that mothers of children dying from leukemia had a significantly greater frequency of antenatal abdominal x-ray examinations than mothers of matched control children (13 percent compared with 7 percent). The x-ray dosage in this type of examination would not exceed 5 r. It is apparent that such a large difference between the two groups of children would not be found if the dose-response relationship in the fetus were that estimated for adults by Court-Brown and Doll. This leads one to estimate from Stewart's data the fetal dose-response which would produce such a difference. This was done as follows:

1) The estimated population under age 10 in England and Wales in 1951 was 6 million. If the percentage of mothers of control children reporting an antenatal x-ray (7 percent) is applied to this population, there would be an estimated 420,000 children exposed in utero to a dose of approximately 5 r and 5.58 million children not so exposed.

2) From 1953 to 1955 there were 792 leukemia deaths in this age group, an annual average of 264.

3) Let R_1 and R_2 be the annual leukemia death rates per 100,000 population under age 10, for children with and without antenatal irradiation, respectively.

4) Since 13 percent of the leukemia children had a history of antenatal x-ray, 34 (0.13×264) children dying of leukemia in one year would have such a history. Therefore

$$R_1 = \frac{34 \times 100{,}000}{420{,}000} = 8.1$$

5) Since the remaining 87 percent of the 264 leukemia deaths would come from the nonirradiated group,

$$R_2 = \frac{230 \times 100{,}000}{5{,}580{,}000} = 4.1$$

6) Thus an estimated annual increment of 4.0 deaths per 100,000 population followed a dose of 5 r. The increment expected from Court-Brown and Doll's dose-response data is only 0.5 per 100,000.

Thus it appears that if Stewart's observation does, in fact, reflect an association between antenatal irradiation and leukemia, the response rate of embryonic tissue is approximately 8 times that of adult tissue.

The estimates which form the basis of this report are not intended to be precise figures, but are offered merely to stimulate interest in a very practical aspect of the problem of investigating the existence of a threshold leukemogenic dose of radiation. This aspect of the problem has so far received little attention.

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19 December 1958

Nitrogen Partition in Excreta of Three Species of Mosquitoes

Abstract. Adults of three species of mosquitoes, *Aedes aegypti, Anopheles quadrimaculatus* and *Culex pipiens,* showed essentially similar patterns of nitrogen output as judged by their excretion of total nitrogen and by their excretion of nitrogen as uric acid, urea, ammonia, amino acid, and protein. About 80 percent of their total nitrogen has been accounted for. Substances that seem on analysis to be like glycoprotein have been found in the excreta of the three species.

In continuing our previous work on the total nitrogen and uric acid patterns in the excreta and body tissues of adult *Aedes aegypti* (*1*) we have further partitioned the nitrogen excretion and have extended the analysis to two other species of mosquitoes, *Anopheles quadrimaculatus* and *Culex pipiens.*

The methods for the collection and extraction of mosquito excreta and lyophilization of the extracts were essentially similar to those employed in previous work (*1*). All analyses were performed in duplicate on extracts (lyophilized dry solid form) of excreta collected for a period of 2 weeks from insects on a diet of 4 percent sucrose and without any source of nitrogen. Total nitrogen was determined by the Kjeldahl method, and uric acid nitrogen was determined by the method of Kalckar (*2*) as modified by Praetorius (*3*). Urea and ammonia nitrogen were determined by the method of Seligson and Seligson (*4*).

The amino acid nitrogen determination was performed on desalted material as follows. A weighed amount of excreta (20 to 50 mg) was dissolved in 25 to 50 ml of distilled water and passed through a column (1 by 4 cm) of Dowex-50 (X-4, 50 to 100 mesh) in the hydrogen form. The column was washed with distilled water until the washings were neutral. The amino acids were eluted with 14 percent ammonia. The excess ammonia was removed in a vacuum at 25°C, and the solution was lyophilized. Amino acid nitrogen was determined by the Ninhydrin method of Moore and Stein (*5*) on a portion of the lyophilized material after the solids had been dissolved in a measured volume of distilled water.

Protein nitrogen was determined as follows. A weighed amount of the dried excreta extract (30 to 100 mg) was dissolved in a minimum volume of water, placed in a cellophane bag, and dialyzed for 3 days at +2°C against several changes of water. The dialyzed material was evaporated to dryness and hydrolyzed with 6*N* HCl at 110°C for 18 to 24 hours. The hydrolyzate, after removal of the HCl in a vacuum, was diluted to a convenient volume, and the amino acid nitrogen was determined by the Ninhydrin method (*5*).

Table 1 shows that the three species of mosquitoes, *Aedes aegypti, Anopheles quadrimaculatus* and *Culex pipiens* have essentially similar patterns of nitrogen excretion. Uric acid N represents approximately half of the total N, urea N about 10 percent, ammonia N about 10 percent, amino acid N about 5 percent, and protein nitrogen about 10 percent. Thus 80 percent of the total N excreted is accounted for.

Paper chromatography (*6*) was also performed on the protein hydrolyzates. The results showed the presence of tyrosine, phenylalanine, leucine, isoleucine, valine, proline, alanine, threonine, glycine, serine, glutamic acid, aspartic acid, arginine, lysine, cysteic acid, galactosamine, glucosamine, and trace amounts of β-alanine, indicating that the nondialyzable material probably contained a glycoprotein. This material was likewise found in the excreta of *Aedes aegypti* during the third and fourth

Table 1. Nitrogen partition in mosquito excreta collected for 2 weeks from mosquitoes on a diet of 4 percent sucrose.

Species	Nitrogen (%)					Total nitrogen accounted for (%)
	Uric acid	Urea	NH_3	Protein*	Amino acid	
Aedes aegypti	47.30	11.90	6.40	10.82	4.40	80.82
Anopheles quadrimaculatus	42.50	9.50	7.80	9.22	4.70	73.72
Culex pipiens	46.90	7.90	10.00	9.67	5.50	79.97

* Since chromatograms of the protein hydrolyzates indicated the presence of hexoseamines, the protein N values include amine N.

weeks. No analyses were made for the other two species beyond the second week. Work is now under way to isolate and characterize this material. As judged visually from the chromatograms, there were quantitative differences in the composition of the protein hydrolyzates. For instance, tyrosine and phenylalanine were present only in faint traces in *Aedes aegypti* as compared with higher concentrations in *Anopheles* and *Culex*.

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24 November 1958

The Clock Paradox

Abstract. In two-dimensional Minkowski space a geodesic arc is longer than every other admissible arc with the same end points. Thus a particle whose worldline is a geodesic—that is, an unaccelerated particle—requires more (local) time to travel between two events than an accelerated particle. Similar results in four-dimensional Minkowski space can be established.

The so-called "clock paradox" (*1*) in Special Relativity may be loosely stated by saying that if a space traveler were to take an extended trip into space, he would find upon returning to earth that he was younger than he would have been if he had remained at home. Since there is no such concept in Relativity as the same place at two different times, it is clear that a more precise formulation of the problem is required.

To simplify the problem, let us consider it in a space of one time dimension and one space dimension, and let the units be so chosen that the velocity of light is 1. In this two-dimensional Minkowski space, the element of local time is the element of arc length

$$ds = (dt^2 - dx^2)^{1/2}$$

The clock paradox is a simple problem in the calculus of variations.

Let P_1 (t_1, x_1) and P_2 (t_2, x_2) be two events, or points in Minkowski space, $t_2 > t_1$. The length of arc between P_1 and P_2 along a curve $x = f(t)$ connecting the two points is given by the line integral

$$s = \int_{t_1}^{t_2} (1 - f'^2)^{1/2} dt$$

The clock problem consists in comparing arc lengths between P_1 and P_2 along different paths.

The extremals of arc length, or geodesics, are given by the solutions of the well-known Euler equations

$$\frac{\partial \theta}{\partial f} - \frac{d}{dt}\left(\frac{\partial \theta}{\partial f'}\right) = 0$$

where $\theta = (1 - f'^2)^{\frac{1}{2}}$, and are readily found to be of the form

$$x = f(t) = at + b,$$

where $-1 < a < 1$. This is a straight line in Minkowski space and represents uniform motion in a straight line in ordinary space. Since Euler's equations are invariant under Lorentz transformations, the concept of geodesic is absolute.

Now comes the crucial difference between geodesics in Euclidean space and geodesics in Minkowski space, a difference which points up the danger of attempting to apply geometric intuition to this problem: In Minkowski space, the extremals (geodesics) represent relative maxima.

To show this, let

$$x = f(t) = at + b$$

be a geodesic connecting P_1 (t_1, x_1) and P_2 (t_2, x_2), $t_2 > t_1$, and let $\omega(t)$ be an admissible function vanishing at t_1 and t_2, so that

$$x = f(t) + \omega(t)$$

is a neighboring arc to the geodesic. The arc lengths along the geodesic and the neighboring arc are, respectively,

$$s = \int_{t_1}^{t_2} (1 - f'^2)^{1/2} dt$$

$$s + \Delta s = \int_{t_1}^{t_2} [1 - (f' + \omega')^2]^{1/2} dt$$

By the Mean Value Theorem

$$[1 - (f' + \omega')^2]^{1/2} = (1 - f'^2)^{1/2} + \frac{-f'}{(1 - f'^2)^{1/2}}\omega' + \frac{1}{2}\frac{-1}{[1 - (f' + \phi\omega')^2]^{3/2}}\omega'^2$$

where $0 < \phi < 1$. Now if $f(t) = at + b$ and $-1 < a < 1$,

$$\int_{t_1}^{t_2} \frac{-f'}{(1 - f'^2)^{1/2}}\omega' dt = \frac{-a}{(1 - a^2)^{1/2}}[\omega(t_2) - \omega(t_1)] = 0$$

so that

$$\Delta s = -\frac{1}{2}\int_{t_1}^{t_2} \frac{\omega'^2 dt}{[1 - (f' + \phi\omega')^2]^{3/2}}.$$

The integrand is never negative, and is 0 only when $\omega(t)$ is a constant and therefore 0. Thus for $t_2 > t_1$, Δs is negative for every neighboring curve. That is, the geodesic $x = at + b$ is an arc along which time is a relative maximum.

Let us assume, then, that we are in a space where there is no gravitation and that the earth is moving with constant velocity in a straight line. Then its world line in Minkowski space is a geodesic. Let P_1 (t_1, x_1) and P_2 (t_2, x_2), $t_2 > t_1$, be two distinct points on this geodesic. Any other arc connecting P_1 and P_2 would represent the time of another traveler whose motion is, for at least part of the journey, accelerated. This arc will be shorter than the geodesic so that a space traveler leaving the earth at P_1 would indeed be younger when he again met the earth at P_2 than he would have been if he had remained at home.

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Note

1. For a bibliography, see E. M. McMillan, *Science* 126, 381 (1957).

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Carotenogenesis and Resistance of Micrococcus pyogenes to Tetracyclines

Abstract. Although reddish-yellow pigments, mainly δ-carotene and rubixanthine, were present in the original strain of *Micrococcus pyogenes* var. *aureus*, mutants highly resistant to tetracyclines were observed to become colorless. All strains lack lipoxidase activity. The colorless strains probably reflect blocking by tetracyclines during carotenogenesis.

When *Micrococcus pyogenes* var. *aureus* (*Staphylococcus aureus*), strain 209 P, was cultured successively in media containing gradually increased amounts of tetracyclines, such as oxytetracycline, chlortetracycline, and tetracycline, acquisition of high resistance to these antibiotics was observed. In contrast to the original strain, which was sensitive to these antibiotics—that is, was killed easily by the antibiotics at a concentration of 0.5 μg/ml—and had a tinge of yellowish color, due to the presence of carotenoid pigments, the resistant strains, which withstood the addition of over 300 μg of tetracycline per milliliter of culture media, were observed to become colorless.

Both sensitive and resistant strains were grown on nutrient agar containing 2 percent glycerol and adjusted to *p*H 7.2 for mass cultivation. After incubation for 1 day at 37°C, and then for 6