Yoshida *et al.* have reached a crucial halfway point by pushing their simulation all the way toward the formation of a primordial protostar, the small core inside the collapsing primordial cloud, where for the first time stellar densities, close to that of liquid water on Earth, are reached and where hydrostatic equilibrium is established, the almost exact balance between gravity and thermal pressure. This protostar will subsequently grow by accreting material from the surrounding envelope until the accretion flow is shut off by the ever more intense radiation emitted by the protostar (*8*, *9*). The protostellar accretion problem marks the next frontier in the field, but the present study provides a firm foundation for addressing this challenge. The ultimate goal of predicting the mass and properties of the first stars is now within reach.

The course of early cosmic history depends on this prediction. Specifically, if the first stars were indeed very massive, they were copious producers of heavy chemical elements that were rapidly dispersed into the cosmic matter in the wake of the first supernova explosions, and of ultraviolet photons that were energetic enough to ionize hydrogen, the most abundant element in the universe. The first stars therefore began the extended process of "reionization," which transformed the universe from a completely neutral state during the dark ages, when the universe was cold and contained no stars, into a fully ionized medium today. Observations of the degree of polarization, resulting from photons scattering off free electrons, in the cosmic microwave background, carried out by the Wilkinson Microwave Anisotropy Probe (WMAP), place constraints on the onset of reionization. The WMAP result indicates that about 30% of the total signal was produced by still unobserved stars during the first billion years after the Big Bang (*6*).

Once we understand the first stars, we can then begin to meaningfully consider the formation of the first galaxies (see the figure), which would contain clusters of many primordial stars (*10*, *11*). These small galaxies are believed to be the most distant sources that can be detected with the upcoming James Webb Space Telescope (JWST), planned for launch around 2013, and it is therefore important to predict their properties, such as colors, sizes, and luminosities. Their assembly is strongly influenced by the feedback from the first stars that formed during the process, due to the radiation and heavy elements produced by them. Again, the character and strength of the stellar feedback during the formation of the first galaxies sensitively depend on the mass distribution of the primordial stars.

Even before the JWST, we can test our theoretical predictions for the first stars by hunting for fossils from the dark ages in our local cosmic neighborhood. In this "stellar archaeology" approach, large surveys of low-mass stars are carried out in our Milky Way that contain only a tiny amount of heavy elements. These would carry the imprint of the first stars that produced those elements with an abundance pattern that strongly depends on their mass (*12*, *13*). Until now, the simulations were not detailed enough to make predictions with the required degree of precision, but Yoshida *et al.* have paved the way for stellar archaeology to fulfill its great potential. The combination of cutting-edge supercomputer simulations, large surveys of low-mass fossils in the Milky Way, and the deep images from the JWST promises to close the final gap in our cosmic worldview in the decade ahead. We are clearly entering a period of rapid discovery.

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CELL SIGNALING

"Make and Brake" in Signaling

Aristides G. Eliopoulos

Proteolysis of one component of a receptor-bound complex releases the complex from the cell surface to the cytoplasm to activate kinase signaling.

Understanding how cytokines and growth factors trigger intracellular signal transduction is a major challenge in cell biology. Of particular interest is the tumor necrosis factor (TNF)/TNF receptor superfamily, which encompasses more than 20 different ligands and 30 receptors involved in fundamental biological processes including apoptosis, immunity, inflammation, and organogenesis. Ligand binding triggers assembly of multiprotein complexes at the corresponding receptor intracellular domains. The cellular signaling pathways that dictate the biological outcome of ligand binding are thought to be initiated within the receptor-bound complexes and terminated by complex dissociation. A paper by Matsuzawa *et al.* on page 663 of this issue (*1*) suggests otherwise. By studying the signaling requirements for CD40, a TNF receptor family member with a prominent role in immune regulation and homeostasis, the authors elegantly demonstrate that activation of the c-Jun N-terminal kinase (JNK) and p38 mitogen-activated protein kinase (MAPK) cascades occurs only upon release of a CD40-tethered signaling complex. This two-step mechanism of signal initiation provides new insight into the intricate regulation of receptor-mediated signal transduction.

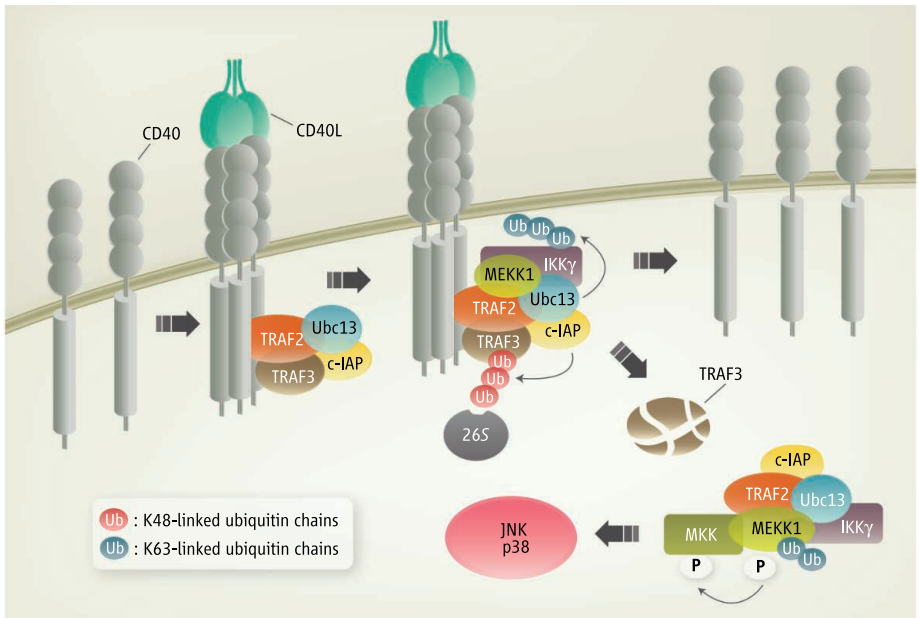
A major component of signaling by members of the TNF receptor superfamily is a group of cytoplasmic adaptor proteins known as TNF receptor–associated factors (TRAFs). Of the seven known mammalian TRAFs, CD40 directly binds TRAF2, TRAF3, and TRAF6 (*2*). TRAF2 and TRAF6 have attracted particular attention because although they lack intrinsic kinase activity, their ablation severely impairs CD40 signaling in lymphoid and nonhematopoietic cells (*3*–*5*). The contribution of TRAFs to signal transduction exceeds, however, their role as molecular bridges between TNF family receptors and intracellular signaling components. Indeed, both TRAF2 and TRAF6 possess E3 ubiquitin ligase activity and function together with the E2 ubiquitin-conjugating enzyme Ubc13 to catalyze the addition of lysine 63 (K63)–linked polyubiquitin chains to various protein kinases and cytoplasmic adaptors, including TRAFs themselves (*6*). Unlike lysine 48 (K48)–linked polyubiquitination, which signals for proteasome-

The University of Crete Medical School and Institute for Molecular Biology and Biotechnology, Foundation of Research and Technology Hellas, Heraklion, Greece. E-mail: eliopag@med.uoc.gr

dependent protein degradation, K63-linked polyubiquitination is nondestructive and activates protein kinases. Highlighting the significance of this modification in CD40 signal transduction, Ubc13-deficient B cells display impaired JNK and p38 MAPK activation in response to CD40 stimulation (*7*). Nevertheless, which protein kinase(s) in the signaling complex are targeted by TRAF2/Ubc13-dependent ubiquitination to activate MAPK signaling remains obscure.

Matsuzawa *et al*. analyzed the composition of the signaling complex assembled at the cytoplasmic tail of CD40 after receptor stimulation. The authors find that the binding of TRAF2 to CD40 is essential for recruitment of the K48-specific E3 ubiquitin ligase cellular inhibitor of apoptosis (c-IAP) and the K63 ligase Ubc13, followed by binding of IκB kinase γ (IKKγ) and MAP or extracellular signal–regulated kinase kinase 1 (MEKK1) (see the figure). IKKγ is best known as the regulatory subunit of the IκB kinase signaling complex involved in activation of the transcription factor nuclear factor κB (NF-κB). More recently, however, K63 ubiquitin-modified IKKγ has been found to influence MAPK activation induced by interleukin-1 and Toll-like receptor stimulation (*7*). Matsuzawa *et al.* show that the ubiquitination of IKKγ by TRAF2/Ubc13 is necessary for the recruitment of MEKK1 to the CD40-bound signaling complex (*1*). MEKK1 is a kinase operating at the start of MAPK cascades with a proven physiological role in CD40-induced MAPK signaling (*8*). It would thus be reasonable to predict that MEKK1 may be ubiquitinated by TRAF2/Ubc13 and activated within the CD40-bound signaling complex. Biological pathways are, however, notoriously unpredictable; thus, as Matsuzawa *et al*. report, MEKK1 associated with CD40 is neither ubiquitinated nor phosphorylated. Instead, ubiquitinated and phosphorylated MEKK1 is found in the cytosol in association with TRAF2, c-IAP, Ubc13, and IKKγ but not CD40. Within this complex, MEKK1 also interacts with and activates its substrates MKK4 and MKK3/6, which serve as JNK and p38 kinases, respectively. Thus, for MAPK signal activation, a complex assembled on CD40 must be released to the cytoplasm without dissociation.

What controls the release of this complex to the cytosol? The key here is shown to be TRAF3, which hetero-oligomerizes with TRAF2 but negatively regulates CD40-induced MAPK activation (*1*). Whereas TRAF2 undergoes both K63- and K48-linked polyubiquitination, TRAF3 undergoes only destructive K48-linked polyubiquitination that is catalyzed by c-IAPs and occurs rapidly at the level of the receptor. Concomitant with the proteasome-mediated degradation of TRAF3, the CD40-tethered signaling complex is released to the cytosol where MEKK1 is K63 polyubiquitin-modified by TRAF2/Ubc13 and activated (see the figure). TRAF2 therefore orchestrates MAPK signal initiation not only by nucleating an "effector" signaling complex at the cytoplasmic tail of CD40 but also by directing its release to the cytosol through the recruitment of c-IAP and the concomitant proteasome-mediated destruction of TRAF3. TRAF3 acts as a "brake" against premature signal initiation that is released when it is ubiquitinated.



A two-stage mechanism of cytokine signaling. Matsuzawa *et al*. suggest that CD40 binding to the CD40 ligand (CD40L) stimulates receptor oligomerization and the formation of a CD40-tethered multiprotein signaling complex that is necessary but not sufficient for JNK and p38 MAPK activation. The latter requires the c-IAP–mediated K48-linked polyubiquitination of TRAF3 and its degradation by the proteasome (26*S*), which causes the release of the signaling complex from CD40 to the cytosol allowing activation of the MEKK1/MKK/MAPK module.

Matsuzawa *et al*. also study TRAF6 and show that it operates in a manner similar to TRAF2 but associates with the apical kinase transforming growth factor–β activated kinase 1 (TAK1) instead of MEKK1 to activate MAPK signaling in CD40-stimulated B cells. Whether the TRAF2 and TRAF6 complexes share components or are formed independently to activate the MAPK pathways remains to be seen (*4*). Moreover, the fate of the cytoplasmic TRAF complexes in the context of signal termination needs to be defined. Although TRAF2 degradation and complex disassembly may determine the termination phase, a role for deubiquitinating enzymes in silencing MAPK signaling cannot be excluded. Further studies are also required to define the mechanism of CD40-induced NF-κB activation, which is critically controlled by TRAF6 (*3*, *4*) but, unlike MAPK, is initiated exclusively at the level of the receptor (*1*). Given that TRAF expression substantially varies among cell lines and tissues (*9*), it is tempting to speculate that the pleiotropic and even biologically opposed consequences of CD40 stimulation may reflect differences in complex assembly and disassembly. There are tantalizing indications that the proposed two-step mechanism of MAPK activation might be more general. Matsuzawa *et al*. show that, similar to CD40, TNF receptor I–mediated MEKK1 and TAK1 phosphorylation are c-IAP–dependent and previous work has demonstrated that TNF stimulates the formation of two TNF receptor signaling complexes: a plasma membrane, receptor-bound complex that rapidly induces NF-κB activation and a slowly formed cytoplasmic complex that mediates apoptotic responses (*10*). Further studies are needed to place a two-step model of MAPK signaling in the context of these findings

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