This paper is available online at www.jbc.org
THE JOURNAL OF BIOLOGICAL CHEMISTRY VOL. 284, NO. 13, p. 8209, March 27, 2009

Regulation of Signaling by Non-degradative Ubiquitination*

Published, JBC Papers in Press, November 13, 2008, DOI 10.1074/jbc.R800070200
Luke A. J. O'Neill[1]
From the School of Biochemistry and Immunology, Trinity College, Dublin 2, Ireland

A central unifying principle in the field of signal transduction has been the covalent modification of proteins by phosphorylation. A wide range of protein kinases are known, and the process is regulated by a more limited although important number of protein phosphatases. Recently, there has been a remarkable increase in the numbers of examples of another reversible covalent modification in proteins during signaling, ubiquitination. Historically, the function of ubiquitination was to cause protein degradation in the 26 S proteasome. The difference lies in the type of ubiquitin chain: if the linkage to the target protein is via Lys^{48} on ubiquitin, this will lead to a series of events that culminate in degradation of the polyubiquitinated protein by the 26 S proteasome. Also, there have to be a minimum of four ubiquitins attached in order to trigger degradation. However, if the polyubiquitin chains are linked via Lys^{63}, this will direct protein-protein interactions via a ubiquitin-binding domain on the interacting target protein. A major function of this type of ubiquitination appears to be to allow proteins with this domain to assemble into multiprotein complexes, which might lead to access to substrates if kinases are also involved, but also regulates processes such as endocytosis and ribosomal protein synthesis (1). The process of ubiquitination is mediated by ubiquitin ligases and is reversed by deubiquitinating enzymes. The best examples of this phenomenon are to be found in the regulation of innate immune signaling, where both phosphorylation and Lys^{63}-linked ubiquitination are the critical covalent modifications that launch signaling pathways activated by innate immune receptors such as the TLRs[2] (2). Here, we present three minireviews on this emerging and exciting topic that provide important examples of ubiquitination/deubiquitination in signaling.

In the first minireview, Sinéad E. Keating and Andrew G. Bowie provide an overview of non-degradative ubiquitination in signaling by innate receptors, particularly the TLRs, but also in response to the pro-inflammatory cytokine interleukin-1 (which, similar to TLRs, signals via the Toll/interleukin-1 receptor domain) (3). They describe how polyubiquitination is a key activation signal for the transcription factor NF-κB. Various ubiquitin ligases participate, notably TRAF6 (TNF receptor-associated factor) and TRAF3, which were formerly thought to be adapters that linked to kinases and are now known to be E3 ligases. Other ligases include Ubc13, which is an E2 ubiquitin-conjugating enzyme, and the Pellino proteins. Keating and Bowie also remind us that bacteria have deubiquitinating proteases that target TRAF6 and TRAF3. The best example of this is the virulence factor YopJ, which is in *Yersinia pestis*. The capacity to target ubiquitinated proteins would be expected to limit host-defense signaling pathways.

In the second minireview, Beatrice Coornaert, Isabelle Carpentier, and Rudi Beyaert describe the fascinating deubiquitinating enzyme A20 (4). Although first described as an inhibitor of TNF-induced apoptosis, we now know that a major function of A20 is to limit TLR signaling and to prevent sepsis. A20 is a dual ubiquitin-editing enzyme again in NF-κB signaling. It has a number of targets, notably TRAF2, TRAF3, TRAF6, and RIP1, all of which are deubiquitinated by A20. Most interestingly, it has been found that A20-deficient mice develop profound colitis (5). If they are crossed with MyD88-deficient mice, however, there is no disease. The authors describe how commensal bacteria activate TLRs, but instead of driving inflammation via MyD88 and the TRAF proteins, they are kept in check by A20. This indicates the importance of deubiquitinating systems to keep pathways under control.

This theme is taken up by Edward T. H. Yeh in the third minireview (6). This minireview describes the SUMO modification and in particular focuses on de-SUMOylation. SUMO is a ubiquitin-like protein that targets many different proteins in multiple processes, including innate immunity, but also the cell cycle, transcriptional control, and viral replication. SUMOylation involves only one conjugating enzyme, Ubc9, and a limited number of ligases. There are a number of de-SUMOylating proteases termed SENP (sentrin/SUMO-specific protease), which are still being characterized. Some respond to inflammatory stimuli such as TNF and are key controllers of SUMOylated proteins.

Much progress has been made in defining the biochemical basis for non-degradative ubiquitination and its role in cellular function. The authors and editors hope that this set of reviews will enable researchers in such areas as innate immunity to appreciate the importance of non-degradative ubiquitination for the control of complex signaling processes and possibly inspire further work on this most important event for protein function.

REFERENCES

1. Pickart, C. M., and Fushman, D. (2004) *Curr. Opin. Chem. Biol.* **8,** 610–616
2. O'Neill, L. A. J. (2008) *Immunity* **29,** 12–20
3. Keating, S. E., and Bowie, A. G. (2009) *J. Biol. Chem.* **284,** 8211–8215
4. Coornaert, B., Carpentier, I., and Beyaert, R. (2009) *J. Biol. Chem.* **284,** 8217–8221
5. Turer, E. E., Tavares, R. M., Mortier, E., Hitotsumatsu, O., Advincula, R., Lee, B., Shifrin, N., Malynn, B. A., and Ma, A. (2008) *J. Exp. Med.* **205,** 451–464
6. Yeh, E. T. H. (2009) *J. Biol. Chem.* **284,** 8223–8227

* This work was supported by Science Foundation Ireland. This minireview will be reprinted in the 2009 Minireview Compendium, which will be available in January, 2010.
[1] To whom correspondence should be addressed. E-mail: laoneill@tcd.ie.
[2] The abbreviations used are: TLR, Toll-like receptor; TNF, tumor necrosis factor; SUMO, small ubiquitin-like modifier.