

generally 4 Å apart, interact directly with the scissile phosphate, stabilize the pentacovalent intermediate, and generate the attacking hydroxide ion, as well as facilitating the departure of the 3' oxyanion⁸. If such a two-metal-ion mechanism is relevant to the 5' nuclease, the possible role, if any, of the more distant site III metal ion can only be guessed.

The source of the thermal stability of *Taq* polymerase is not obvious from structural comparison with KF, but the number of hydrogen bonds has increased by four, and two salt bridges between subdomains in the polymerase domain become hydrophobic; the ratio of leucine to isoleucine has increased by 4.4-fold, and arginine to lysine by 1.3-fold, which may result from the higher G+C content of the leucine and arginine codons (giving a more thermostable DNA), rather than an effect on the protein.

An important question concerning the pol I family of enzymes is how the polymerase and 5'-nuclease active sites work together to generate a duplex DNA product containing only a nick: the present structure raises at least as many questions as it answers, because we observe that these two active sites are separated by over 70 Å. The unusually elongated shape of the molecule seen here led us to examine its overall fold in solution. Preliminary measurements of the radius of gyration (R_g) of *Taq* polymerase by solution X-ray-scattering methods yield an experimental value of R_g that is substantially smaller than that calculated from the coordinates of the crystal structure (S.H.E. *et al.*, unpublished observations). Thus the 5' nuclease domain is not positioned in solution as shown in Fig. 2, but must be located much closer to the centre of mass of the Stoffel fragment. Presumably its orientation in these crystals is adventitious and governed by crystal-packing interactions. Two packing interactions between the 5' nuclease and neighbouring molecules bury 1,100 and 1,466 Å² of solvent-accessible area, larger than the intramolecular interaction surface. A structural basis for understanding how these two activities work together must await the crystal structure of a complex with the appropriately nicked DNA substrate. □

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1. Lawyer, F. C. *et al.* *J. Biol. Chem.* **264**, 6427–6427 (1989).
2. Ollis, D. L., Brick, P., Hamlin, R., Xuong, N. G. & Steitz, T. A. *Nature* **313**, 762–766 (1985).
3. Derbyshire, V. *et al.* *Science* **240**, 199–201 (1988).
4. Derbyshire, V., Grindley, N. D. F. & Joyce, C. M. *EMBO J.* **10**, 17–24 (1991).
5. Lundquist, R. C. & Olivera, B. M. *Cell* **31**, 53–60 (1982).
6. Gutman, P. D. & Minton, K. W. *Nucleic Acids Res.* **21**, 4406–4407 (1993).
7. Lyamichev, V., Brow, M. A. D. & Dahlberg, J. E. *Science* **260**, 778–783 (1993).
8. Freemont, P. S., Friedman, J. M., Beese, L. S., Sanderson, M. R. & Steitz, T. A. *Proc. natl. Acad. Sci. U.S.A.* **85**, 8924–8928 (1988).
9. Beese, L. S. & Steitz, T. A. *EMBO J.* **10**, 25–33 (1991).
10. Kim, E. & Wyckoff, H. H. *J. molec. Biol.* **218**, 449–464 (1991).
11. Kankare, J. *et al.* *Protein Engng* **7**, 823–830 (1994).
12. Teplyakov, A. *et al.* *Protein Sci.* **7**, 1098–1107 (1994).
13. Yang, W., Hendrickson, W. A., Crouch, R. J. & Satow, Y. *Science* **249**, 1398–1405 (1990).
14. Davies, J. F., Hostomska, Z., Homstomsky, Z., Jordan, S. R. & Matthews, D. A. *Science* **252**, 88–95 (1991).
15. Steitz, T. A. *Curr. Opin. struct. Biol.* **3**, 31–38 (1993).
16. Pelletier, H., Sawaya, M. R., Kumar, A., Wilson, S. H. & Kraut, J. *Science* **264**, 1891–1903 (1994).
17. Steitz, T. A., Smerdon, S. J., Jäger, J. & Joyce, C. M. *Science* **266**, 2022–2025 (1994).
18. Ariyoshi, M. *et al.* *Cell* **78**, 1063–1072 (1994).
19. Dyda, F. *et al.* *Science* **266**, 1981–1986 (1994).
20. Yang, W. & Steitz, T. A. *Structure* **3**, 131–134 (1995).
21. Eom, S. H. *et al.* *Acta crystallogr.* **D51** (in the press).
22. Matthews, B. C. W. *J. molec. Biol.* **33**, 491–497 (1968).
23. Fitzgerald, P. M. D. *J. appl. Crystallogr.* **21**, 273–278 (1988).
24. Brünger, A. T. *X-PLOR, version 3.1* (Yale Univ. Press, New Haven, CT, 1992).
25. Otwinowski, Z. *ML—PHARE CCP4 Proc.* 80–88 (Daresbury Laboratory, Warrington, 1991).
26. Zhang, K. Y. J. & Main, P. *Acta crystallogr.* **A46**, 377–381 (1990).
27. Jones, T. A. & Kjeldgaard, M. *O version 5.9* (Department of Molecular Biology, BMC, Uppsala Univ., Sweden, 1993).
28. Kraulis, P. J. *J. appl. Crystallogr.* **24**, 946–950 (1991).
29. Merrit, E. A. & Murphy, M. E. P. *Acta crystallogr.* **D50**, 869–873 (1994).

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ERRATUM

Crystal structure of a replication fork single-stranded DNA binding protein (T4 gp32) complexed to DNA

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AN error in the production process resulted in Fig. 1a and b of the paper by Kim *et al.* on page 613 of this issue being substituted for Fig. 2a and b of the earlier paper by Shamoo *et al.* The correct panels of Fig. 2 are shown here.

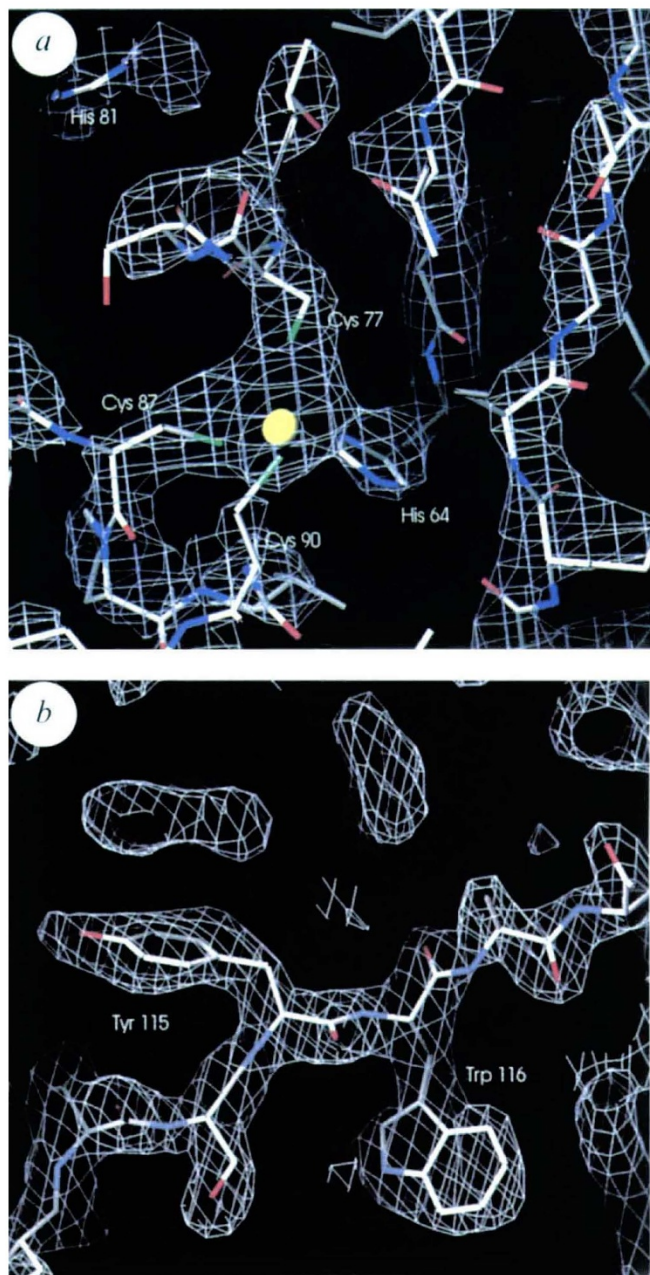


FIG. 2 a, Experimental electron density map to 3.1 Å contoured at 2.5 σ and calculated using the combined MAD and MIR phases that have been solvent flattened. The coordination of the Zn²⁺ ion (yellow) is tetrahedral with His 64, Cys 77, Cys 87 and Cys 90 as ligands. b, 2F₀ - F_c electron density map contoured at 1.3 σ showing a stretch of β -strand 4 that includes the current partly refined model and all the data to 2.2 Å.