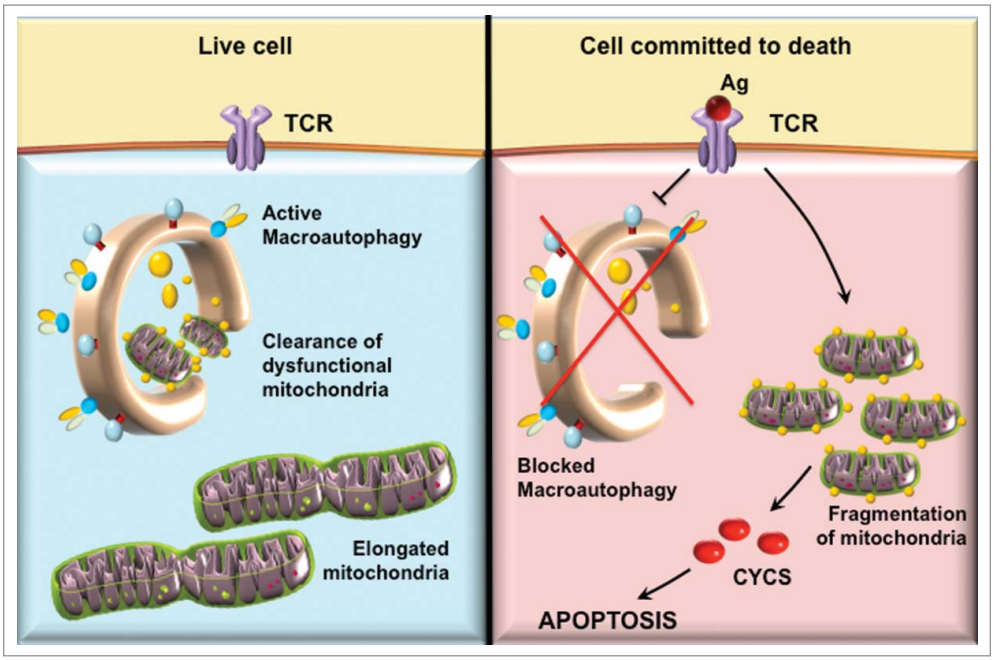

Figure 1. In live cells active autophagy removes dysfunctional PARK2 (yellow small balls)-decorated mitochondria, while healthy organelles maintain elongated morphology. In a cell committed to apoptosis, autophagy is inhibited. This blockage results, in turn, in the accumulation of damaged fragmented mitochondria, which release pro-apoptotic molecules unleashing the intrinsic cell death program. Ag, antigen; TCR, T-cell receptor.

Such an autophagy block allows in fact the accumulation of damaged organelles and apoptosis progression (Fig. 1). It is noteworthy that pharmacological reactivation of general autophagy is able in our experimental setups to restore a "normal" threshold level of mitochondrial morphology and function in order to reduce apoptosis. Thus, we could conclude that it is the autophagy program, rather than selective mitophagy, that is the mechanism that protects T cells from cell death by removing damaged mitochondria. In support of our hypothesis, we showed that T cells from mouse models in which autophagy was genetically ablated (*Ambra1*$^{gt/+}$ and Cre-infected *atg7*$^{fl/fl}$ T lymphocytes) are more sensitive to AICD.

Our report highlights, based on a number of molecular fine controls, the involvement of mitochondria and autophagy regulation in a physiological model of apoptosis (Fig. 1) that affects the extent and the functionality of the adaptive immune response. We show that cristae remodeling and inhibition of autophagy are both hallmarks of AICD. Moreover, we highlight that—at variance with what was expected—bulk autophagy and not mitophagy is necessary to remove dysfunctional mitochondria during apoptosis. Indeed, in conditions in which we specifically block bulk autophagy at very early stages, thus preventing autophagosome formation/maturation, we also block any other selective type of autophagy including mitophagy. This latter cannot proceed although the specific cargoes are in place and ready to be "selectively" degraded. By the way, such a dependence of mitophagy on the autophagy machinery is highly evolutionary conserved: in *S. cerevisiae* the cargo selectivity is apparently provided by the strong binding of its receptor, via its multiple binding sites, to the clustered Atg8 on the phagophore membrane. This binding increases the probability of interaction between the 2 molecules, and, by consequence, the amount of damaged organelles identified and carried within a canonical autophagosome.

We think that the molecular understanding of how mitochondria and autophagy work in AICD, and the identification of a predominant role for autophagy in an ideally "mitophagic context," could also have a more direct translational effect when dealing with various physiological or pathological conditions. In particular, this molecular knowledge could be exploited to pharmacologically modulate autophagy, or mitochondrial morphology, in the context of both autoimmune disease, in which the autoreactive immune response has to be shut down or, vice versa, in immunotherapy, where long-lived functional T cells are required to counteract cancer or pathogen attack.

ORCID

Silvia Campello http://orcid.org/0000-0003-0536-2484