Description of USP12 nuclear export sequence
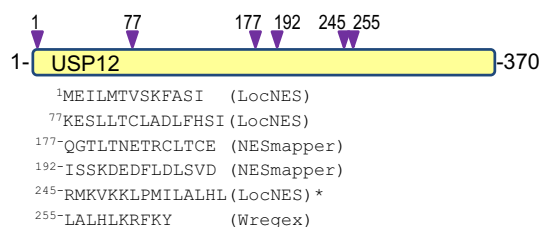
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In PNAS, Jahan et al. (1) describe a new role for the ubiquitin-specific protease 12 (USP12) deubiquitinase in T-cell receptor (TCR) signaling. Among other findings, the authors report that USP12 is exported from the nucleus to the cytosol upon TCR activation. Although the experimental evidence they provide is consistent with this model, I believe that some of their results would require a more detailed description to support their claims better.

Jahan et al. (1) analyze USP12 localization using cell membrane permeabilization followed by centrifugation and collection of supernatant and pellet fractions. In resting T cells, they detect USP12 mostly in the pellet fraction, whereas in cells stimulated with anti-CD3, USP12 is predominantly detected in the supernatant fraction. Assuming that the pellet fraction contains mostly nuclear proteins, the authors deduce from these observations that USP12 relocalizes from the nucleus to the cytosol upon TCR activation. They go on to show that the translocation of USP12 from the pellet to the supernatant fraction is blocked by treatment with leptomycin B, an inhibitor of the nuclear export receptor CRM1, or by mutation of a nuclear export signal (NES) in USP12. These findings provide further support to their model. However, this support is weakened, in my opinion, by the exceedingly vague description of the USP12 NES mutation used in their experiments. Jahan et al. (1) state that USP12 (and the related deubiquitinase USP46) are equipped with NESs, but they do not describe these motifs. We have previously tested candidate NESs in USP12 and USP46, and reported that these motifs are nonfunctional in a nuclear export assay (2). To the best of my knowledge, no reports of functional NES in USP12 have been published to date. Importantly, analysis of the USP12 amino acid sequence using NES prediction tools (3–5) reveals several candidate NES motifs (Fig. 1), and, therefore, the sequence referred to by the authors as NES is not obvious. In my view, a description of USP12 NES, and the specific amino acid mutations that render it inactive, would provide stronger support to the model of USP12 nuclear export proposed by Jahan et al. (1), and would facilitate independent confirmation of their interesting findings.

Fig. 1. Candidate NES motifs in USP12. Schematic representation of the USP12 protein shows the position of six different candidate NES motifs (arrowheads) predicted by three NES prediction tools: Wregex (weighted regular expression; "relaxed" configuration), LocNES, and NESmapper. The numbers above the arrowheads indicate the first residue of the predicted NES. The amino acid sequence and the program that predicted each motif are indicated below. One of these candidate NES motifs (marked with an asterisk) has been previously tested and reported to be nonfunctional in a nuclear export assay (2).

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