13, 1146 (1979); R. Risebrough, B. Delappe, B. Walker, in *Marine Pollutant Transfer*, H. Windom and R. Duce, Eds. (Skidaway Institute of Oceanography, Savannah, GA, 1976), pp. 261–321; G. R. Harvey and W. G. Steinhauer, in *Environmental Biogeochemistry*, J. Nriagu, Ed. (Ann Arbor Science, Ann Arbor, MI, 1976); W. Kramer *et al.*, *Chemosphere* **13**, 1255 (1984).
4. National Research Council, *Disposal of Industrial and Domestic Wastes: Land and Sea Alternatives* (National Academy Press, Washington, DC, 1984).
5. C. N. Statham, C. R. Elcombe, S. P. Szyjka, J. J. Lech, *Xenobiotica* **8**, 65 (1978); J. J. Stegeman, A. V. Klotz, B. R. Woodin, A. M. Pajor, *Aquat. Toxicol.* **1**, 197 (1981); M. O. James and J. R. Bend, *Environ. Prot. Agency Rep. 600/9-82-013* (1982), p. 172.
6. J. F. Payne, *Science* **191**, 945 (1976); J. J. Stegeman, A. V. Klotz, B. R. Woodin, A. M. Pajor, *Aquat. Toxicol.* **1**, 197 (1981); G. L. Foureman, N. B. White, J. R. Bend, *Can. J. Fish. Aquat. Sci.* **40**, 854 (1983); R. L. Binder and J. J. Lech, *Fundam. Appl. Toxicol.* **4**, 1042 (1984).
7. *Coryphaenoides armatus* were collected from a depth of 3200 m near 38°35′N, 69°58′W (near Hudson Canyon) and from a depth of 1700 to 2300 m near 45°25′N, 48°35′W (near Carson Canyon). Livers were excised and frozen in liquid N_2 within 20 minutes of retrieval and held at −80°C until microsome preparation.
8. R. R. Wilson and R. S. Waples, *Mar. Biol.* **80**, 227 (1984).
9. J. J. Stegeman, *Can. J. Fish. Aquat. Sci.* **40**, 78 (1983).
10. A. V. Klotz, J. J. Stegeman, C. Walsh, *Arch. Biochem. Biophys.* **226**, 578 (1983); D. F. Williams and D. R. Buhler, *Biochem. Pharmacol.* **33**, 3743 (1984); A. V. Klotz, J. J. Stegeman, C. Walsh, *Mar. Environ. Res.* **14**, 402 (1984).
11. F. J. Weibel, J. C. Leutz, L. Diamond, H. V. Gelboin, *Arch. Biochem. Biophys.* **144**, 76 (1971).
12. Data presented at the Third International Conference on Microsomes and Drug Oxidations, Brighton, England (1984), describing strong inhibition of BNF-induced AHH and EROD activities in several teleost species by antibodies to cytochrome P-450E from the marine fish scup (*Stenotomus chrysops*). Only the catalytic activities induced by MC or by BNF were inhibited. At the same ratios of IgG to P-450, both EROD and AHH were inhibited about 95 percent, ECOD was inhibited about half as much, and aminopyrine metabolism was not inhibited (J. J. Stegeman, B. R. Woodin, S. S. Park, P. J. Kloepper-Sams, H. V. Gelboin, *Mar. Environ. Res.*, in press).
13. Precipitin formation on double-diffusion analysis was evident only with cytochrome P-450E and not other isozymes from scup. Antibodies to P-450E reacted only with P-450E on immunoblot of the five forms of P-450 representing the total complement resolved from scup liver. A single band in the P-450 region coincident with P-450E (54.3K) was recognized in BNF-induced scup. Cross-reactivity of antibodies to P-450E was indicated by strong reaction with a single band in liver microsomes of BNF-treated trout (*Salvelinus fontinalis*). The same amount of microsomal P-450 from control trout had barely detectable staining.
14. The lesser inhibition of EROD activity suggests a lower affinity of antibodies to P-450E for the enzyme in *C. armatus*.
15. Chlorobiphenyls and chlorinated camphenes were identified by glass capillary GC and glass capillary GC-MS (EI and pulsed positive-negative ion chemical ionization with CH_4). Chlorobiphenyls were quantified by glass capillary GC by using response curves generated by a standard of each chlorobiphenyl. An SE-52 column (J & W Scientific) installed in a Carlo Erba Model 2150 gas chromatograph equipped with a split-splitless injector and ^{63}Ni electron-capture detector interfaced with a Columbia Scientific Instruments Supergrator 3 electronic integrator was used. A Finnegan 4510 quadrupole mass spectrometer interfaced with a Carlo Erba 4160 gas chromatograph and a Finnegan INCOS 2300 data system and standard EI/CI ion source and PPNICI accessory was used for GC-MS analyses. Further details are available from the authors. Abbreviations: *pp′*-DDE: 1,1′-(2,2-dichloroethenylidene)bis[4-chlorobenzene]; *pp′*-DDD: 1,1′-(2,2-dicloroethylidene)bis[4-chlorobenzene].
16. A median effective dose for induction of AHH activity by polychlorinated biphenyls in larval *Fundulus heteroclitus* was 1.4 ppm, measured as Aroclor 1254 in the organism [R. L. Binder, J. J. Stegeman, J. J. Lech, *Chem.-Biol. Interact.* **55**, 185 (1985)].
17. S. Bandiera, S. Safe, A. B. Okey, *ibid.* **35**, 259 (1982).
18. J. J. Stegeman, in *Polycyclic Aromatic Hydrocarbons and Cancer*, H. V. Gelboin and P. O. P. T'so, Eds. (Academic Press, New York, 1981), vol. 3, pp. 1–60.
19. G. N. Somero and J. F. Siebenaller, *Nature (London)* **282**, 100 (1979).
20. W. L. Backes, M. Hogaboom, W. J. Canady, *J. Biol. Chem.* **257**, 4063 (1982); B. Bösterling and J. R. Trudell, *ibid.*, p. 4783.
21. G. N. Somero, J. F. Siebenaller, P. W. Hochachka, in *The Sea*, G. T. Rowe, Ed. (Wiley, New York, 1983), pp. 261–330.
22. S. S. Park, A. V. Klotz, H. V. Gelboin, J. J. Stegeman, *Pharmacologist* **26**, 124 (1984).
23. H. Towbin, T. Staehelin, J. Gordon, *Proc. Natl. Acad. Sci. U.S.A.* **76**, 4350 (1979).
24. M. D. Mullin *et al.*, *Environ. Sci. Technol.* **18**, 468 (1984).
25. We thank R. L. Haedrich, G. N. Somero, and E. Copeland for assistance in collecting *C. armatus* and B. R. Woodin, J. T. Tirnauer, N. M. Frew, C. Johnson, and A. Davis for laboratory assistance. Supported by NSF grants OCE80-18569 and OCE83-10505, NOAA grant WASC-8300220, the Richard King Mellon Foundation, and by a Woods Hole Oceanographic Institution independent study award. This is Woods Hole Oceanographic contribution 6014.

19 August 1985; accepted 20 December 1985

Characterization of Highly Immunogenic p66/p51 as the Reverse Transcriptase of HTLV-III/LAV

FULVIA DI MARZO VERONESE, TERRY D. COPELAND, ANTHONY L. DEVICO, RUKHSANA RAHMAN, STEPHEN OROSZLAN, ROBERT C. GALLO, M. G. SARNGADHARAN

Approximately 80 percent of all human sera that react with antigens of HTLV-III, the etiologic agent of the acquired immune deficiency syndrome (AIDS), recognize protein bands at 66 and 51 kilodaltons. A mouse hybridoma was produced that was specific to these proteins. Repeated cloning of the hybridoma did not separate the two reactivities. The p66/p51 was purified from HTLV-III lysates by immunoaffinity chromatography and subjected to NH_2-terminal Edman degradation. Single amino acid residues were obtained in 17 successive degradation cycles. The sequence determined was a perfect translation of the nucleotide sequence of a portion of the HTLV-III *pol* gene. The purified p66/51 had reverse transcriptase activity and the monoclonal immunoglobulin G specifically removed the enzyme activity from crude viral extract as well as purified enzyme.

HUMAN T-CELL LYMPHOTROPIC VIRUS (HTLV-III/LAV) is the etiologic agent of the acquired immune deficiency syndrome (AIDS) (*1*). By the use of the Western blot technique, several antigens in lysates of purified HTLV-III have been recognized as major targets of antibody reactivity with sera of HTLV-III infected individuals (*2*). Antigens of 120, 66, 51, 41, 31, 24, and 17 kD are recognized in these blots (Fig. 1A). Of these antigens, those of 17 and 24 kD (p17 and p24) have been identified as proteins encoded by the HTLV-III *gag* gene, and gp120 and gp41 as products of the *env* gene (*3, 4*).

The nature of p51 and p66 remains to be determined. Reactivity to these proteins is present in at least 80 percent of randomly tested human sera that are HTLV-III antibody positive. These proteins are antigenically unrelated to HTLV-III *gag* proteins p24 and p17, or *env* proteins gp120 and gp41 (*5*). If one assumes that these proteins are encoded by the virus, they may be derived from a long open reading frame (LOR) other than the LOR's of the *gag* and *env* genes.

In one approach to this question we prepared hybridomas secreting monoclonal antibodies that recognized these two proteins. BALB/c mice were immunized by several intraperitoneal injections of detergent-lysates of purified HTLV-III strain 3B produced in H9 cells (*6*). Splenic lymphocytes were fused with the NS-1 mouse myeloma line as described (*7*). Supernatant fluids from the hybrids obtained were screened for antibody to HTLV-III proteins by an enzyme-linked immunosorbent assay (ELISA) in which we used detergent-disrupted HTLV-III as antigen. Hybrids that were scored positive in this assay were expanded and tested further by the immunoblot technique to define specificities.

F. di Marzo Veronese, A. L. DeVico, R. Rahman, M. G. Sarngadharan, Department of Cell Biology, Bionetics Research, Inc., Rockville, MD 20850.
T. D. Copeland and S. Oroszlan, Laboratory of Molecular Virology and Carcinogenesis, LBI-Basic Research Program, NCI–Frederick Cancer Research Facility, Frederick, MD 21701.
R. C. Gallo, Laboratory of Tumor Cell Biology, National Cancer Institute, Bethesda, MD 20205.

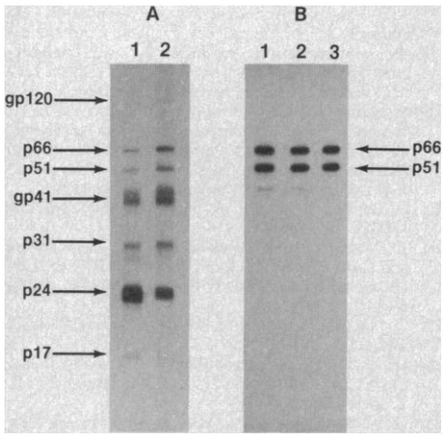

Fig. 1. Detection of p66/p51 by HTLV-III–positive human sera and mouse monoclonal antibody in the immunoblot assay. The assay was performed as described (*3*). (A) Lanes 1 and 2, sera from AIDS patients. (B) Lanes 1, 2, and 3, supernatant fluids from three individual clones of M3364.

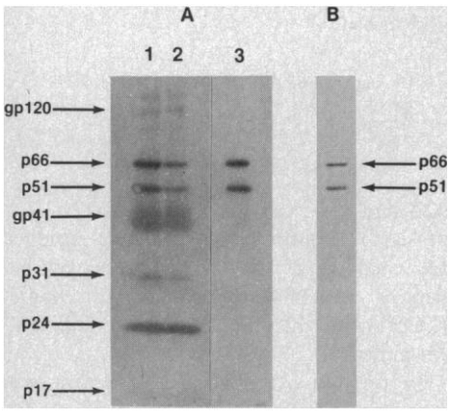

Fig. 2. Purification of p66/p51 by immunoaffinity chromatography. Purified IgG from M3364 (4 mg) was coupled to activated CH-Sepharose (5 ml) by incubation for 16 hours at 4°C in 10 ml of 0.1*M* sodium bicarbonate buffer, *p*H 8, containing 0.5*M* NaCl. The gel was washed and further incubated for 1 hour at room temperature in 10 ml of a 5% suspension of nonfat dry milk in phosphate-buffered saline (PBS) containing 0.01% Antifoam A and 0.001% Merthiolate to block remaining protein binding sites on the gel. The gel was extensively washed and packed into a column and was equilibrated with PBS containing 0.1% Triton X-100. An extract of HTLV-III was applied to the IgG-Sepharose column. The unbound proteins were collected and the column was washed with the equilibration buffer. Proteins bound to the column were eluted with 0.2*M* glycine, *p*H 2.8, containing 0.1 m*M* PMSF and 4-ml fractions were collected. (A) Western blot profiles on the unfractionated sample (lane 1), the fraction that did not bind to the column (lane 2), and a fraction eluted from the column with glycine buffer (lane 3). The antibody source was a serum from an AIDS patient. (B) A sample identical to the one used in lane 3 of (A) was analyzed by acrylamide gel electrophoresis and stained with Coomassie blue.

One hybridoma, designated M3364, was found to secrete antibodies recognizing both p66 and p51 in HTLV-III preparations (Fig. 1B). The antibody did not react with uninfected H9 cells. M3364 was subjected to repeated cycles of cloning in attempts to separate the two reactivities. Even after the fifth cloning, seeding only one cell every three wells, the two antibody reactivities could not be separated. This indicated that p66 and p51 have common determinants. Ascitic fluids, produced in pristane-treated BALB/c mice by intraperitoneal injection of M3364 cells, were used to further characterize the antibody and the proteins reactive with it. By double-immunodiffusion analysis M3364 was shown to secrete immunoglobulins of the IgG_1 subclass. Immunoblot analysis of cross-reactive epitopes of M3364 with HTLV-I and -II produced negative results, defining the specificity of the monoclonal antibody for HTLV-III.

To further characterize chemically and biochemically the antigen recognized by M3364, we purified the antigen by immunoaffinity chromatography using M3364 immunoglobulins coupled to activated CH-Sepharose. The immunoglobulin-bound antigens were eluted with 0.2*M* glycine, *p*H 2.8, containing 0.1 m*M* phenylmethylsulfonyl fluoride (PMSF). Portions from unfractionated virus extract, the fraction not bound to the immunoaffinity column, and the fractions eluted from the column were then subjected to electrophoresis on SDS-polyacrylamide slab gels. The protein bands were either stained with Coomassie blue or blotted to a nitrocellulose sheet. The blot was reacted with a human serum positive for antibodies to HTLV-III antigens. In Fig. 2A, lane 1 shows the antigenic composition of the unfractionated HTLV-III extract. Reactive antigens recognized are gp120, p66, p51, gp41, p31, and p24. With a longer exposure, p17 was also recognized. Lane 2 represents the unbound fraction. The same reactive antigens are present as in lane 1, but the reactivities toward p66 and p51 are noticeably decreased. Figure 2A, lane 3, and the lane in Fig. 2B show a fraction eluted from the immunoaffinity column and analyzed by the Western blot (lane 3) and stained by Coomassie blue (Fig. 2B). The results show that p66 and p51 are the only proteins recognized in this fraction by a polyvalent human serum and by Coomassie blue stain. Another portion of this fraction was dialyzed against 10 m*M* ammonium bicarbonate and lyophilized, then twice dissolved in water and relyophilized. While still in solution, a portion was removed for protein quantitation by amino acid analysis. The remainder of the sample was subjected to gas phase Edman degradation in an Ap-

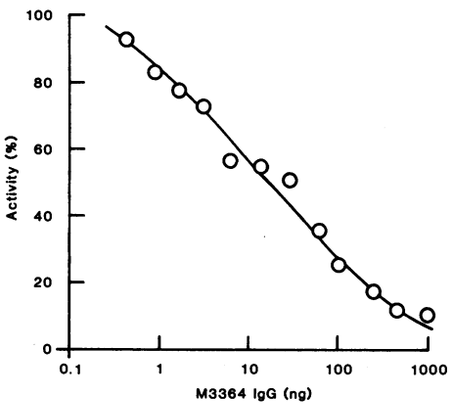

Fig. 3. Kinetics of reverse transcriptase activity. Portions of fractions eluted from the immunoaffinity column (Fig. 2) were neutralized with 0.1*M* NaOH and brought up to 0.1% Triton X-100, 1 mg/ml of bovine serum albumin, 8 m*M* dithiothreitol (DTT), 10% glycerol, and 0.1 m*M* PMSF. In the experiment described, 100 μl of one eluted fraction was incubated at 37°C with 50 m*M* tris-HCl, *p*H 7.8; 10 m*M* $MgCl_2$; 10 m*M* DTT; 0.1*M* NaCl; 20 μ*M* [^{3}H]dTTP (9600 cpm/pmol) and 50 μg of $(dT)_{\sim 15} \cdot (A)_n$ in a final volume of 1 ml. Portions (100 μl) were withdrawn at indicated times and were treated with 100 μg of yeast transfer RNA and about 3 ml of chilled 10% trichloroacetic acid containing 0.02*M* sodium pyrophosphate. The precipitates were collected on glass fiber filters, washed, and dried, and their radioactivity was determined with a Packard scintillation spectrometer.

Table 1. The NH_2-terminal amino acid sequence of HTLV-III reverse transcriptase as determined by Edman degradation of purified p66/p51.

Predicted amino acid sequence*	Sequence determined†		
	Cycle	Amino acid‡	Yield‡ (pmol)
Nucleotide numbers 2130–2132			
Pro	1	Pro	181
Ile	2	Ile	208
Ser	3	Ser	65
Pro	4	Pro	210
Ile	5	Ile	133
Glu	6	Glu	197
Thr	7	Thr	115
Val	8	Val§	
Pro	9	Pro	158
Val	10	Val§	
Lys	11	Lys	142
Leu	12	Leu	125
Lys	13	Lys	150
Pro	14	Pro	140
Gly	15	Gly	80
Met	16	Met	50
Nucleotide numbers 2178–2180			
Asp	17	Asp	65

*Sequence predicted from Ratner *et al.* (*10*). †The input was 300 pmol of protein. ‡Identified as the phenylthiohydantoin (PTH) derivative. The repetitive yield based on the quantitative recoveries of Ile (cycle 2) and Leu (cycle 12) is 95%, which is typical and reproducibly obtained in microanalyses on the gas phase sequenator. Residues which significantly differ from this yield are those whose PTH derivative tend to be unstable. §Qualitative assignment only. PTH-valine could not be accurately quantitated because of the presence of an artifact peak in the high-performance liquid chromatography analysis of each cycle of the Edman degradation.

plied Biosystems sequenator (*8*). The phenylthiohydantoin derivative of each amino acid was identified and quantitated by high-performance liquid chromatography on a phenylalkyl support (*9*). The results obtained in the first 17 cycles are described in Table 1, along with the amino acid sequence predicted from a segment of the nucleotide sequence of the *pol* gene of HTLV-III, clones BH10 and BH5 (*10*). The sequence determined is a perfect translation of the nucleotide segment 2130–2180, identifying the immunoreactive protein as a product of HTLV-III *pol* gene.

Immunoaffinity purified p66/51 was assayed for reverse transcriptase activity by using the primer-templates $(dT)_{\sim 15} \cdot (A)_n$, $(dT)_{\sim 15} \cdot (dA)_n$, $(dG)_{\sim 15} \cdot (C)_n$, and $(dG)_{\sim 15} \cdot (Cm)_n$ (*11*). Figure 3 shows the enzyme activity of a fraction for which we used the primer-template $(dT)_{\sim 15} \cdot (A)_n$. The enzyme reaction was linear for more than 60 minutes at 37°C. Relative to the activity with $(dT)_{\sim 15} \cdot (A)_n$, the enzyme activities were as follows: with $(dT)_{\sim 15} \cdot (dA)_n$, 2.9 percent, with $(dG)_{\sim 15} \cdot (C)_n$, 31.6 percent, and with $(dG)_{\sim 15} \cdot (Cm)_n$, 3.1 percent. Terminal deoxyribonucleotidyl transferase was absent in this preparation because it failed to catalyze the incorporation of radioactivity from tritiated deoxyguanosine triphosphate into polydeoxyadenylate. The results clearly indicate that at least one of the two proteins recognized by M3364 is active reverse transcriptase of HTLV-III.

The effect of the antibody M3364 on the enzyme activity in HTLV-III extracts was studied. Incubation of the reverse transcriptase with different concentrations of M3365 IgG did not inhibit the enzyme activity. Assuming that antibody binding might not inhibit the enzyme if the antigenic determinant were located distantly from the active site, we repeated the experiment as follows. The virus extract was incubated with serial dilutions of M3364 IgG in the presence of protein A–Sepharose. The immune complex was removed by centrifugation and the residual reverse transcriptase activity in the supernatant was measured. The results (Fig. 4) indicated a concentration-dependent removal of enzyme activity, greater than 90 percent of the enzyme being precipitated by 1 μg of the M3364 IgG. A titration of the effect of M3364 IgG on the purified reverse transcriptase gave similar results. Under the same conditions, no such reaction was observed with the reverse transcriptase activity present in extracts of HTLV-I.

The results presented here indicate that p66 and p51 share common epitopes recognized by the monoclonal antibody M3364. Both p66 and p51 are present in fairly equal amounts in virus preparations because they both appear as equally intense bands in Western blots of all batches of virus analyzed with the monoclonal antibody. In 17 cycles of Edman degradation the mixture of p66 and p51 gave a single amino acid residue in each cycle. This could mean that the two proteins have a common NH_2-terminus or that one of the proteins has a blocked NH_2-terminus. The latter is unlikely, because the quantitative recovery of the amino acids in each degradative cycle (Table 1) was significantly higher than the theoretical yield if only half the input protein was undergoing degradation.

It is not known whether both p66 and p51 are enzymatically active. They do not appear to be an S-S linked dimer because their electrophoretic mobilities are not influenced by treatment with β-mercaptoethanol. It is also not known whether they are products of a single gene or encoded by two separate but highly related genes present in the virus-producing cell. It is worth noting that the monoclonal antibody M3364 recognizes the same pair of proteins in another independent isolate of HTLV-III grown in H9 cells, namely, $HTLV\text{-}III_{RF}$ (*6*). If they are indeed derived from a single gene, then p51 should be a product of COOH-terminal cleavage of p66.

The high immunogenicity of p66 and p51 in HTLV-III antibody-positive individuals is remarkable, with the presence of antibodies to these proteins appearing to be independent of the clinical status of the patient or risk group to which the individual belongs. Such an immunogenicity of reverse transcriptase has not been demonstrated for other animal retroviruses, including HTLV-I, in their natural hosts.

Reverse transcriptases have been purified from avian myeloblastosis virus and Moloney murine leukemia virus and their amino acid sequences determined (*12*). We have now purified the reverse transcriptase of HTLV-III and determined its NH_2-terminal amino acid sequence. A complete biochemical characterization of the enzyme, including the determination of its active site, may now be possible. Reverse transcriptase appears to be a major target of antiviral agents, and many such agents are enzyme inhibitors in vitro (*13*). The availability of homogeneous reverse transcriptase will make possible the direct analysis of the molecular interactions between effective drugs and this critical viral component.

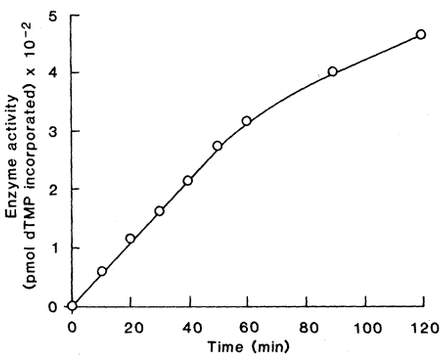

Fig. 4. Immunoprecipitation of HTLV-III reverse transcriptase activity with M3364 IgG. Ten microliters of a HTLV-III extract (1 μg) in 50 m*M* sodium phosphate, *p*H 7.5, containing 0.8*M* NaCl, 0.1% Triton X-100, and 0.1 m*M* PMSF was incubated in duplicate with indicated concentrations of M3364 IgG for 60 minutes at room temperature in 100 μl of PBS containing 16 μl of Protein A Sepharose that had been previously treated with rabbit antiserum to mouse κ light chain (100 μl per milliliter of Protein A Sepharose). The IgG concentration was maintained at 1 μg in each incubation by supplementing with IgG from an unrelated mouse hybridoma (M25). Portions (55 μl) were then transferred from the incubation mixtures and treated with 45 μl of a mixture that produced final concentrations of 50 m*M* tris-HCl, *p*H 7.8; 10 m*M* $MgCl_2$; 10 m*M* dithiothreitol; 0.1*M* NaCl; 20 μ*M* [^{3}H]dTTP (9600 cpm/pmol); and 50 μg/ml of $(dT)_{\sim 15} \cdot (A)_n$. The reaction mixture was incubated at 37°C for 60 minutes and processed as described in Fig. 3. Ten microliters of the virus extract incubated with 1 μg of the irrelevant IgG from the M25 hybridoma incorporated 30 pmol of [^{3}H]dTMP per milliliter under the conditions described. This was taken as 100%. M25 IgG caused no more than 7% inhibition of reverse transcriptase activity compared to a control in which phosphate-buffered saline was used instead of IgG.

REFERENCES AND NOTES

1. M. G. Sarngadharan, M. Popovic, L. Bruch, J. Schüpbach, R. C. Gallo, *Science* **224**, 506 (1984); S. Z. Salahuddin *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **82**, 5530 (1985).
2. M. G. Sarngadharan *et al.*, *Cancer Res. (Suppl.)* **45**, 4574 (1985).
3. F. di Marzo Veronese, A. L. DeVico, T. D. Copeland, S. Oroszlan, R. C. Gallo, M. G. Sarngadharan, *Science* **229**, 1402 (1985).
4. J. S. Allan, J. E. Coligan, F. Barin, M. F. McLane, J. G. Sodroski, C. A. Rosen, W. A. Haseltine, T. H. Lee, M. Essex, *Science* **228**, 1091 (1985); W. G. Robey *et al.*, *ibid.*, p. 593; F. di Marzo Veronese *et al.*, in preparation.
5. M. G. Sarngadharan, unpublished data.
6. M. Popovic, M. G. Sarngadharan, E. Read, R. C. Gallo, *Science* **224**, 497 (1984).
7. C. L. Reading, *J. Immunol. Methods* **53**, 261 (1982).
8. R. M. Hewick, M. M. Hunkapiller, L. E. Hood, W. J. Dryer, *J. Biol. Chem.* **254**, 7990 (1981).
9. L. E. Henderson, T. D. Copeland, S. Oroszlan, *Anal. Biochem.* **102**, 1 (1980).
10. L. Ratner *et al.*, *Nature (London)* **313**, 277 (1985).
11. M. G. Sarngadharan, H. S. Allaudeen, R. C. Gallo, *Methods Cancer Res.* **12**, 3 (1976).
12. M. Golomb and D. P. Grandgenett, *J. Biol. Chem.* **254**, 1606 (1979); T. D. Copeland, D. P. Grandgenett, S. Oroszlan, *J. Virol.* **36**, 115 (1980); G. F. Gerard, *Biochemistry* **20**, 256 (1981); T. D. Copeland, G. F. Gerard, C. W. Hixson, S. Oroszlan, *Virology* **143**, 676 (1985).
13. H. Mitsuya, M. Popovic, R. Yarchoan, S. Matsushita, R. C. Gallo, S. Broder, *Science* **226**, 172 (1984); H. Mitsuya *et al.*, *Proc. Natl. Acad. Sci. U.S.A.* **82**, 7096 (1985); P. Chandra *et al.*, *Cancer Res. (Suppl.)* **45**, 4677 (1985).

18 December 1985; accepted 21 January 1986