compared to the density of the vapor, the fraction of the total volume which they occupy is exceedingly small. Accordingly we change the properties of a vapor only slightly by a small cooling, as from A' to C'. Only the optical properties are noticeably changed. We commonly call such a vapor a mist.

A slight cooling below the liquid branch, say from A to C, results in a correspondingly small mass fraction of the liquid vaporizing into small bubbles. Since the density of the vapor is very low compared to the density of the liquid, the fraction of the total volume occupied by the vapor may be very high. Thus, if we adiabatically cool water from 25° to 24°C, the mass fraction that vaporizes is only 0.00168, but this small mass fraction of vapor occupies 98.7 percent of the volume. The volume fraction of the liquid is so small that it is confined to the corners of the vapor cells. The pure liquid phase at 25°C has changed into a foam by cooling 1°C. More generally, we define a foam as a mixture of liquid and vapor in which the overwhelming volume percentage is in the vapor phase, the overwhelming mass percentage is in the liquid phase, and the vapor is contained in cells bounded by liquid films. The bulk of the liquid is contained at the corners of these cells.

With further cooling below 24°C, more liquid vaporizes, resulting in still smaller overall density, ρ. It is this extremely low density that gives rise to a remarkably large enthalpy change

$$\Delta H = \int_{P_A}^{P_B} \frac{dP}{\rho}$$

when the pressure is reduced from P_A = 0.458 pound per square inch (psi) (31,600 dyne/cm^2), corresponding to the vapor pressure of water at 25°C, to P_B = 0.126 psi, corresponding to the vapor pressure of water at 5°C. This enthalpy change is just the area of the triangle ABC in Fig. 1 and is given by ΔH = −2.90 joule/g. It is sufficient to raise the 1 g of water 972 feet (296 m). When the plant design is optimized to achieve the lowest possible capital cost per unit power output the actual head will be reduced below the theoretical value of 972 feet.

The thermodynamic quantities P, ρ, and ΔH are given as a function of T in Table 1. These quantities have been computed from steam tables (*2*).

We visualize a foam SSPP as depicted in Fig. 2. Consider that the air is removed from the device, so that only water (liquid and vapor) exists within. The seawater intake provides warm surface water at 25°C with a vapor pressure of 0.458 psi. The condenser temperature is brought to 5°C by cooling with deep ocean water. This corresponds to a vapor pressure of 0.126 psi. There will be a flow of vapor from the warm water (25°C) to the condenser (5°C). However, if the water between 25° and 5°C exists as a foam, the flow of vapor from the warm region to the cool region will necessarily be accompanied by a flow of the whole foam structure. This foam structure consists not only of the individual foam walls, but also of the cell edges where two walls meet, and of the cell corners. The cell corners are of particular importance, for they hold the bulk of the fluid. If the liquid and vapor are separated by the foam breaker at the top of the tower, the liquid may be channeled into the central water column, where it is used to drive the turbine before being exhausted back into the sea.

Table 1. Properties of a foam having an upper limiting temperature of 25°C.

T (°C)	X (weight fraction of vapor)	P (psi)	ρ (g/cm^3)	ΔH (joule/g)
25	0	0.458	1	0
24	0.0017	0.432	0.013	−0.0063
22	0.0052	0.383	0.0037	−0.061
20	0.00865	0.339	0.0020	−0.18
15	0.0167	0.247	0.00077	−0.726
10	0.0248	0.178	0.000385	−1.63
5	0.0320	0.126	0.000208	−2.8

The maximum height of 972 feet is attained only when the available enthalpy is all converted into potential energy. But under these conditions the foam rises with a velocity approaching zero, and hence no power is developed. The maximum power per unit horizontal area is developed when the foam breaker is at two-thirds of its maximum height, 648 feet. For this height of the foam breaker, power is generated at the rate of 1.62 kw per square foot.

It is pertinent to ask how long the foam takes in rising 648 feet. This may be calculated directly from Bernoulli's equation

$$\int \frac{dP}{\rho} + \frac{1}{2}v^2 + gz = \frac{1}{2}v_0^2$$

where v is the velocity of the rising foam, z is the height the foam has risen, v_0 is the value of v at $z = 0$, and g is the acceleration of gravity. We take the differential of this equation

$$\frac{dP}{\rho} + vdv + gdz = 0$$

and multiply by ρ

$$dP + \dot{m}dv + g\dot{m}dt = 0$$

where $\dot{m}$ denotes the mass flux, ρv. Since the mass flux is constant, this equation may be integrated to give

$$\Delta P + \dot{m}\,\Delta v + g\dot{m}\,\Delta t = 0$$

Upon substituting the appropriate constants into this equation, we deduce that Δt, the time required for the foam to reach the foam breaker at 648 feet, is 21 seconds.

CLARENCE ZENER
Carnegie-Mellon University, Pittsburgh, Pennsylvania 15213
JOHN FETKOVICH
Department of Physics, Carnegie-Mellon University

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Oxidation of the Carcinogens Benzo[*a*]pyrene and Benzo[*a*]anthracene to Dihydrodiols by a Bacterium

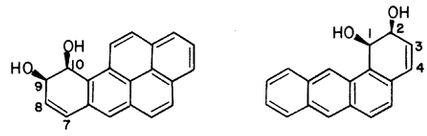
Abstract. *A mutant strain of* Beijerinckia, *after growth with succinate plus biphenyl, contains an enzyme system that oxidizes benzo*[a]*pyrene and benzo*[a]*anthracene to mixtures of vicinal dihydrodiols. The major dihydrodiol formed from benzo*[a]*pyrene was identified as* cis*-9,10-dihydroxy-9,10-dihydrobenzo*[a]*pyrene by comparison with a synthetic sample. Benzo*[a]*anthracene was metabolized to four dihydrodiols, the major isomer being* cis*-1,2-dihydroxy-1,2-dihydrobenzo*[a]*anthracene.*

Almost 200 years ago Percivall Pott reported a possible causal relationship between coal soot and the incidence of scrotal skin cancer in English chimney sweeps (*1*). Since that time several different polycyclic aromatic hydrocarbons (PAH) have been implicated as chemical carcinogens (*2*). Today, it is generally accepted that the carcinogenic properties of PAH are only manifested after metabolic activation by microsomal monooxygenases. Arene oxides, the initial oxidation products of aromatic rings, in mammals are possibly the ultimate chemical carcinogens (*3*). The present study explores the fate of carcinogenic PAH on oxidation by micro-

organisms. Previous reports indicate that bacteria will degrade PAH (*4*), but the structures of the metabolites have not been elucidated. We now provide the first structural information on the products formed by the action of a strain of *Beijerinckia* on benzo[*a*]pyrene (BaP) and benzo[*a*]anthracene (BA).

A bacterium which has been tentatively identified as a *Beijerinckia* species was isolated from a polluted stream by virtue of its ability to grow with biphenyl as its sole source of carbon and energy. Treatment of this organism with *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine led to the isolation of a mutant strain, *Beijerinckia* B-836, that oxidized biphenyl (*5*) to *cis*-2,3-dihydroxy-2,3-dihydrobiphenyl (46 percent), phenanthrene (*6*) to *cis*-3,4-dihydroxy-3,4-dihydrophenanthrene (45 percent), and anthracene (*6*) to *cis*-1,2-dihydroxy-1,2-dihydroanthracene (15 percent). Both BaP and BA failed to induce significant oxygenase activity in cells of *Beijerinckia* B-836. However, cells of this organism, after growth on succinate in the presence of biphenyl, oxidized both polycyclic hydrocarbons to polar products. *Beijerinckia* B-836 was grown on succinate in the presence of biphenyl for 4 hours (*5*). The cells were harvested (16 g, wet weight), washed three times with buffer (0.05*M* KH_2PO_4) at *p*H 7.2, and suspended in 400 ml of the same buffer. After addition of 30 mg of BaP in 3.0 ml of acetone, the suspension was incubated at 25°C on a rotary shaker for 48 hours prior to extraction with ethyl acetate. Preparative thin-layer chromatography of the extract (chloroform and acetone, 80 : 20) allowed isolation of a fraction (1.7 percent yield, $R_F = 0.30$) that was homogeneous in several solvents. Chemical ionization mass spectrometry with isobutane gave an ($M + 1$) peak at 287 (7 percent) with the base peak at 269 (100 percent) corresponding to loss of water, and the electron impact spectrum gave a parent ion at 286 (100 percent) and an ($M - 18$) peak at 268 (45 percent), indicating the presence of dihydrodiol. Acid-catalyzed dehydration (6*N* HCl, 100°C, 15 minutes) of the dihydrodiol produced material with the ultraviolet spectrum and chromatographic properties of 9-hydroxybenzo[*a*]pyrene (*7*), suggesting the structure 9,10-dihydroxy-9,10-dihydrobenzo[*a*]-pyrene. Examination of the Fourier transform proton magnetic resonance spectrum (Table 1) of the metabolite confirmed this assignment. The hydrogen assigned at C-10 is shifted downfield about 0.6 ppm from the position typical of a benzylic carbinol and has a chemical shift (5.63 δ) typical for a dihydrodiol that is proximate to a bay position (*6*). The coupling constant of $J_{9,10} = 5.2$ hertz requires *cis* conformation (*6*, *8*). Preponderant dehydration to the 9-phenol is consistent with earlier steric (*6*) and electronic (*9*) arguments.

Table 1. Proton magnetic resonance spectra of the major metabolites from BaP and BA. Spectra were recorded in acetone-d_6 (Varian 220-Mhz spectrometer equipped with Fourier transform) after exchange of hydroxyl protons with CD_3OD. The synthetic diol from BaP had an identical spectrum.



Chemical shift δ (ppm)	Coupling constants (hertz)	Chemical shift δ (ppm)	Coupling constants (hertz)
H_7 6.85	$J_{7,8} = 10.0$	H_1 5.46	$J_{1,2} = 5.4$
H_8 6.11	$J_{7,9} = 2.0$	H_2 4.72	$J_{2,3} \sim 2$
H_9 4.77	$J_{8,9} \sim J_{8,10} < 1.0$	H_3 6.05	$J_{2,4} = 2.8$
H_{10} 5.63	$J_{9,10} = 5.2$	H_4 6.55	$J_{3,4} = 9.9$

The ultraviolet spectrum of the metabolite was comparable to that reported for the *trans*-9,10-dihydrodiol obtained as a mammalian metabolite of BaP (*10*, *11*). However, the bacterial diol had an absorption of about 370 nm, which was reported by only one of the above groups (*11*). To establish whether this peak was due to an impurity, *cis*-9,10-dihydrodiol was synthesized. 7,8-Dihydrobenzo[*a*]-pyrene (*12*) was oxidized to *cis*-9,10-dihydroxy-7,8,9,10-tetrahydro-BaP with osmium tetroxide, and the product was acetylated. Bromination at C-7 with *N*-bromosuccinimide was followed by dehydrohalogenation and hydrolysis of the ester groups. A comparable route provided the *trans*-7,8-dihydrodiol from *trans*-7,8-dihydroxy-7,8,9,10-tetrahydro-BaP (*13*). The ultraviolet spectrum of the synthetic *cis*-9,10-dihydrodiol (Fig. 1) lacked the absorption at 370 nm. High-pressure liquid chromatography of the bacterial product on two coupled 1-m columns (Du Pont Permaphase ODS) with 43 percent methanol in water as the mobile phase allowed partial separation of a major (early) and a minor (about 10 percent) component. The major component no longer had the 370-nm absorption and was identical with the synthetic *cis*-9,10-dihydrodiol (Fig. 1), while the minor component had an ultraviolet spectrum identical with the synthetic 7,8-dihydrodiol (*14*). The minor bacterial dihydrodiol probably has *cis* configuration, but this cannot be proved until sufficient material is available for a proton magnetic resonance spectrum. This is the first instance in which *Beijerinckia* B-836 has produced a mixture of position isomers.

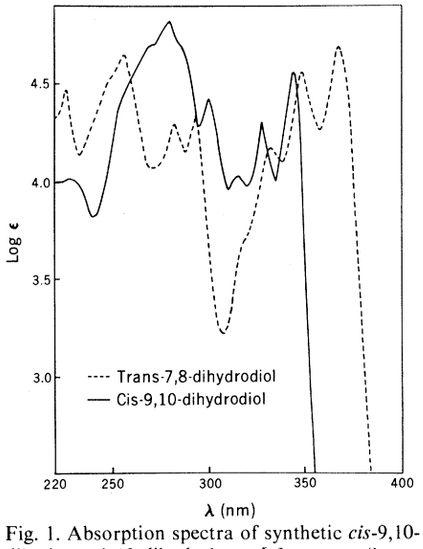


Fig. 1. Absorption spectra of synthetic *cis*-9,10-dihydroxy-9,10-dihydrobenzo[*a*]pyrene (λ_{max} 280 nm, $\epsilon = 6.65 \times 10^4$) and synthetic *trans*-7,8-dihydroxy-7,8-dihydrobenzo[*a*]pyrene (λ_{max} 368 nm, $\epsilon = 4.82 \times 10^4$) measured in ethanol (Cary model 14 recording spectrophotometer). The corresponding biosynthetic dihydrodiols at these ring positions gave essentially identical spectra after purification by high-pressure liquid chromatography.

Incubation of 1.2 liters of suspended *Beijerinckia* B-836 cells with 312 mg of BA in 62 ml of polyethylene glycol–20 for 14 hours at 25°C, followed by extraction with ethyl acetate, provided 330 mg of a yellow residue. Column chromatography of this residue on deactivated silica gel (20 percent water) and elution with a chloroform and acetone system (9 : 1) allowed isolation of 120 mg of an unstable, yellow dihydrodiol fraction which appeared homogeneous in several solvent systems on thin-layer chromatography. High-pressure liquid chromatography on a 0.25-m column (Du Pont, analytical Zorbax-Sil) and elution within a hexane, dioxane, and ethanol system (95 : 4 : 1) indicated that four partially resolved components were present. Repeated injection of the fraction enriched in the major component allowed isolation of pure *cis*-1,2-dihydroxy-1,2-dihydrobenzo[*a*]anthracene, as deduced from its proton magnetic resonance spectrum (Table 1). Shielding of the C-1 proton proximate to the bay position and the value of $J_{1,2} = 5.4$ are particularly diagnostic. Assignment of the other three dihydrodiols will be made when they are purified. The loss of positional specificity by *Beijerinckia* B-836 when oxidizing BaP and BA is in marked contrast to the high specificity observed with the smaller and noncarcinogenic PAH.

The identification of *cis*-dihydrodiols as

metabolites formed from BaP and BA by *Beijerinckia* B-836 suggests that these intermediates are not formed from arene oxide precursors since *trans* configuration would then pertain (*3*). It is possible that eukaryotic microorganisms utilize a monooxygenase enzyme system for the oxidation of aromatic hydrocarbons. Thus, *Cunninghamella bainieri* oxidizes naphthalene through *trans*-1,2-dihydroxy-1,2-dihydronaphthalene (*15*), and various fungi produce phenolic metabolites similar to those formed by hepatic microsomes (*16*). In contrast, prokaryotic organisms utilize a dioxygenase enzyme system, and *cis*-dihydrodiols have been identified as intermediates in the bacterial oxidation of several different nonphenolic aromatic hydrocarbons (*17*). The generality of this contrast awaits the isolation of bacteria from different genera that are capable of oxidizing such substrates. Bacterial metabolism to dihydrodiols is of particular interest since several recent reports have established that dihydrodiols of BaP and BA can be further activated by mammalian monooxygenases and can subsequently bind to DNA (*18*).

DAVID T. GIBSON
VENKATANAYARANA MAHADEVAN
Department of Microbiology, University of Texas at Austin, Austin 78712
DONALD M. JERINA
HARUHIKO YAGI
HERMAN J. C. YEH
National Institute of Arthritis, Metabolism, and Digestive Diseases, Bethesda, Maryland 20014

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Isolation of Type C Virions from a Normal Human Fibroblast Strain

Abstract. *Type C virions were spontaneously released from cultures of a diploid human cell strain. The virions have properties of known type C RNA tumor viruses and share antigenic determinants with the major interspecies-specific antigen (p30) of simian sarcoma virus. Antiserum to reverse transcriptase of gibbon ape leukemia virus inhibits the reverse transcriptase of the putative human virions and that of simian sarcoma virus, but has no effect on the corresponding enzymes of avian or murine RNA tumor viruses.*

It has been postulated that the genetic information for type C tumor viruses resides in the DNA of normal cells (*1*). Type C viruses can be activated from selected mammalian cell cultures by prolonged propagation in vitro or by exposure to x-irradiation, halogenated pyrimidine derivatives, or inhibitors of protein synthesis (*2*). We now report on the characterization of so-called HEL-12 virions which are released spontaneously from a human fibroblast cell strain.

HEL-12 cells were derived in August 1974 from the lungs of a spontaneously aborted 8-week embryo. The cells grew as fibroblast like monolayers in medium containing fetal calf serum and antibiotics (*3, 4*). Weekly tests for mycoplasma contamination were negative (*5*). Type C virions were not detected by electron microscopy or assays of culture fluids for RNA-instructed DNA polymerase (reverse transcriptase) after the cells were subcultured for a few generations (*6, 7*). However, type C virions were released spontaneously after HEL-12 cells were serially cultured for 6 months. Spontaneous virion production was observed on four different occasions after virus-free, frozen cells were reinitiated in culture (*8*). Despite their conversion to virion releasers, HEL-12 cells maintained a modal distribution of 46 normal chromosomes throughout their life span in culture.

Thin-section electron microscopy of HEL-12 cells revealed typical type C virions, both free in the extracellular space and budding from plasma membranes (Fig. 1) (*6*). The HEL-12 virions incorporated [^{3}H]uridine; in sucrose equilibrium

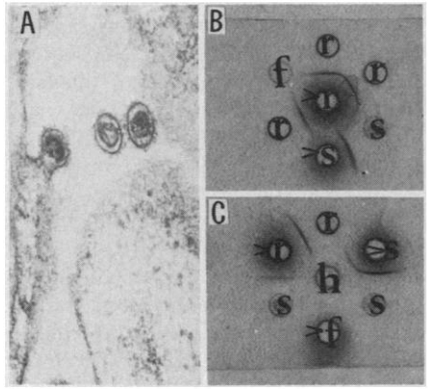

Fig. 1. Morphological and immunological properties of HEL-12 virions. HEL-12 cells were fixed with 2 percent glutaraldehyde in sodium cacodylate buffer, *p*H 7.3, and with 1 percent osmium tetroxide in collidine buffer, *p*H 7.3. Fixed cells were dehydrated in ethanol, embedded in Epon, and thin-sectioned. Sections were stained with uranyl acetate and lead citrate and examined in an RCA-EMU 4 electron microscope (*6*). (A) Electron micrograph of free and budding HEL-12 virions (× 62,000). (B and C) Double immunodiffusion analysis. The HEL-12 virions were purified by equilibrium centrifugation in sucrose gradients and examined for relatedness to R-MuLV (*r*), SiSV (*s*), and R-FeLV (*f*) with the use of monospecific antiserums to purified R-MuLV p30 (>*r*), SiSV p30 (>*s*), and R-FeLV p27 (>*f*). Slides contained 1 percent noble agar and 2 percent polyethylene glycol. Wells were filled with a total of 30 μl of undiluted antiserum or 15 μg of gradient purified virus. After overnight at room temperature, slides were washed for 3 days with six changes of normal saline, dried, and stained with amido black (*9*).