(ref. 15 and M. McCoy, J. J. Toole, E.H.C. and R.A.W., in preparation) as well as fibroblastic tumours (L. Parada and R.A.W., unpublished results) have been found to carry oncogenes derived from activation of cellular Ki-*ras* genes. As discussed above, the N-terminal regions of the Ha-*ras* and Ki-*ras* proteins are extremely similar, and the Ki-*ras* proteins of many of these tumours also exhibit aberrant electrophoretic migration rates (manuscript in preparation). We predict that activation of many of these other oncogenes will depend on structural alterations very similar to those reported here.

Most amino acid sequence alterations are either neutral or deleterious to protein function. Few are able to actively potentiate the normal functions of a protein. We suggest that only a small number of sites on the p21 protein can be altered in a fashion leading to oncogenic activation. Most mutations will affect other residues whose alteration will be unproductive for oncogenic conversion. The target for oncogenic conversion may therefore be exceedingly small, and may even be confined to the Gly-12 codon. In such a case, the restriction site for *Nae*I, which totally spans that codon, may provide an ideal diagnostic tool for detecting a critical change in the genome.

The present data suggest that the alteration of one nucleotide in one bladder cell leads to the creation of an activated oncogene. There are three possible point mutations at this position, and it is perhaps not coincidental that the G-T transversion observed here is precisely that mutation favoured by many suspected bladder carcinogens[39]. The oncogene resulting from this mutation was probably an important determinant in the subsequent outgrowth of a lethal neoplasm. The point mutation implicated as a central event in this oncogenic transformation represents the first demonstration of a lesion in cellular DNA whose occurrence is directly related to the carcinogenic process.

We thank Dr James G. Rheinwald and Terry O'Connell (Sidney Farber Cancer Institute) for preparing cultures of normal human bladder cells; Drs B. Ballou and T. Hakala for the bladder biopsy; Dr Mark Furth for providing monoclonal anti-p21 antisera; R. Kooler and L. Donehower for help in sequencing; R. Chipperfield for assistance in constructing recombinant clones; Dr G. Zieve for photographing cell lines; Dr J. Essigmann for helpful discussions; Dr M. Ruta for communicating data before publication and S. Posner for secretarial assistance. This work was supported by grants CA26717, CA31649 and CA17537 from the USNCI and by a grant from the Whitehead Institute for Biomedical Research.

Received 13 September; accepted 5 October 1982.

1. McCann, J., Choi, E., Yamasaki, E. & Ames, B. N. *Proc. natn. Acad. Sci. U.S.A.* **72,** 5135–5139 (1975).
2. McCann, J. & Ames, B. N. *Proc. natn. Acad. Sci. U.S.A.* **73,** 950–954 (1976).
3. Bridges, B. A. *Nature* **261,** 195–200 (1976).
4. Bouck, N. & diMayorca, G. *Nature* **264,** 722–727 (1976).
5. Shih, C., Shilo, B., Goldfarb, M. P., Dannenberg, A. & Weinberg, R. A. *Proc. natn. Acad. Sci. U.S.A.* **76,** 5714–5718 (1979).
6. Cooper, G. M., Okenquist, S. & Silverman, L. *Nature* **284,** 418–421 (1980).
7. Shih, C., Padhy, L. C., Murray, M. J. & Weinberg, R. A. *Nature* **290,** 261–264 (1981).
8. Krontiris, T. G. & Copper, G. M. *Proc. natn. Acad. Sci. U.S.A.* **78,** 1181–1184 (1981).
9. Perucho, M. *et al. Cell* **27,** 467–476 (1981).
10. Lane, M. A., Sainten, A. & Cooper, G. M. *Proc. natn. Acad. Sci. U.S.A.* **78,** 5185–5189 (1981).
11. Shilo, B. & Weinberg, R. A. *Nature* **289,** 607–609 (1981).
12. Goldfarb, M., Shimizu, K., Perucho, M. & Wigler, M. *Nature* **296,** 404–409 (1982).
13. Shih, C. & Weinberg, R. A. *Cell* **29,** 161–169 (1982).
14. Pulciani, S. *et al. Proc. natn. Acad. Sci. U.S.A.* **79,** 2845–2849 (1982).
15. Der, C., Krontiris, T. G. & Cooper, G. M. *Proc. natn. Acad. Sci. U.S.A.* **79,** 3637–3640 (1982).
16. Parada, L. F., Tabin, C. J., Shih, C. & Weinberg, R. A. *Nature* **297,** 474–479 (1982).
17. Santos, E. *et al. Nature* **298,** 343–347 (1982).
18. Scolnick, E. M. & Parks, W. P. *J. Virol.* **13,** 1211–1219 (1974).
19. Shih, T. Y. *et al. J. Virol.* **27,** 45–55 (1978).
20. DeFeo, D. *et al. Proc. natn. Acad. Sci. U.S.A.* **78,** 3328–3332 (1981).
21. Chang, E. H., Gonda, M. A., Ellis, R. W., Scolnick, E. M. & Lowy, D. R. *Proc. natn. Acad. Sci. U.S.A.* 4848–4852 (1982).
22. Ellis, R. W. *et al. J. Virol.* **36,** 408–420 (1980).
23. Wu, Y-J., *et al. Cell* (submitted).
24. Ellis, R. W., DeFeo, D., Furth, M. E. & Scolnick, E. M. *Cell* (in the press).
25. Furth, M. E., Davis, L. J., Fleurdelys, B. & Scolnick, E. M. *J. Virol.* **43,** 294–304 (1982).
26. Mulligan, R. & Berg, P. *Proc. natn. Acad. Sci. U.S.A.* **78,** 2072–2076 (1981).
27. Wigler, M. *et al. Cell* **16,** 777–785 (1979).
28. Shih, T. Y. *et al. J. Virol.* **42,** 253–261 (1982).
29. Seif, I., Khoury, G. & Dhar, R. *Nucleic Acids Res.* **8,** 2225–2238 (1980).
30. Maxam, A. H. & Gilbert, W. *Proc. natn. Acad. Sci. U.S.A.* **74,** 560–564 (1977).
31. Dhar, R. *et al. Science* **217,** 934–936 (1982).
32. Shih, T. Y., Weeks, M. O., Young, H. A. & Scolnick, E. M. *Virology* **96,** 64–79 (1979).
33. Tsuchida, N., Ryder, T. & Ohtsubo, E. *Science* **217,** 937–939 (1982).
34. Cantor, C. R. & Schimmel, P. R. in *Biophysical Chemistry* Vol. 1, 303–304 (Freeman, San Francisco, 1980).
35. Neel, B. G., Hayward, W. S., Robinson, H. L., Fang, J. & Astrin, S. M. *Cell* **23,** 323–334 (1981).
36. Payne, G. S. *et al. Cell* **23,** 311–322 (1981).
37. Blair, D. G. *et al. Science* **212,** 941–943 (1981).
38. Chang, E. J., Furth, M. E., Scolnick, E. M. & Lowy, D. R. *Nature* **297,** 479–483 (1982).
39. Swenson, D. H. & Kadlubar, F. F. in *Microbial Testers Probing for Carcinogenesis* (ed. Felkner, I. C.) 3–33 (Dekker, New York, 1981).
40. Varmus, H. E., Quintrell, N. & Ortiz, S. *Cell* **25,** 23–36 (1981).
41. Rigby, P. W. *et al. J. molec. Biol.* **13,** 237–251 (1977).
42. Wahl, G. M., Stern, M. & Stark, G. R. *Proc. natn. Acad. Sci. U.S.A.* **76,** 3683–3687 (1979).
43. Manley, J. L. *et al. Proc. natn. Acad. Sci. U.S.A.* **77,** 3855–3859 (1980).
44. Graham, F. L. & van der Eb, A. J. *Virology* **52,** 456–471 (1973).
45. Andersson, P., Goldfarb, M. P. & Weinberg, R. A. *Cell* **16,** 63–75 (1979).

A point mutation is responsible for the acquisition of transforming properties by the T24 human bladder carcinoma oncogene

E. Premkumar Reddy, Roberta K. Reynolds, Eugenio Santos & Mariano Barbacid

Laboratory of Cellular and Molecular Biology, National Cancer Institute, National Institutes of Health, Bethesda, Maryland 20205, USA

The genetic change that leads to the activation of the oncogene in T24 human bladder carcinoma cells is shown to be a single point mutation of guanosine into thymidine. This substitution results in the incorporation of valine instead of glycine as the twelfth amino acid residue of the T24 oncogene-encoded p21 protein. Thus, a single amino acid substitution appears to be sufficient to confer transforming properties on the gene product of the T24 human bladder carcinoma oncogene.

DNA-MEDIATED gene transfer techniques have made it possible to identify the presence of dominant transforming genes (oncogenes) in a variety of human tumours (for review see ref. 1). Although only a small number of human tumour DNAs have been shown to be capable of transforming normal cells in transfection assays, oncogenes have been detected in tumours representative of each of the major forms of human cancer[2–8]. To date, more than 10 different human oncogenes have been identified. One of them, present in T24 and EJ bladder carcinoma cell lines, has been isolated by molecular cloning techniques[8–10]. Preliminary characterization of this oncogene has revealed that it is small (less than 4.6 kilobase, kb), and that it has not undergone major genetic rearrangements[9–11]. Comparative analysis of this bladder carcinoma oncogene with retroviral transforming (*onc*) genes has revealed that an internal fragment of the T24 oncogene is closely related to the *onc* genes of

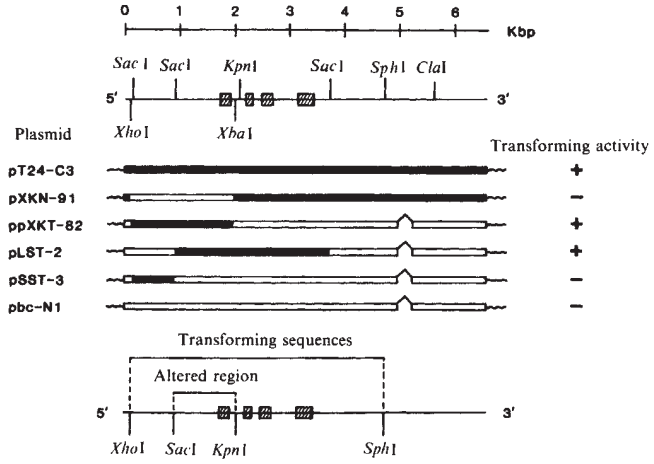

Fig. 1 Location of the region of the c-*has/bas*-1 gene that underwent the genetic alterations that led to the activation of the T24 oncogene. Hybrid plasmids containing sequences derived from pT24-C3 (■) and pbc-N1 (□) were tested for their biological activity. pT24-C3, pXKT-82 and pLST-2 transformed NIH 3T3 cells in transfection assays[8], with specific transforming activities of 5×10^4, 1×10^4 and 0.6×10^4 ffu pmol^{-1}, respectively. In contrast, pbc-N1, pXKN-91 and pSST-3 exhibited no transforming activity. The hatched boxes located within the restricted endonuclease map shown in the top of the figure represent the predicted exons of the T24 oncogene and its normal homologue, the c-*has/bas*-1 gene. Deletions (⋀), and plasmid sequences (〰), are also indicated in the diagram. See text for definition of the regions encompassing the transforming sequences and the activated domain of the T24 oncogene.

Harvey and BALB murine sarcoma viruses (designated v-*has* and v-*bas*, respectively)[11–13]. Characterization of human DNA sequences homologous to v-*has* and v-*bas* has demonstrated that the normal c-*has*/c-*bas*-1 gene is in fact an allele of the T24 oncogene[11–14]. Moreover, we found that these genes, despite their different biological properties, were indistinguishable by both heteroduplex and restriction enzyme analysis[11–13]. In an effort to understand the molecular events involved in human carcinogenesis, studies were undertaken to determine the number and nature of the genetic alterations that have resulted in the activation of the T24 human bladder carcinoma oncogene.

A domain within the T24 oncogene responsible for its activation

Previously, we have reported that a plasmid (pT24-C3) known to contain a 6.6-kb *Bam*HI fragment of T24 cellular DNA was capable of transforming NIH 3T3 mouse cells with a specific activity of 5×10^4 focus-forming units (ffu) per pmol. A restriction endonuclease map of this DNA fragment is depicted in Fig. 1. The transforming activity of this molecule was localized to a region within the 4.6-kb *Xho*I–*Sph*I DNA fragment based on the observations that cleavage with these two restriction endonucleases did not affect the transforming activity of pT24-C3 (ref. 11). To establish the region of the normal human c-*has/bas*-1 gene that underwent the genetic alterations leading to its malignant activation in T24 bladder carcinoma cells, we constructed a series of plasmids in which fragments of the T24 transforming gene were substituted with the homologous normal sequences of c-*has/bas*-1. In reciprocal experiments, regions of the normal c-*has/bas*-1 gene were replaced by the corresponding domains of its transforming allele. Figure 1 summarizes the plasmids constructed in the present studies as well as their respective biological activities. Substitution in pT24-C3 of a DNA fragment located between the *Xho*I and *Kpn*I cleavage sites by normal sequences, completely abolished its transforming activity. In contrast, replacement of the normal *Xho*I–*Kpn*I region of pbc-N1, a plasmid that contains the 6.4-kb *Bam*HI DNA clone of the normal human c-*has/bas*-1 gene, by the corresponding domain of pT24-C3, generated a plasmid (pXKT-82) that transformed NIH 3T3 mouse cells with an efficiency of 10^4 ffu pmol^{-1}.

Two additional plasmids were generated by replacing the internal 0.7-kb and 3.0-kb *Sac*I DNA fragments of pbc-N1 by the corresponding regions of the T24 oncogene. As shown in Fig. 1, only the latter plasmid (pLST-2) was able to induce malignant transformation on transfection to NIH 3T3 cells. These results, taken together, establish that the genetic alterations that activated the T24 oncogene are located within the 0.9-kb *Sac*I–*Kpn*I DNA fragment of pT24-C3 DNA.

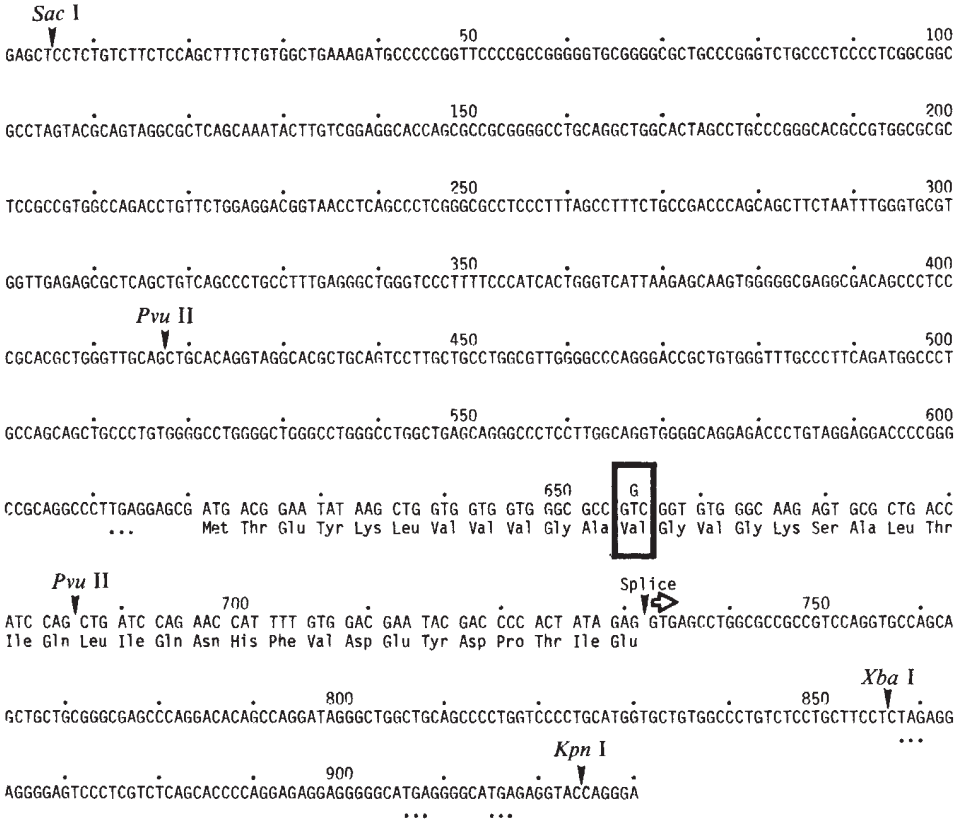

Fig. 2 Comparative sequence analysis of the 0.93-kb *Sac*I–*Kpn*I DNA fragment of the T24 oncogene and its normal human homologue c-*has/bas*-1. The upper line shows the sequence of the T24 oncogene proceeding in the 5′ to 3′ direction with respect to the polarity of the closely related v-*has* and v-*bas* retroviral mRNAs. The corresponding sequence of the 0.93-kb *Sac*I–*Kpn*I DNA fragment of c-*has/bas*-1 has also been determined and found to be identical to that of the T24 oncogene with the exception of the nucleotide located at position 653, where we detected a G instead of a T. These nucleotide chains result in the incorporation of glycine, instead of valine, at position 12 in the p21 protein coded for by the c-*has/c-bas*-1 gene. Termination codons are indicated (···). The nucleotide sequence analysis was performed according to the procedures of Maxam and Gilbert[23].

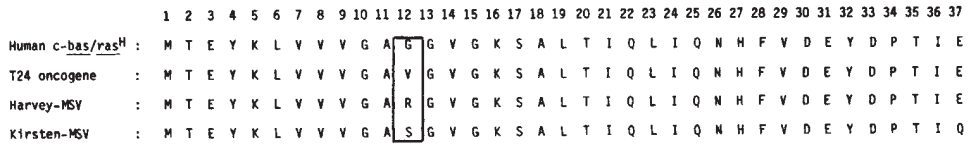

Fig. 3 Predicted amino acid sequence of the first exon of the normal human c-*has/bas*-1 gene and its transforming allele, the T24 oncogene. Comparison with the first 37 amino acids predicted for the p21 proteins coded for by Harvey-MSV[15] and Kirsten-MSV[18]. The letter code for the amino acids is: A, Ala; D, Asp; E, Glu; F, Phe; G, Gly; H, His; I, Ile; K, Lys; L, Leu; M, Met; N, Asn; P, Pro; Q, Gln; R, Arg; S, Ser; T, Thr; V, Val; and Y, Tyr.

Comparative sequence analysis of the 0.9-kb *Sac*I–*Kpn*I DNA fragments of pT24-C3 and pbc-N1

The nucleotide sequence of the 0.9-kb *Sac*I–*Kpn*I DNA fragments of the T24 oncogene and its normal homologue was next established. To our surprise there was only a single (nucleotide) change, located at position 655 from the *Sac*I cleavage site. Figure 2 shows that the guanosine residue present in the normal c-*has/bas*-1 gene has been changed into a thymidine residue in the T24 oncogene. Thus the activation of the c-*bas/ras*-1 gene in T24 human bladder carcinoma cells was caused by a single point mutation.

The mutated guanosine residue was found to be part of the hexanucleotide sequence GCCGGC, which is specifically recognized by the restriction endonuclease *Nae*I. Thus, we used this enzyme to verify the results obtained by nucleotide sequence analysis. We observed that *Nae*I cleaved the 6.4-kb *Bam*HI insert of pbc-N1 five times, yielding four internal DNA fragments, of sizes 0.2, 0.4, 0.6 and 0.9 kb. In contrast, digestion of pT24-C3 with *Nae*I yielded only three internal fragments, of sizes 0.2, 0.6 and 1.3 kb (data not shown). The loss of the *Nae*I cleavage site within the 1.3-kb internal fragment confirms the nucleotide sequence shown in Fig. 2.

Heteroduplex analysis of the T24 oncogene and its normal allele with v-*has* and v-*bas* had indicated that the putative coding sequences of the human genes are distributed among four exons, of which only the first one lies within the 0.9-kb *Sac*I–*Kpn*I fragment[11,14]. We have sequenced the entire coding region of the T24 oncogene and identified the four exons by comparison of these sequences with those of v-*has*[15] and v-*bas*[16]. This sequence analysis demonstrated that the first exon ends at position 731 from the *Sac*I cleavage site (Fig. 2). At this position, we detected the consensus donor splice sequence AGAGG[17]. Thus, in agreement with previous heteroduplex analysis and restriction enzyme data[11,14], the first exon of the T24 oncogene contained the coding information for 37 amino acids extending from position 621 to 731. These observations located the mutated G residue at the second base of a triplet coding for glycine, the predicted twelfth amino acid residue of the normal human p21 protein. Substitution of this guanosine residue for thymidine results in the change of the glycine-coding triplet GGC to GTC, which codes for valine. Thus, a single amino acid substitution (Gly→Val) seems to be sufficient to confer transforming properties to the gene product of the T24 human bladder carcinoma oncogene.

A common genetic alteration in human and retroviral transforming genes

The amino acid sequences of Harvey and BALB-MSV-encoded p21 proteins have been identified by nucleotide sequence analysis of their corresponding *onc* genes, v-*has* and v-*bas*, respectively[15,16]. Comparison of the first 37 amino acids of the p21 protein coded for by the normal c-*has/bas*-1 human gene with their retroviral counterparts shows complete identity except at position 12, the same amino acid residue that is altered in the T24 oncogene (Fig. 3). Whereas arginine is present in Harvey-MSV p21[15], lysine was found in BALB-MSV p21[16]. Interestingly, the normal rat homologue of human c-*has/bas*-1, the gene that presumably recombined with Moloney-MuLV to generate Harvey-MSV, also possesses glycine at position 12 (M. Ruta, personal communication). These results suggest that mutations affecting the coding properties of the twelfth codon of normal c-*has/bas*-1 genes may be sufficient for the acquisition of malignant properties.

Recently, Tsuchida *et al.* have also reported the nucleotide sequence of v-*kis*, the *onc* gene of Kirsten-MSV[18]. When the first 37 amino acids of the deduced sequence of Kirsten p21 were compared with those of c-*has/bas*-1, only two changes were detected: Gly→Ser in position 12, and Glu→Gln in position 37. Thus, it is possible that changes in the glycine residue at position 12 may have also activated c-*kis* genes. This hypothesis would predict that another human transforming oncogene, present in a large variety of human carcinomas and known to contain the human c-*kis*-2 gene,[5,6,8,12,14,19] has been activated by the same mechanism that originated the T24 oncogene.

Activation of the human c-*has/bas*-1 gene: a qualitative or quantitative mechanism?

That each of the known p21 transforming proteins has a different amino acid at residue 12 suggests that their malignant properties are the result of the elimination of the glycine residue present in their normal counterparts, rather than due to a specific transition from one type of amino acid residue to another. Glycine residues are known to alter the α-helical structure of proteins. We compared the predicted secondary structures of the p21 proteins coded for by the T24 oncogene and its normal human homologue using the computerized method of Garnier *et al.*[20]. Such an analysis revealed that the N-terminal region of the c-*has/bas*-1-encoded p21 protein contains two α-helical regions of 10 and 13 amino acids joined by a hinge region of 7 amino acids that provides flexibility for folding. When the glycine at position 12 is replaced by valine in the transforming p21 protein, the size of the hinge region decreases to a stretch of two amino acids, resulting in an increased helical content of the N-terminal region of this protein. This change should result in the prominent projection of the N-terminal domain of transforming p21 proteins away from the central core of the molecule.

The biological significance of the structural alteration of human and retroviral p21 transforming proteins remains to be determined. Shih *et al.* have recently reported that the Harvey-MSV p21 protein is processed before its association with membranous structures[21]. Whether the presence of a glycine residue in position 12 confers on the normal cellular p21 protein a configuration that affects either its processing or its association with putative cellular targets remains to be determined. Whatever the mechanism might be, our findings indicate that the difference between the normal c-*has/bas*-1 and the T24 oncogene-encoded p21 proteins is of a qualitative nature.

However, it is also possible that quantitative alterations in the expression of the normal p21 protein induce malignant transformation. Chang *et al.* have shown that the normal c-*has/bas*-1 can transform NIH 3T3 mouse cells when this gene comes under the regulatory influence of a retroviral LTR[22]. In this case high levels of normal p21 protein can be detected in the transformed cells. Thus it is possible that increasing amounts of normal p21 protein may overcome the structural limitations imposed by the presence of a glycine residue at position 12. However, the possibility that over-expression of the normal c-*has/bas*-1 genes may induce morphological transformation by a different mechanism than that of their transforming counterparts cannot be ruled out at present.

Implications

The present studies have located the genetic changes leading to the activation of the T24 bladder carcinoma oncogene within a 0.9-kb *Sac*I–*Kpn*I DNA fragment of the human c-*has/bas*-1 gene. This region contains some 5′ putative regulatory sequences, as well as the first exon (Fig. 2). Comparative sequence analysis of these 0.9-kb DNA fragments derived from the T24 oncogene and its normal counterpart revealed a single base pair difference: a point mutation (G→T) within the exon sequences that led to the substitution of a glycine residue in the normal c-*has/bas*-1 gene-encoded p21 protein to a valine residue in the corresponding translational product of the T24 oncogene. These results, taken together, indicate that a single point mutation was responsible for the activation of this oncogene in T24 human bladder carcinoma cells.

These findings may have important practical applications. The mutated guanosine residue is part of a sequence (GCCGGC) recognized by the restriction endonucleases *Nae*I (GCCGGC), *Hpa*II (CCGG) and *Msp*I (CCGG). The G→T change observed in the T24 oncogene eliminated the cleavage site for these three enzymes. Neither *Nae*I nor *Hpa*II appear to cleave this sequence in human DNA, probably due to DNA methylation. However it is possible that *Msp*I, which recognizes the CCGG sequence whether methylated or not, could be used for the screening of human DNAs for the presence of T24-like oncogenes. Similarly, antibodies capable of specifically recognizing the altered domain of transforming p21 proteins may also be used to detect activated c-*has/bas* genes.

Demonstration that a single genetic change can activate a human transforming gene poses the dilemma as to how to relate these observations with the widely accepted multi-stage model for the development of human neoplasias. It is possible that activation of the T24 oncogenes may represent a late, irreversible step in oncogenesis. Support for this hypothesis comes from the fact that NIH 3T3 mouse cells, the cells used to identify this oncogene, are thought to be pre-neoplastic rather than normal cells. However, the possibility that this oncogene may have a role in the onset of certain human cancers cannot be excluded at present.

Development of biochemical and immunological reagents capable of specifically detecting the presence of the T24 bladder carcinoma oncogene (or its gene product) in both naturally and experimentally induced neoplasms should help to define the role of this dominant transforming gene in carcinogenesis.

We thank Richard Feldmann for help in the computer analysis of the p21 proteins, Anne V. Lauver and Linda K. Long for technical assistance, and S. A. Aaronson for support.

Received 23 September; accepted 8 October 1982.

1. Cooper, G. M. *Science* **217,** 801–806 (1982).
2. Krontiris, T. G. & Cooper, G. M. *Proc. natn. Acad. Sci. U.S.A.* **78,** 1181–1184 (1981).
3. Shih, C., Padhy, C., Murray, M. & Weinberg, R. A. *Nature* **290,** 261–264 (1981).
4. Lane, M. A., Sainten, A. & Cooper, G. M. *Proc. natn. Acad. Sci. U.S.A.* **78,** 5185–5189 (1981).
5. Murray, M. J. *et al. Cell* **25,** 355–361 (1981).
6. Perucho, M. *et al. Cell* **27,** 467–476 (1981).
7. Lane, M. A., Sainten, A. & Cooper, G. M. *Cell* **28,** 873–880 (1982).
8. Pulciani, S. *et al. Proc. natn. Acad. Sci. U.S.A.* **79,** 2845–2849 (1982).
9. Goldfarb, M., Shimizu, K., Perucho, M. & Wigler, M. *Nature* **296,** 404–409 (1982).
10. Shih, C. & Weinberg, R. A. *Cell* **29,** 161–169 (1982).
11. Santos, E., Tronick, S. R., Aaronson, S. A., Pulciani, S. & Barbacid, M. *Nature* **298,** 343–347 (1982).
12. Der, C. J., Krontiris, T. G. & Cooper, G. M. *Proc. natn. Acad. Sci. U.S.A.* **79,** 3637–3640 (1982).
13. Parada, L. F., Tabin, C. J., Shih, C. & Weinberg, R. A. *Nature* **297,** 474–478 (1982).
14. Chang, E. H., Gonda, M. A., Ellis, R. W., Scolnick, E. M. & Lowy, D. R. *Proc. natn. Acad. Sci. U.S.A.* **79,** 4848–4852 (1982).
15. Dhar, R. *et al. Science* **217,** 934–937 (1982).
16. Reddy *et al.* (in preparation).
17. Sharp, P. A. *Cell* **23,** 643–646 (1981).
18. Tsuchida, N., Ryder, T. & Ohtsubo, E. *Science* **217,** 937–939 (1982).
19. Pulciani, S., Santos, E., Lauver, A. V., Long, L. K. & Barbacid, M. *Nature* (in the press).
20. Garnier, J., Osguthorpe, D. J. & Robson, B. *J. molec. Biol.* **120,** 97–120 (1978).
21. Shih, T. Y. *et al. J. Virol.* **42,** 253–261 (1982).
22. Chang, E. H., Furth, M. E., Scolnick, E. M. & Lowy, D. R. *Nature* **297,** 479–483 (1982).
23. Maxam, A. M. & Gilbert, W. *Proc. natn. Acad. Sci. U.S.A.* **74,** 560–564 (1977).

LETTERS TO NATURE

UV observations of TT Arietis and the magnetic rotator hypothesis

R. F. Jameson, M. R. Sherrington, A. R. King & J. Frank

Astronomy Department, University of Leicester, Leicester LE1 7RH, UK

TT Arietis, one of the brightest cataclysmic variables, $m_v \sim 10.2 \rightarrow 11.0$, is usually classed as a novalike variable, but after a recent dip in brightness to $m_v \sim 14.5$, Krautter *et al.*[1] suggested it to be a dwarf nova in an extended high state. Photometric light curves[2–4] generally show a sinusoidal light variation with a period of 0.1329 days and no eclipses[5]. Cowley *et al.*[6] have measured radial velocity curves and find a period of 0.13755 days. We have made[7] IR measurements of the system from which we deduce a distance for TT Ari of 100 ± 20 pc based on a colour change for the system from V to K of +0.4 while the system is faint to $V-K=-0.17$ when brighter. The red colour is caused by dilution of the primary 2.2-μm light by the light of the secondary, giving $m_K = 11.75^{+0.45}_{-0.25}$ for the secondary star; Bailey's method[8] then leads to the distance. The sinusoidal light variations have generally been interpreted as due to a hot spot on the edge of an accretion disk. Since good quality IUE spectra can be made with exposure times of about 15 min, we decided to observe as many IUE spectra as possible round the binary period with a view to studying the behaviour of the hot spot at UV wavelengths. We now report that the variations at these and X-ray wavelengths round the orbital cycle rule out a hot spot/accretion disk model for TT Ari. Instead we propose that it has a magnetized white dwarf primary rotating with the photometric period of 0.1329 days.

Four short and four long wavelength low dispersion spectra were obtained with the IUE satellite spectrometer on 9 January, 1982. Between each exposure the fine error sensor (FES) was used to measure the white light variation of TT Ari. The spectra taken nearest to maximum light, are shown in Fig. 1, together with an optical spectrum measured by M. J. Ward with the Anglo Australian Telescope. Although the spectra appear noisy, most of the features are repeatable from one spectrum to the next. The strongest features are: Lyα 1,216 e (geocoronal?); N V 1,238 a; Si IV 1,393 a; C IV 1,549 e (2.9); He II 1,640 a; and Mg II 2795 e (2.1) where 'a' denotes absorption, 'e' emission and the number in parentheses the variation in line strength (maximum/minimum). The variation of the emission lines roughly follows that of the continuum variation. Regions as free as possible of absorption or emission lines were used to measure the continuum light variations. The continuum was averaged over a bandwidth of 50 Å. The only significant errors in the measurements are due to the photometric accuracy of the IUE cameras, namely $\lesssim \pm 10\%$.

Figure 2 shows the four point continuum light curves at 1,265, 1,700, 2,110, 2,510 and 3,010 Å, together with the nine point white light FES light curve. The amplitude of the white light curve corresponds to $m_v = 11.86 \rightarrow 12.36$; thus TT Ari was unusually faint. The solid curves are an attempt to fit a curve of the same shape as the white light curve but with variable amplitude through the UV points. The main difficulty in getting a good fit to the UV points is that the FES points represent ~1 min of integration and are thus prone to the flickering whereas the UV points result from spectra with 15-min exposure times. Whichever numbers are used for the amplitude of the variations the trend is clear; maximum amplitude being found for white light and the hard UV with a minimum variation in between at 3,010 Å.

0028-0836/82/450152—04$01.00