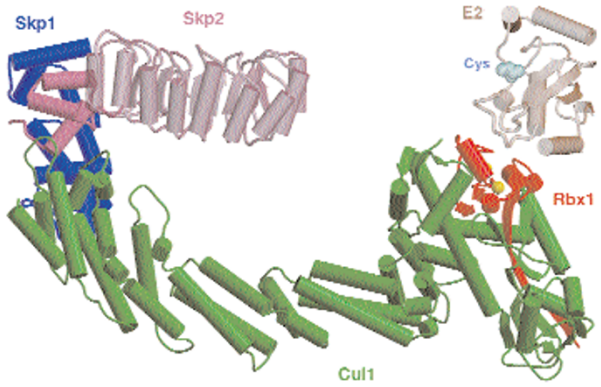
1. McParland, V.J., Kalverda, A.P., Homans, S.W. & Radford, S.E. *Nature Struct. Biol.* **9**, 326–331 (2002).
2. Hoshino, M. *et al. Nature Struct. Biol.* **9**, 332–336 (2002).
3. Kelly, J.W. *Curr. Opin. Struct. Biol.* **6**, 11–17 (1996).
4. Rochet, J.-C. & Lansbury, P.T.Jr *Curr. Opin. Struct. Biol.* **10**, 60–68 (2000).
5. Sunde, M. & Blake, C.C.F. *Q. Rev. Biophys.* **31**, 1–39 (1998).
6. Dobson, C.M. *Trends Biochem. Sci.* **24**, 329–332 (1999).
7. Wetzel, R. *Cell* **86**, 699–702 (1996).
8. Kelly, J.W. *Curr. Opin. Struct. Biol.* **8**, 101–106 (1998).
9. Pepys, M.B. in *Immunological Diseases*, Vol. 1 (ed. Samter, M.) 631–674 (Little, Brown & Co., Boston/Toronto, 1988).
10. Jacobson, D.R. & Buxbaum, J.N. *Adv. Hum. Genet.* **20**, 69–123 (1991).
11. Kisilevsky, R. *Lab. Invest.* **49**, 381–390 (1983).
12. Kisilevsky, R. *J. Struct. Biol.* **130**, 99–108 (2000).
13. Selkoe, D.J. *Science* **275**, 630–631 (1997).
14. Hammarstrom, P., Schneider, F. & Kelly, J.W. *Science* **293**, 2459–2461 (2001).
15. Anderson, O. & Wallin, G. in *Second International Symposium On Familial Amyloidotic Polyneuropathy And Other Transthyretin Related Disorders* Vol. 1 49 (Skelleftea, Sweden; 1992).
16. Ando, Y. *Rinsho Byori* **48**, 425–429 (2000).
17. Coelho, T. *Curr. Opin. Neurol.* **9**, 355–359 (1996).
18. Suhr, O.B., Herlenius, G., Friman, S. & Ericzon, B.G. *Liver Transp.* **6**, 263–276. (2000).
19. Lamber, M.P. *et al. Proc. Natl. Acad. Sci. USA* **95**, 6448–6453 (1998).
20. Snyder, S.W. *et al. Biophysical J.* **67**, 1216–1228 (1994).
21. Klein, W.L., Krafft, G.A. & Finch, C.E. *Trends Neurosci.* **24**, 219–224 (2001).
22. Harper, J.D., Wong, S.S., Lieber, C.M. & Lansbury, P.T.Jr *Chem. Biol.* **4**, 119–125 (1997).
23. Yong, W. *et al. Proc. Natl. Acad. Sci. USA* **99**, 150–154 (2002).
24. Ohashi, K. *Pathol. Intl.* **51**, 1–10 (2001).
25. Gejyo, F. *et al. Biochem. Biophys. Res. Comm.* **129**, 701–706 (1985).
26. Shirahama, T. *et al. Lab. Invest.* **53**, 705–709. (1985).
27. Connors, L.H., Shirahama, T., Skinner, M., Fenves, A. & Cohen, A.S. *Biochem. Biophys. Res. Comm.* **131**, 1063–1068. (1985).
28. Gorevic, P.D. *et al. Proc. Natl. Acad. Sci. USA* **83**, 7908–7912 (1986).
29. Colon, W. & Kelly, J.W. *Biochemistry* **31**, 8654–8660 (1992).
30. Booth, D.R. *et al. Nature* **385**, 787–793 (1997).
31. Hurle, M.R., Helms, L.R., Li, L., Chan, W. & Wetzel, R. *Proc. Natl. Acad. Sci. USA* **91**, 5446–5450 (1994).
32. Qu, B.-H., Strickland, E. & Thomas, P.J. *J. Biol. Chem.* **272**, 15739–15744 (1997).
33. Chen, C.D. *et al. EMBO J.* **20**, 6277–6287 (2001).
34. Kazmirski, S.L. *et al. Nature Struct. Biol.* **9**, 112–116 (2002).
35. McParland, V. *et al. Biochemistry* **39**, 8735–8746 (2000).
36. Chiti, F. *et al. J. Biol. Chem.* **276**, 46714–46721 (2001).
37. Hasebe, Y. Kanazawa Daigaku Juzen Igakkai Zasshi **102**, 228–237 (1993).
38. Liu, K., Cho, H.S., Lashuel, H.A., Kelly, J.W. & Wemmer, D.E. *Nature Struct. Biol.* **7**, 754–757 (2000).
39. Tycko, R. *Curr. Opin. Chem. Biol.* **4**, 500–506 (2000).
40. Tycko, R., Antzutkin, O.N., Balbach, J.J. & Reed, J. *Polym. Mater. Sci. Eng.* **82**, 133–134 (2000).
41. Benzinger, T.L. *et al. Biochemistry* **39**, 3491–3499 (2000).
42. Gregory, D.M. *et al. Solid State NMR* **13**, 149–166 (1998).
43. Benzinger, T.L. *et al. Proc. Natl. Acad. Sci. USA* **95**, 13407–13412 (1998).
44. Sunde, M. & Blake, C. *Adv. Protein Chem.* **50**, 123–159 (1997).
45. Jimenez, J.L. *et al. EMBO J.* **18**, 815–821 (1999).
46. Serpell, L.C. *et al. J. Mol. Biol.* **254**, 113–118 (1995).
47. Serpell, L.C. *et al. J. Mol. Biol.* **300**, 1033–1039 (2000).
48. Kheterpal, I., Zhou, S., Cook, K.D. & Wetzel, R. *Proc. Natl. Acad. Sci. USA* **97**, 13597–13601 (2000).
49. Sunde, M. *et al. J. Mol. Biol.* **273**, 729–739 (1997).
50. Lynn, D.G. & Meredith, S.C. *J. Struct. Biol.* **130**, 153–173 (2000).
51. Blake, C. & Serpell, L. *Structure* **4**, 989–998 (1996).
52. Walsh, D.M. *et al. Nature* **416**, 535–539 (2002).
53. Bucciantini, M. *et al. Nature* **416**, 507–511 (2002).

Locked but not loaded

Eukaryotic cells possess a highly regulated pathway whereby proteins are specifically targeted for degradation. Ubiquitin-dependent proteolysis is a two-step process involving the covalent attachment of ubiquitin (Ub), which tags a protein for degradation, followed by the selective degradation of the tagged protein by the 26S proteasome. The mechanism of protein ubiquitination that triggers Ub-dependent proteolysis depends on a cascade of three enzymes, E1, E2 and E3. The covalent attachment of Ub to the target protein is mediated by E3, the ubiquitin-protein ligase complex; however the exact mechanism by which these protein complexes mediate ubiquitination is not clear.

In a recent issue of *Nature*, Zheng, *et al.* (*Nature* **416**, 703–709; 2002) report the crystal structure of one E3 complex, the Cul1–Rbx1–Skp1–F box SCF complex, and offer some insight into the arrangement of the players. The SCF complex consists of four subunits and represents the largest family of E3 ubiquitin-protein ligases. The largest subunit, Cul1 (green) acts as a rigid scaffold, organizing the substrate recognition and catalytic domains at opposite ends of the complex with a separation of ~100 Å. The globular Cul1 C-terminal domain and the Rbx1 (red) make up the catalytic domain. Rbx1 sits in



a V-shaped groove formed by the Cul1 C-terminal domain and forms the interface with the ubiquitin-conjugating E2 enzyme (brown, transparent).

At the opposite end of Cul1, the N-terminal tip of the Cul1 stalk is the binding site for the Skp1–F box. Skp1 (blue) is an adapter protein linking Cul1 to the F box motif (pink) of an F box protein such as Skp2 (pink, transparent) — modeled here by superimposing the previously reported structure of Skp1–Skp2. The F box protein is the site of substrate binding, and the identity of the F box protein varies to accommodate a wide range of diverse substrate proteins.

By modeling the Skp2 domain and E2 into their relative binding sites on the SCF complex, Zhang, *et al.* were able to estimate the separation between the region of substrate binding and the Ub-binding Cys residue of E2 at ~50 Å. While this gap could easily accommodate $p27^{Kip1}$, a known ubiquitination target of Skp2, it is not clear exactly how the covalent linkage is achieved. The lack of obvious regions of flexibility within the SCF architecture does not shed light on the mechanism of Ub transfer and progression of the polyUb chain. While it seems that some flexibility would be necessary to accommodate diverse substrates and Ub chain growth, the work of Zheng *et al.* suggests that rigidity might be important for SCF function.

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