Interaction of the Anaphase-promoting Complex/Cyclosome and Proteasome Protein Complexes with Multiubiquitin Chain-binding Proteins*

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Fission yeast Rhp23 and Pus1 represent two families of multiubiquitin chain-binding proteins that associate with the proteasome. We show that both proteins bind to different regions of the proteasome subunit Mts4. The binding site for Pus1 was mapped to a cluster of repetitive sequences also found in the proteasome subunit SpRpn2 and the anaphase-promoting complex/cyclosome (APC/C) subunit Cut4. The putative role of Pus1 as a factor involved in allocation of ubiquitylated substrates for the proteasome is discussed.

Ubiquitin-dependent protein degradation is a mechanism employed in eukaryotic cells not only to recycle damaged and misfolded proteins but also to control cellular processes by specific breakdown of regulatory proteins (1). Ubiquitylation is accomplished in multiple steps (2). Initially ubiquitin is activated by a ubiquitin-activating enzyme (E1) in an ATP-dependent process whereby the E1 forms a thiol ester bond with the C terminus of ubiquitin. Subsequently the ubiquitin molecule is transferred to a ubiquitin-conjugating enzyme (E2). E2s carrying ubiquitin associate with substrate binding ubiquitin protein ligases (E3s) resulting in the covalent attachment of ubiquitin to the substrate protein. Several rounds of this conjugation process produce substrates carrying a chain of ubiquitin molecules. There is a range of different E2s that associate with various E3s, incorporating an element of substrate specificity to this process.

Ubiquitin protein ligases can be divided in two major groups, HECT domain E3s and RING finger domain E3s. The anaphase-promoting complex/cyclosome (APC/C)1 belongs to a family of multimeric ubiquitin protein ligases of the RING finger type that also include the SCF and the VCB (2, 3). It has a molecular mass of 700 kDa and consists of 11 different subunits. Mitotic events are controlled by the APC/C via multiubiquitylation of cell cycle regulators like Cut2/Pds1p-securin and B-type cyclins. Once a ubiquitin chain is conjugated to these proteins they become targets for degradation by the 26 S proteasome enabling the continuation of downstream mitotic events.

The 2.5 MDa 26 S proteasome catalyzes the degradation of cellular proteins in an ATP-dependent manner (4). Its proteolytic component is the 20 S core complex, a cylindrical structure comprising four stacked rings each containing seven proteins. The inner rings enclose a central chamber harboring the catalytic sites. Access to the lumen is provided via the outer rings and regulated by the 19 S regulatory complex that is attached to one or both ends of the 20 S core (5).

The 19 S regulatory complex can be dissociated in two subcomplexes called the base and the lid (6, 7). Six ATPases subunits are presumed to form a structure that associates with the 20 S core. Together with the two largest subunits of the proteasome SpRpn2/Rpn2/S1 and Mts4/Rpn1/S2 these ATPases form the base complex that was suggested to be participating in the unfolding and translocation of substrates. The lid complex is believed to contact the base via the subunits SpRpn2/Rpn2/S1, Mts4/Rpn1/S2, and Pus1/Rpn10/S5a. It is composed of a number of non-ATPase subunits whose function remains rather enigmatic. Proteasomes lacking the lid can cleave small peptides in an ATP-dependent manner, but they are unable to degrade ubiquitylated proteins (6). Thus the lid is believed to be involved in the recognition and processing of those substrates.

To date, one subunit of the proteasome has been found to possess the ability to recognize multiubiquitylated substrates, namely Pus1/Rpn10/S5a (8). However, given the fact that in yeast the corresponding RPN10+pus1+ genes are not essential for cell viability, other mechanisms must clearly exist for this crucial step in the degradation process (9). Recently, it has been shown that the UBA/UBL domain proteins Rhp23/Rad23p and Dph1/Dsk2p can interact with both the proteasome and with multiubiquitin suggesting that these factors play a role in the recognition and delivery of substrates for proteolysis (10–13). The Pus1/Rpn10p protein contains a stretch of 20 amino acids recently defined as ubiquitin-interacting motif (UIM) that is involved in multiubiquitin chain binding, whereas in Dph1/Dsk2p and Rhp23/Rad23p the task is thione S-transferase; GFP, green fluorescent protein; HA, hemagglutinin.
accomplished by a C-terminal ubiquitin pathway associated (UBA) domain (9, 10, 14, 15, 16). Both Dph1/Dsk2p and Rhp23/ Rad23p bind to the proteasome with their N-terminal UBA domain (10, 17). As Pus1/Rpn10p does not have a UBL domain it must use different structures to associate with other protea-

somal subunits. Lamberton et al. (18) were able to precipitate the proteasome from cells deleted for RPN10 and RAD23 using tagged Rad23tp or Rpn10p, respectively, demonstrating that these proteins bind to the proteasome independently.

So far little is known about how ubiquitinylated substrates associate directly with the proteasome machinery and the proteasome as implied by the finding that HPLC proteins, the human homologues of Dph1/Dsk2p, associate with E3 proteins (24). However, previously neither Rhp23/Rad23p nor Pus1/Rpn10p have been re-

ported to interact with ubiquitinylating enzymes.

To improve our understanding of how the proteasome re-
cruits ubiquitin substrates we started to investigate the asso-
ciation of these proteins with the 19 S regulatory complex. We characterize regions in proteasome subunits that bind Pus1 and Rhp23 and describe the interaction of these multiubiqu-
in-binding proteins with the APCC.

EXPERIMENTAL PROCEDURES

Bioinformatics—Sequence similarity searches were carried out using Psi-BLAST (version 2.2.3–3) (25) using the BLOSUM62 substitution ma-
trix for the first iteration. Sequences identified with a BLAST E-value of <0.0001 were filtered to remove overly similar se-
quences (80% identity in alignment) and aligned using ClustalW (1.74). The HMMer 2.0 package (version 2.2.9) was used to generate and optimize Hidden Markov models (HMMs) from the multiple sequence alignment. HMMsearch was used with the HMM to identify further examples of the UBL binding region in the SPTR data base.

Schizosaccharomyces pombe Strains and Techniques—

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Plasmids—The plasmid pREP1Rhp23 was produced as described (10). To generate Mts4 constructs FL (full-length), N

–1 (amino acids

744) and Cut4–(873 –243), and C

–193) were constructed by

inserting in-frame into the pGEX-KGPus1 plasmids were generated by ampli-
fying in-frame from genomic

DNA and

subcloned into pQE30 (Qiagen) except Mts4PC1, which was

inserted in-frame into the pGEX-KG plasmid.

RESULTS

The UBL Domain of Rhp23 Binds to the Proteasomal Subunit Mts4—As a first step to characterize the interactions of Rhp23 with the proteasome we employed the yeast two-hybrid system. We tested the ability of Rhp23 to bind to the proteasomal subunits Mts1 (Rpn9), Mts2 (Rpt2p), Mts3 (Rpn12p), SpRpn2 (Rpn2), Mts4 (Rpn1), Pad1 (Rpn11), Pus1 (Rpn10), SpRpt1 (Rpt1), SpRpt6 (Rpt6), and to the proteasome-associated ubiquitin hydrolase Uch2. The two-hybrid assay detected an interaction of Rhp23 with Mts4, a subunit of the proteasomal base complex (data not shown). To test whether there is also a genetic interaction between rhp23 and mts4 we used the fission yeast mts4-1 strain that carries a mutation in the gene encoding Mts4 (27). By transforming mts4-1 cells with the plasmid pREP1Rhp23, in which Rhp23 expression is driven by the thiamine inducible nmt1 promoter, we were able to partially suppress the temperature sensitive (ts) phenotype of the mutant (Fig. 1A). As a control we also introduced the plasmid

into mts2-1 cells, which have a mutation in the gene for the proteasome subunit Mts2 and display a temperature sensitive phenotype similar to mts4-1 (29). However, transformation of mts2-1 with pREP1Rhp23 could not rescue the mutant phenotype indicating that the interaction between rhp23 and mts4 is specific. In addition, the observed genetic interaction was dependent on the Rhp23 UBL domain as when this domain was deleted from the rhp23 gene suppression of the mts4-1 mutant no longer occurred.2

In order to determine whether this genetic interaction re-
flected a direct physical interaction between Rhp23 and Mts4, we performed an in vitro binding assay. A GST-Rhp23 fusion protein coupled to glutathione-Sepharose was incubated with His-tagged full-length Mts4 as well as N- and C-terminal truncated versions of the protein expressed in Escherichia coli. As shown, GST-Rhp23 precipitated Mts4 from the bacterial ex-
tracts (Fig. 1B) demonstrating a direct interaction between these proteins. The use of truncated versions of Mts4 revealed

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that Rhp23 binds within a region stretching from amino acid position 181 to 407.

It has been shown that Rhp23 binds the proteasome via its UBL domain (10). Therefore the binding of Rhp23 to Mts4 should also be dependent upon the UBL domain. Truncated versions of Rhp23 fused to GST were used in an in vitro binding assay similar to the one described above. We found that only the truncation of Rhp23 containing the Rhp23 protein. We could narrow down the binding site for Pus1 on Mts4 and analyzed as described above.

To demonstrate that binding to the Mts4/Rpn1 subunit of the 26 S proteasome is a general property of proteins that contain a UBL domain we assayed the fission yeast Udp7 protein (SwissProt accession number Q94580). The Udp7 protein, like the Rhp23 protein, contains a UBL domain in its N terminus.

When assayed in vitro GST-Udp7 was able to precipitate Mts4 protein from the bacterial extracts demonstrating an interaction (Fig. 1D). This result is consistent with the hypothesis that the UBL domain is a general Mts4/Rpn1 interaction domain.

Pus1 Binds Mts4 in a Region Containing Repetitive Sequences—It has been demonstrated in budding yeast that Rpn10p (Pus1) interacts with Rpn1 (Mts4) in an in vitro binding assay (20). Thus we investigated interactions between these two subunits. Previously, a genetic interaction was detected between the genes encoding these two proteins. The mts4-1 allele was found to be synthetically lethal with a null allele of pus1 (9). The Pus1 binding site within Mts4 was determined in an in vitro binding assay using the same Mts4 constructs employed for mapping the Rhp23 binding site. An N-terminal GST fusion of Pus1 immobilized on glutathione-Sepharose was used to precipitate full-length Mts4 as well as truncated versions of the protein. We could narrow down the binding site for Pus1 on Mts4 to a stretch of amino acids ranging from position 408 to 582 (Fig. 2, A and B). This particular region contains a cluster of repeats that can also be found in SpRpn2, the fission yeast homologue of the budding yeast Rpn2p subunit of the proteasome, and in Cdt4 a subunit of the APC/C or anaphase-promoting complex (30). Although the repeats are rather weakly conserved they appear to be present only in orthologues of the proteins mentioned. In the Prosite database (www.expasy.ch/prosite/), this structure is referred to as APC_SEN3_REPEAT.
(accession number PS50248), whereas the Pfam data base (www.sanger.ac.uk/Pfam/) lists it as PC_rep for proteasome/cyclosome repeat (accession number PF01851). We will therefore call it PC repeat.

**Pus1 Also Binds to PC Repeat Modules of SpRpn2 and Cut4**—Having shown that Pus1 binds to one of the PC repeat modules found in Mts4 we asked whether Pus1 might also interact with the PC repeat modules present in SpRpn2 and Cut4. Moreover we wanted to narrow down the region of Pus1 that is responsible for binding the PC repeat structures. We expressed truncated versions of Pus1 with an N-terminal GST fusion. These constructs were tested in a binding assay using His-tagged PC repeat modules of Mts4, SpRpn2 and Cut4. Both the full-length Pus1 and PusCΔ1 were capable of binding to all three PC repeat modules tested, whereas no binding was observed using Pus1Δ1 and Pus1Δ2 (Fig. 2B). From these data we conclude that the N-terminal region of Pus1 up to amino acid position 84 appears to contain a region necessary for binding PC repeat modules.

**Pus1 and Rhp23 Interact with APC/C**—We have demonstrated that Pus1 can bind to the PC repeat module present in the APC/C subunit Cut4 in *vivo*. If Pus1 binds to Cut4 in *vivo*, it should be possible to use the GST-Pus1 fusion protein to precipitate the APC/C from fission yeast extracts. Therefore we incubated GST-Pus1 and GST-Rhp23 with extracts from *cut9HA* cells that carry a genomically HA-tagged version of the APC/C subunit Cut9 and tested whether these GST fusions could precipitate the APC/C and/or the proteasome. Because Rhp23 and Pus1 bind to multiubiquitin chains it had to be considered that they might interact with multiubiquitinylated substrates attached to the APC/C. Taking that into account, we also used Pus1N5 and Rhp23PP, which have mutations in their ubiquitin binding domains and do not bind multiubiquitin (9, 10). Furthermore, we tested truncated versions of Pus1, which lack the C-terminal multiubiquitin binding motif (Pus1CΔ1) as well as versions of Rhp23 lacking either its UBL domain (Rhp23NΔ1) or both UBA domains (Rhp23CΔ1). We were able to precipitate the APC/C and the proteasome using Pus1, Pus1N5, or Pus1CΔ1 fusion proteins but not Pus1NΔ1 or Pus1NΔ2 (Fig. 3A). These findings indicate that the interaction of Pus1 with the APC/C and the proteasome is independent of its ability to bind ubiquitin conjugates but can be assigned to the Pus1 N terminus up to amino acid position 84, which is consistent with the results of the mapping studies involving the Cut4 PC repeat region.

Rhp23, Rhp23PP, and Rhp23 but not Rhp23NΔ1 were found to precipitate the proteasome, confirming that the UBA domains of Rhp23 are not involved in proteasome binding (10). However, the APC/C binding activity of Rhp23 appears to be mediated by its multiubiquitin binding UBA domains as we could detect APC/C when beads coated with Rhp23 or Rhp23NΔ1 but not with Rhp23PP or Rhp23CΔ1 were used. Thus it is likely that Rhp23 binds via multiubiquitinylated substrates associated with the APC/C.

Given that Pus1 and Cut4 interact directly, we investigated whether any genetic interactions exist using existing mutant alleles of these genes. We crossed the *pus1* strain to the temperature sensitive mutant *cut4*Δ-333 (31). Spores carrying both the *cut4*-333 and the *apus1* allele were not viable. As a control Δ *pus1* was crossed to *cut9*-665 another temperature sensitive APC/C mutant (31). Synthetic lethality was not observed between *cut9*-665 and *apus1* indicating a specific genetic interaction between *pus1* and *cut4*Δ.

In order to elucidate whether the genetic data mirror an *in vivo* interaction between Pus1 and the APC/C we performed co-precipitation studies. Extracts from *cut9HA* cells expressing either GST or GST-Pus1 were incubated with glutathione beads and analyzed by SDS-PAGE and Western blotting. As presented in Fig. 3B we detected the APC/C subunit Cut9 in precipitates from *cut9HA/GST-Pus1* but not from *cut9HA/GST-Pus1* extracts was loaded, and on the left panel 10 μl of the precipitates were loaded and analyzed by anti-HA Western blotting.

**DISCUSSION**

We have characterized the interactions of Pus1 and Rhp23, representing two families of multiubiquitin-binding proteins, with the proteasome and the APC/C. Our studies demonstrate that Rhp23 binds to Mts4, a base component of the proteasomal 19 S regulatory complex. The UBL domain of Rhp23, which is responsible for the interaction with the proteasome, appears to bind the Mts4 protein between amino acid positions 181 and 407. Using this sequence to search the data base demonstrates that it is only found in other Mts4/Rpn1 orthologues from other species. Interestingly, this sequence comprises part of a pfamB domain, pfam B_4211. A multiple sequence alignment of the conserved pfamB domain is shown in Fig. 4. A homology search of the recently sequenced *S. pombe* genome with this pfamB domain demonstrated that it was only present in the Mts4 protein. Considering that there are a number of proteins carrying UBL domains two conclusions could be drawn. Either the structures that interact with the different UBL domains do not resemble each other, or all proteins containing a UBL domain can bind to Mts4/Rpn1. In this study we have shown that the UBL domain containing protein Ubp7 is also able to interact directly with Mts4/Rpn1 indicating that the latter hypothesis seems more probable. Consistent with this hypothesis the Dph1/Dsk2, Bag1, and Ubp6/Usp14 UBL containing proteins...
have already been shown to bind the proteasome, presumably by interaction with the Mts4/Rpn1 subunit, although this has only been shown for the budding yeast Dsk2 protein (32–34).

Using protein cross-linking studies it has been reported that the budding yeast Dsk2 and Rad23 UBls interact with both the Rpn1 and Rpn2 subunits (35). In contrast, in our *in vitro* binding experiments we found no interaction with the fission yeast Rpn2 orthologue and the fission yeast Rad23 orthologue, Pus1 interacts with the PC repeat clusters with five and three repeats, respectively, which we call here PC-1 and PC-2. The UBL domain of the Rhp23 protein interacts with the Mts4/Rpn1 protein in a region 5′ to the PC domain. Ubiquitin conjugates are thus brought to a region between the Lid subcomplex and the ATPase ring by binding to the UBA or UIM multiubiquitin binding domains.

FIG. 4. Multiple sequence alignment of the conserved pfamB region within the Rhp23 UBL binding region. Aligned sequences are defined by a genus/species abbreviation followed by the SPTR accession number. Sp, S. pombe; Sc, Saccharomyces cerevisiae; Hs, Homo sapiens; Dn, Drosophila melanogaster; Ce, Caenorhabditis elegans; Eh, Entamoeba histolytica; Pt, Plasmodium falciparum; Tc, Trypanosoma cruzi; At, Arabidopsis thaliana. Black background indicates 100% identity, dark gray 80%, and light gray 60% identity. Numbers to the right of the alignment show the corresponding amino acid coordinate for the protein sequence (as defined by the SPTR accession).

FIG. 5. Interaction of the multiubiquitin chain-binding proteins with the proteasome. Scheme summarizing the interactions between the Pus1 and Rhp23 multiubiquitin-binding proteins and the 19 S regulatory complex of the proteasome. Pus1 interacts with the PC domains of Mts4/Rpn1 and SpRpn2 via the PC binding region (PCB) located within the N-terminal 84 amino acids of the Pus1 protein. The UBL domain of the Rhp23 protein interacts with the Mts4/Rpn1 protein in a region 5′ to the PC domain. Ubiquitin conjugates are thus brought to a region between the Lid subcomplex and the ATPase ring by binding to the UBA or UIM multiubiquitin binding domains.

not able to detect binding between those proteins (data not shown). In addition, in mammalian cells it has been shown that the hHR23 and S5a proteins, the human versions of the fission yeast Rhp23 and Pus1 proteins, can directly interact with each other (37). Using deletion analysis the S5a binding site was determined to be in the second UIM binding domain present in this protein. This domain is not present in the simpler versions of the fission yeast Pus1 and budding yeast Rpn10 proteins (9, 38). Consistent with this we observed no direct interaction between the fission yeast Pus1 and Rhp23 proteins.3

The proteasome subunits SpRpn2 and Mts4 appear to play a central role in the structural organization of the 19 S regulatory complex. They link the ATPases of the base and components of the lid and also harbor binding sites for proteins that recognize multiubiquitinated substrates (See Fig. 5). This provides a simple model for the delivery of ubiquitinated substrates between the lid subcomplex and the ATPase ring. Presumably upon release from the multiubiquitin-binding proteins the ubiquitinated substrates are captured by the multiubiquitin binding Rpt5/S6′ ATPase, before the substrate protein is unfolded by the action of the ATPase ring and the polypeptide translocated to the 20 S catalytic complex for degradation (39).

Characterization of the Pus1 association with Mts4 revealed a specific interaction between Pus1 and a cluster of PC repeats in Mts4. These repetitive sequences contain an alternating pattern of large aliphatic residues and glycine or alanine (30). It has been suggested that these repeats form a structure possessing a concave surface of a hydrophobic nature which might represent a binding site. According to the PDB data base, Mts4 contains two PC repeat clusters with five and three repeats, respectively, which we call here PC-1 and PC-2. The mapping experiments show that PC-1 is sufficient to bind Pus1. It is conceivable that PC-1 and PC-2 together form one binding site. This would imply that deletion of PC-2 does not interfere...
with the integrity of the binding site, but that PC-2 on its own is unable to form a functional binding structure itself. Mts4 binds not only Rph23 and Pus1, its human homologue S2 was also shown to associate with the HECT E3 KIAA10 (22). SpRpn2 can bind Pus1 and for its budding yeast orthologue Rpn2p, an interaction with the RING finger E3 Ubr1p was demonstrated (20).

Data base searches revealed that PC repeats can be found only in the two largest subunits of the proteasome Mts4/Rpn1 and SpRpn2 and in the APC/C component Cut4/APC1. In binding studies we demonstrated that Pus1 could bind also to the PC repeat clusters of SpRpn2 and Cut4. This indicates that binding of Pus/Rpn10 represents a general and conserved function of these PC repeat structures.

The Pus1/Rpn10 protein represents a special case among regulatory subunits of the proteasome as it is the only subunit that also occurs in low molecular weight fractions of cell extracts (9, 38, 40). Furthermore it has been suggested that Rpn10 might have a role in linking the proteasome base and tract (9, 38, 40). It has been suggested that also occurs in low molecular weight fractions of cell excretion of these PC repeat structures.

PC repeat clusters of SpRpn2 and Cut4. This indicates that only in the two largest subunits of the proteasome Mts4/Rpn1 and SpRpn2 demonstrated (20).

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REFERENCES
1. Herschk, A., and Ciechanover, A. (1998) Annu. Rev. Biochem. 67, 425–479
2. Weissman, A. M. (2001) Nat. Rev. Mol. Cell. Biol. 2, 169–178
3. Page, A. M., and Hieter, P. (1999) Annu. Rev. Biochem. 68, 583–609
4. Voges, D., Zwickl, P., and Baumeister, W. (1999) Annu. Rev. Biochem. 68, 1015–1046
5. Grell, M., Ditzel, L., Lowe, J., Stock, D., Bochtler, M., Bartunik, H. D., and Huber, R. (1997) Nature 386, 463–471
6. Glickman, M. H., Rubin, D. M., Coux, O., Weses, I., Pfeifer, G., Cjeka, Z., Baumeister, W., Freed, V. A., and Finley, D. (1998) Cell 94, 615–623
7. Ferrell, K., Wilkinson, C. R., Dubiel, W., and Gordon, C. (2000) Trends Biochem. Sci. 25, 85–88
8. Deveraux, Q., Ustrell, V., Pickart, C., and Rechsteiner, M. (1994) J. Biol. Chem. 269, 7059–7061
9. Wilkinson, C. R., Ferrell, K., Penney, M., Wallace, D., Dubiel, W., and Gordon, C. (2000) J. Biol. Chem. 275, 15182–15192
10. Wilkinson, C. R., Steeger, M., Hartmann-Petersen, R., Stone, M., Wallace, M., Semple, C., and Gordon, C. (2001) Nat. Cell Biol. 3, 939–943
11. Funakoshi, M., Sanaki, T., Nishimoto, T., and Kobayashi, H. (2002) Proc. Natl. Acad. Sci. U. S. A. 99, 745–750
12. Rao, H., and Sastry, A. (2002) J. Biol. Chem. 277, 11691–11695
13. Sasaki, Y., Saih, Toh, C., and Yosokawa, H. (2002) Biochem. Biophys. Res. Commun. 293, 986–992
14. Pu, H., Sadis, S., Rubin, D. M., Glickman, M., van Noeker, S., Finley, D., and Vierstra, R. D. (1996) J. Biol. Chem. 271, 1970–1981
15. Haracska, L., and Udvary, A. (1997) FEBS Lett. 412, 331–336
16. Hofmann, K., and Pasquet, L. (2001) Trends Biochem. Sci. 26, 347–350
17. Schauber, C., Chen, L., Tongaonkar, P., Vega, I., Lambertson, D., Potts, W., and Madura, K. (1998) Nature 391, 715–718
18. Lambertson, D., Chen, L., and Madura, K. (1999) Genes 153, 69–79
19. Orito, T., Tongaonkar, P., Lambertson, D., Chen, L., Schauber, C., and Madura, K. (2000) Nat. Cell Biol. 2, 601–608
20. Xu, Y., and Varshavsky, A. (2000) Proc. Natl. Acad. Sci. U. S. A. 97, 2497–2502
21. Tongaonkar, P., Chen, L., Lambertson, D., Ko, B., and Madura, K. (2000) Mol. Cell. Biol. 20, 4691–4698
22. You, J., and Pickart, C. M. (2001) J. Biol. Chem. 276, 19871–19878
23. Young, P., Deveraux, Q., Beal, E. R., Pickart, C. M., and Rechsteiner, M. (1998) J. Biol. Chem. 273, 5461–5467
24. Kleijnen, M. F., Shih, A. H., Zhou, P., Kumar, S., Socci, N. B., Eshers, L., Gill, G., and Howley, P. M. (2000) Mol. Cell. 6, 409–419
25. Altschul, S. F., Madden, T. L., Schaffer, A. A., Zhang, J., Zhang, Z., Miller, W., and Lipman, D. J. (1997) Nucleic Acids Res. 25, 3389–3402
26. Moreno, S., Klar, A., and Nurse, P. (1991) Methods Enzymol. 194, 795–823
27. Wilkins, C. R., Wallace, M., Steeger, M., Dubiel, W., and Gordon, C. (1997) J. Biol. Chem. 272, 25768–25777
28. Madronell, K. (1999) J. Biol. Chem. 265, 10857–10964
29. Gordon, C., McGurk, G., Dillon, P., Rosen, C., and Hastie, N. (1993) Nature 366, 355–357
30. Lupsa, A., Baumeste, W., and Hofmann, K. (1997) Trends Biochem. Sci. 22, 195–196
31. Hiram, T., Funahashi, S., Uemura, T., and Yanagida, M. (1986) EMBO J. 5, 2973–2979
32. Elssasser, S., Gali, R. R., Schwickhart, M., Larsen, C. N., Leggett, D. S., Muller, B., Feng, M. T., Tubing, F., Dittmar, G. A., and Finley, D. (2002) Nat. Cell Biol. 4, 725–730
33. Luders, J., Deand, M., and Hohfeld, J. (2000) J. Biol. Chem. 275, 4611–4617
34. Leggett, D. S., Hanna, J., Borodovsky, V., Croas, B., Schmidt, M., Baker, T., Walz, T., Foege, H., and Finley, D. (2002) Mol. Cell. 10, 495–507
35. Sasaki, Y., Tobe, A., and Yosokawa, H. (2002) Biochem. Biophys. Res. Commun. 296, 813–819
36. Walters, K. J., Kleijnen, M. F., Goh, A. M., Wagner, G., and Howley, P. M. (2002) Biochemistry 41, 1767–1777
37. Hiyama, H., Yuki, M., Masutani, C., Sugawara, K., Makawa, T., Tanaka, K., Hoejmakers, J. H., and Hanaoka, F. (1999) J. Biol. Chem. 274, 28019–28025
38. van Noeker, S., Sadis, S., Rubin, D. M., Glickman, M., Pu, H., Coux, O., Weses, I., Finley, D., and Vierstra, R. D. (1996) Mol. Cell. Biol. 16, 6020–6028
39. Lam, Y. A., Lawson, T. G., Velkyutham, M., Zweer, I. J., and Pickart, C. M. (2002) Nature 416, 763–767
40. Haracska, L., and Udvary, A. (1995) Eur. J. Biochem. 231, 720–725
41. Dai, R. P., and Li, C. C. (2001) Nat. Cell Biol. 3, 740–744
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