lated is irradiated (Fig. 2*A*). However, bacteria (*S. marcescens*) on the irradiated surface are rapidly killed as indicated by leaf disk prints (*2*) made by pressing the inoculated and irradiated surface against nutrient agar. After 5 to 10 minutes there was a noticeable reduction in the number of colonies, after 15 minutes there were very few, and after 20 minutes there were none.

The use of UV to determine if bacteria are on the leaf surface or in the intercellular leaf spaces is a useful tool in the study of microorganisms in this environment.

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Sedimentation Velocity Experiments: Position and Motion of Schlieren Peaks

Abstract. *There are four possible cases for single solute peaks in ultracentrifuge sedimentation velocity experiments. The peak can be either above or below the baseline and can move toward or away from the cell bottom. With appropriate solvents these cases are demonstrated for a single polymeric solute.*

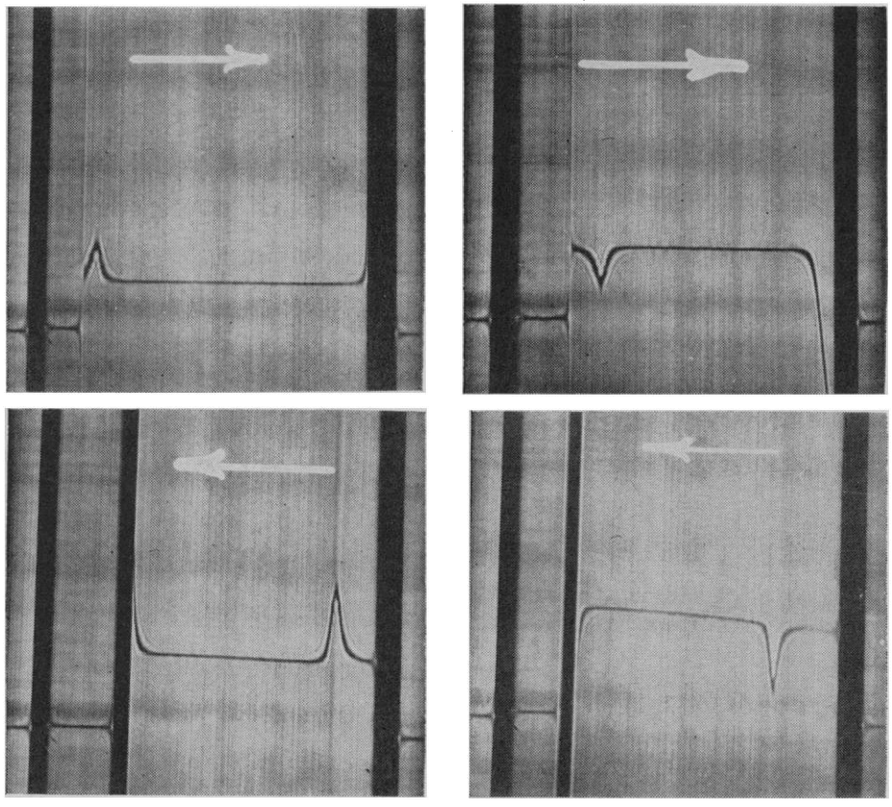
Conventionally, in ultracentrifugation the solute is not only more dense than the solvent, but it also has a higher refractive index. With the usual schlieren optics this leads, in velocity sedimentation, to a peak moving from the meniscus to the bottom and rising above the baseline (Fig. 1). If the solute is lighter than the solvent, the peak must move in the opposite direction—that is, from bottom to meniscus. In such a case (flotation) the refractive index of the solute is often less than that of the solvent, and the peak appears above the baseline (Fig. 3). However, it may not be as obvious that the sedimentation velocity peak can also exist below the baseline in either sedimentation or flotation. Such possibilities are rather infrequent (*1*) and seem to be unfamiliar to many people working with ultracentrifugation. These four distinct cases have not been explicitly described before, and it is our purpose to demonstrate them with a single solute in four appropriate solvents.

The key to the position of sedimentation peaks lies in the fact that the observed schlieren pattern is really a graph of dn/dx plotted against the distance from the center of rotation. We may represent this differential change of refractive index with distance as the product of two factors:

$$\frac{dn}{dx} = \frac{dn}{dc} \quad \frac{dc}{dx}$$

Each of the quantities dn/dc (change of refractive index with concentration) and dc/dx (change of concentration with distance) may be either positive or negative, giving rise to four cases, which are schematically represented as $++$, $+-$, $-+$, and $--$. These possibilities can be experimentally demonstrated with a single solute if one picks appropriate solvents. As an example we have taken polyisodecyl acrylate with density 1.02 and refractive index 1.473 at room temperature. Figure 1 shows its sedimentation in hexanol-1 ($[n]_D^{20} = 1.4135$; density, $\rho = 0.82$). Here both dn/dc and dc/dx are positive. This corresponds, of course, to the most usual case ($++$). In benzene ($[n]_D^{20} = 1.5017$, $\rho = 0.879$) the polymer still sediments but its refractive index is less than the solvent ($-+$), so dn/dc is negative; consequently the peak is just reversed about the baseline from the normal case (Fig. 2).

In chlorobenzene ($[n]_D^{20} = 1.525$, $\rho = 1.107$) both dn/dc and dc/dx are negative ($--$), so the product dn/dx is positive. This means the peak is upright but moves toward the meniscus (Fig. 3). Finally, in *n*-propyl bromide ($[n]_D^{20} = 1.4341$, $\rho = 1.354$) the polymer floats but has a higher refractive index than the solvent, giving the case ($+-$); hence the peak is



Figs. 1–4. Sedimentation peaks for polyisodecyl acrylate. Fig. 1 (upper left). Sedimentation in hexanol-1. Peak moves from the meniscus to the bottom and rises above the baseline. Fig. 2 (upper right). Sedimentation in benzene. The peak is reversed about the baseline from the normal case. Fig. 3 (lower left). Sedimentation in chlorobenzene. The peak is upright but moves toward the meniscus. Fig. 4 (lower right). Sedimentation in *n*-propyl bromide. The peak is downward but moves toward the meniscus.

downward but moving toward the meniscus (Fig. 4).

For any particular polymer it is often quite difficult to find appropriate solvents to illustrate the four cases. An example of another system which we have found is atactic polypropylene oxide in methanol, benzene, chlorobenzene, and $CCl_2F—CClF_2$ (*2*).

The existence of peaks above and below the baseline leads to some interesting possibilities in the ultracentrifugation of solute mixtures. For example, if two solutes in a single solvent correspond to the cases (+ +) and (− −), sedimentation peaks should form at both bottom and meniscus, both upright and moving toward each other; if it is assumed that the peaks are stable for a long time they should meet near the center of the cell, forming a single large peak which then splits into two again. So far, unfortunately, we have not been able to find the right combination of solvent and solutes to demonstrate this case experimentally. Similarly, if one starts with peaks on opposite sides of the baseline and moving toward each other, a canceling out should occur, and separate peaks would emerge again as they pass each other. A possible system for this case would be polyvinyl acetate and polyisobutylene in chlorobenzene, but the peaks we observed do not move rapidly enough (in relation to diffusion) to show the expected cancellation and emergence phenomena.

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1. For example, inverted peaks are obtained with certain lipoprotein fractions: see H. K. Schachman, *Ultracentrifugation in Biochemistry* (Academic Press, New York, 1959), pp. 126–127. As pointed out by a referee, similar phenomena can be observed in diffusion; L. G. Longsworth, in *Electrochemistry in Biology and Medicine*, T. Shedlovsky, Ed. (Wiley, New York, 1955), pp. 235–236, and in electrophoresis: H. A. Hoch and A. Chanutin, *J. Biol. Chem.* **200**, 241 (1953).
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Carcinogenic Aromatic Hydrocarbons: Special Vulnerability of Rats

Abstract. *Compared with other species, the rat is unusually vulnerable to polynuclear aromatic hydrocarbons. In the rat, relatively small amounts of carcinogenic aromatics (i) profoundly depress incorporation of thymidine in DNA, (ii) greatly induce menadione reductase in liver, and (iii) then kill the rat.*

Mammary glands of young adult female rats of the Sprague-Dawley (S-D) strain are foremost among cells of living creatures in their susceptibility to induction of cancer by irradiation (*1*) or by hydrocarbons and also in the speed with which cancers become evident. Mammary glands of the rat are surpassed in these respects only by cells of chickens inoculated in vivo with Rous sarcoma virus I.

The cell-free filtrate of an avian sarcoma injected in other fowls leads to a palpable tumor within 10 to 21 days (*2*). We have now found that a single large but tolerable dose of any of a number of polynuclear aromatic hydrocarbons or aromatic amines invariably elicited mammary cancer in S-D female rats aged 50 days (*3*); the first palpable cancer was detected in 20 days after administration of the hydrocarbon, and a large proportion of the tumors was manifest before the 30th day. Of these carcinogens, 7,12-dimethylbenz[a]anthracene (7,12-DMBA) is more efficient than all others by ten times. Whereas a single feeding of 7,12-DMBA under simple conditions (*4*) always induces breast cancer, intravenous injection of a concentrated lipid emulsion of 7,12-DMBA is equally effective, less hazardous to laboratory personnel, more convenient, and less costly in that smaller doses are required.

Cancers elicited by 7,12-DMBA differ between rat and mouse. In newborn mice, subcutaneous injection of 7,12-DMBA increased the incidence and the rate of development of both lymphomas and pulmonary tumors (*5*), whereas cancer of the breast was not elicited. In the newborn rat, 7,12-DMBA evoked mammary cancer (*6*) in high incidence whereas lymphomas were seldom induced. In adult S-D rats, a single feeding of 7,12-DMBA (133 mg/kg) induced mammary cancer in every female (*3*) but ovarian tumors did not develop; the dosage causing death (after a single feeding) of one-half the rats (LD_{50}) was 270 mg. Morii and Kuwahara (*7*) fed a single meal of 7,12-DMBA (500 mg/kg) to mice of two different strains without inducing mammary cancer; 60 percent of their mice of each strain developed cancers—predominantly ovarian in C_3H/HeJ mice and mainly lymphomas in C_{57} black mice. Accordingly there are two central problems in the vulnerability of animals to 7,12-DMBA: differences in (i) species and (ii) target areas.

Pathologic changes induced in mice by large doses of 7,12-DMBA have been described (*8*), but the minimum dosage causing death has not been determined. The LD_{50} in rodents was derived by the probit method of Gaddum (*9*) after a single intravenous injection of a lipid emulsion of 7,12-DMBA. Rats of two strains succumbed to much lower doses of 7,12-DMBA than did mice or *Citellus tridecemlineatus* (Table 1). For female mice of Carworth Farms No. 1 (CF1) strain,

Table 1. Toxicity of 7,12-DMBA in rat, mouse, and *Citellus*. The animals were given a single intravenous injection of lipid (15 percent) emulsion of 7,12-DMBA (0.5 percent) and observed for 21 days. S-D, Sprague-Dawley; CF1, Carworth Farms No. 1.

Strain	Sex	Age (days)	Number	LD_{50} (mg/kg)
		Rat		
S-D	♂	25	71	58
S-D	♀	25	100	54
Long-Evans	♀	25	54	73
		Mouse		
CF1	♂	44	44	382
CF1	♀	120	34	443
C_{57} black	♂	57	51	340
		Citellus		
	♂	>1 yr	11	>227
	♂, ♀	65	20	>227

Table 2. Incorporation of tritium (microcuries per gram of tissue, fresh weight) in the DNA fraction (perchloric acid–insoluble) of tissues of rats and mice 24 hours after injection with $TdRH^3$. Groups were of four or five females: Sprague-Dawley rats, about 130 g, aged 43 days; CF1 mice, about 20 g, aged 56 days. Intravenous injection with a lipid emulsion of 7,12-DMBA (0.5 percent) preceded by 4 hours the $TdRH^3$ (0.5 μc/g) injection. Adrenals were pooled; other tissues were processed individually, and the tritium content of washed perchloric acid-insoluble residues was determined (*10*).

7,12-DMBA (mg/kg)	Rat		Mouse	
	Ileum	Adrenal	Ileum	Adrenal
None	1.39 (1.01–1.79)	0.12	0.88 (0.7–1.2)	0.11
30	0.04 (.03–.05)	.05	0.82 (0.6–1.1)	.09
300			0.31 (.2–.5)	.07