

ance was not observed under the discrimination task used here primarily because no compensatory responses were readily available. The delayed matching-to-sample task incorporates a memory component not present in simpler behavioral tasks. Several studies have demonstrated that short-term memory in both animals and humans is disrupted by short-term administrations of Δ^9 THC (7, 8). To the extent that memory impairment is not easily overcome by high probability compensatory behaviors, our failure to obtain tolerance may be considered as an instance which supports the importance of drug-behavior interactions in the development of marijuana tolerance.

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- Synthetic Δ^9 THC was made available in a dehydrated alcohol solution by Dr. M. Braude of NIMH. The 1.0 mg/kg doses were taken from NIMH batch SSC-66907 (96.3 percent Δ^9 THC, 1.2 percent Δ^8 THC, and 2.2 percent cannabinol). The 4.0 mg/kg doses were taken from NIMH batch SSC-69060 (93 percent Δ^9 THC, 3.2 percent Δ^8 THC, and 3.1 percent cannabinol). The synthetic Δ^9 THC was put into solution (0.1 g/ml) with sesame oil and kept refrigerated in darkness until use.
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Epstein-Barr Virus: Detection of Genome in Somatic Cell Hybrids of Burkitt Lymphoblastoid Cells

Abstract. *Somatic cell hybrids of Burkitt lymphoblastoid cells, from which Epstein-Barr virus can be recovered, were examined for the presence of virus DNA by DNA-RNA hybridization. Four clones of hybrid cells, each negative for virus antigens by immunofluorescence, contained virus DNA in varying genomic equivalents. The number of virus genome equivalents increased in the hybrid cells after induction of virus with iododeoxyuridine.*

The Epstein-Barr virus (EBV) and EBV-specific antigens have been observed in several lymphoblastoid cell lines (1). There are lymphoblastoid cell lines that do not contain either virus particles detected by electron microscopy or EBV-specific antigens detected by immunofluorescence. When cells from one of these lymphoblastoid cell lines, as well as cells from an EBV-positive cell line, were treated with 5-bromodeoxyuridine, EBV-specific antigens and virus were induced in both lines (2).

The Burkitt lymphoblastoid cell lines EB₃ and P3J-HR-1 (HR-1) can be hybridized to mouse and human cells, respectively (3). After D98/HR-1 (a somatic cell hybrid of HR-1 cells and D98, a human sternal marrow cell line) was exposed to 5-iododeoxyuridine (IdU), EBV-specific antigens were detected by immunofluorescence and virus particles were found by electron microscopy (4).

The technique of DNA-RNA hybridization on nitrocellulose filters can be used to detect integrated virus DNA associated with host cell DNA. Studies with both DNA-DNA and DNA-RNA hybridization have shown that Burkitt lymphoblastoid cells containing EBV antigens and so-called "normal" lymphoblastoid cell lines (without EBV antigens) contain virus DNA in varying amounts (5). We report here that DNA of EBV was detected in somatic cell hybrids of Burkitt lymphoblastoid cells, whereas no evidence of EBV in the hybrid cells was found either by immunofluorescence or electron microscopy until after treatment with IdU.

In each experiment, cells were grown in HAT selective medium (3, 6) or in Eagle's medium. Cells from five 250-ml plastic tissue culture flasks were

pooled in saline buffered with tris-(hydroxymethyl)aminomethane (pH 9). Clones 1, 2, 3, and 8 of the hybrid cells as well as the parental monolayer cell type, D98, were tested in the assay. Clone 1 of the hybrid cells was grown for 3 days in Eagle's medium containing IdU (60 μ g/ml), the medium was then replaced with normal Eagle's medium, the cells were grown for an additional 7 days, and the cells were examined for EBV genomes. The procedures for the preparation of DNA from EBV for use as template, the preparation of RNA complementary to virus DNA, and the determination of the number of genome equivalents per cell from DNA-RNA hybridization have been described (5).

Clones 1, 2, 3, and 8 of D98/HR-1

Table 1. DNA-RNA hybridization tests. An amount of cRNA (RNA complementary to DNA of Epstein-Barr virus) containing 1.5×10^5 count/min was used in each test. The amount of radioactivity bound to DNA of D98 cells (159 count/min) was subtracted from the other values before the number of genome equivalents was estimated. Cell lines not described in the text are Raji, a Burkitt lymphoblastoid cell line that was negative for virus by immunofluorescence, and Hep-2, a cell line not associated with Burkitt's lymphoma.

DNA on filter	cRNA hybridized (count/min per 50 μ g of DNA)	Genome equivalents per cell (No.)
D98	159	0
HR-1	21,282	610
Raji	2,411	60
D98/HR-1 clone:		
1	562	11
2	299	4
3	829	18
8	709	15
1, treated with IdU	3,499	97
Hep-2	148	0

hybrid cells were simultaneously tested for the presence of EBV antigens by the indirect immunofluorescence test and by electron microscopy. Cover slip monolayer cultures of the four clones of D98/HR-1 and of D98 cells were prepared. Hybrid and D98 cells were grown in 250-ml tissue culture flasks, treated in a similar manner as the cells in the hybridization experiment, and examined by electron microscopy. The cells were examined for virus antigens and virus particles 7 days after removal of medium containing IdU, a time corresponding to the assay of virus DNA in IdU-treated cells. Cells grown on cover slips were fixed in acetone and examined for EBV antigens with an antiserum against EBV (obtained from K. Traul, Pfizer, Inc.). Simultaneously, the hybrid cells grown in tissue culture flasks were fixed and examined by electron microscopy.

The results of the DNA-RNA hybridization experiments are shown in Table 1. All four clones tested contained DNA of EBV in varying genomic equivalents. Earlier experiments indicated that the D98 human parent cell contained no virus genomes, whereas the HR-1 Burkitt lymphoblastoid cell line, which was actively synthesizing virus, contained at least 680 genomes per cell (5). The number of genome equivalents in the D98/HR-1 hybrids ranged from 4 per cell in clone 2 to 18 per cell in clone 3. The number of genome equivalents in clone 1, initially 11 per cell, increased to 97 per cell after treatment with IdU. The IdU-treated cells were positive for EBV antigens by immunofluorescence, with up to 20 percent of cells containing EBV antigens in the nucleus or the cytoplasm. Virus particles were also observed by electron microscopy (4).

The data from the DNA-RNA hybridization experiments reveal that the somatic cell hybrids contain EBV genomes, even when the cells express no EBV markers, an observation also true of EBV-negative cell lines NC37 and F265 (2). The induction of virus antigens and virus particles with IdU indicates that the latent EBV genome can be induced to replicate. The induction of virus DNA is shown by the increase in hybridizable virus DNA in cells treated with IdU. Complete and incomplete virus particles, detectable by electron microscopy, also appear after IdU treatment.

The EBV genome is repressed in the

somatic cell hybrids, although it is expressed in the parental HR-1 cells. This would suggest that information supplied by chromosomes of the parental D98 cells aids in suppressing expression of EBV information. The infectivity and oncogenic potential of the EBV recovered from the somatic cell hybrids following induction by IdU remains to be established.

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Sexual Behavior in Rhesus Monkeys after Vasectomy

Abstract. *Vasectomy had no statistically significant effect on the sexual behavior of vigorous adult rhesus monkeys. Of two groups of males matched for level of sexual performance, one was vasectomized and the other underwent sham vasectomy. In sex tests administered within the month after surgery, the groups showed no difference ($P > .05$) in rate of mounting, intromission, or ejaculation, or in any other measure of sexual or sex-related behaviors.*

Fifty years ago hundreds of men in the United States, lured by the hope of experiencing sexual rejuvenation, were vasectomized by overenthusiastic clinicians who used as their rationale the results of poor research (1). Vasectomy proved to be no fountain of sexual vigor. In 1970 an estimated 750,000 men were vasectomized in this country as a contraceptive measure (2). The number of vasectomies has been rising rapidly despite complaints during the past decade of such side effects as impotency, decreased control over ejaculation, and other psychiatric symptomatology (3). However, Thompson and Illsley (4) noted that impotency is not a likely consequence of vasectomy and that if it does occur, it is probably only temporary; they conceded, however, that "the problem has not been adequately researched" (4, p. 2).

Psychological factors are often cited as being particularly important in determining the advisability of vasectomy as a method of birth control. In addition, numerous questions about the anatomical, neuroendocrinological, and immunologic consequences of vasectomy remain unanswered. Appropriate pre- and postsurgical care and operative techniques need to be standardized if the operation is to be widely adopted

as a satisfactory contraceptive measure and if the effects of vasectomy are to be correctly assessed. Not only do we need to know what aspects of sexual behavior change after vasectomy and to what extent, but we must identify which changes are due directly to physiological factors and which to cultural and psychological factors.

To this end we vasectomized a number of adult male rhesus monkeys whose sexual behavior was studied before and after surgery. Despite their resemblance to man, these primates can be expected to be free of such emotions

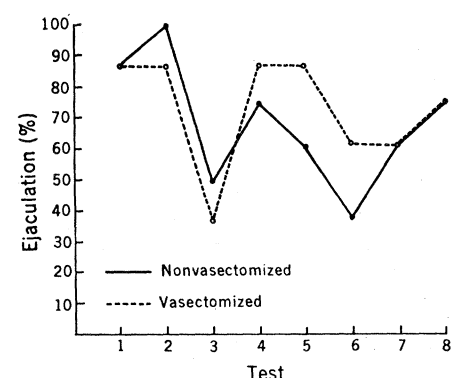


Fig. 1. Mean percentage of vasectomized and nonvasectomized males ejaculating in eight tests (two per week) of sexual behavior after surgery.