

quantitative electron microscopy (4), and the average dry mass of the fiber of  $11.61 \times 10^{-16}$  g/ $\mu$ m, a total fiber length of 457  $\mu$ m per chromatid is calculated. A measured radius of 0.3025  $\mu$ m and a measured length of 3.025  $\mu$ m yield a volume of 0.87  $\mu$ m<sup>3</sup> for a chromatid. Knowledge of fiber diameter (365 Å) and fiber length (457  $\mu$ m) permits calculation of total fiber volume per chromatid which is 0.48  $\mu$ m<sup>3</sup>. Fiber volume divided by chromatid volume yields 0.55; that is, 55 percent of the volume of a chromosome is occupied by chromatin fibers. Preliminary calculations for some other chromosomes for which detailed quantitative evaluations of total and fiber masses and dimensions are available (3, 5) indicate that the percentage of fiber packing in human metaphase chromosomes varies from 30 to 70 percent.

For scanning electron microscopy, electrically nonconducting objects should be coated with a suitable conductor that permits the greatest exposure of the fine structure of the object. Deposition of 150 Å of metal undoubtedly obscured fine structural detail, but we were able to inspect the

three-dimensional aspect of chromosomes dried by the critical-point method in a more useful and demonstrable manner than is possible by stereoscopic electron micrography. Our study supports the folded fiber model of chromosome structure proposed by DuPraw and Rae (6).

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7. Supported in part by a research contract, project No. 3A013001A91C, from the Medical Research and Development Command, U.S. Army, Washington, D.C., and by grant P-259-j from the American Cancer Society. The opinions or assertions contained herein are the private views of the authors and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense.

13 November 1970

## Primitive Earth Synthesis of Nicotinic Acid Derivatives

**Abstract.** *Nicotinonitrile, 2-cyanopyridine, and 4-cyanopyridine can be synthesized under primitive earth conditions by the action of electric discharges on ethylene and ammonia. The electric discharge first synthesizes pyridine and hydrogen cyanide, which react in the discharge to form the cyanopyridines. Nicotinonitrile would have hydrolyzed in the primitive ocean to nicotinamide and nicotinic acid.*

The synthesis under primitive earth conditions of organic compounds such as amino acids, purines, and pyrimidines has been demonstrated by a number of investigators, but the synthesis of a vitamin has not been reported until recently (1). We find that nicotinonitrile (3-cyanopyridine) can be synthesized in small yield by the action of electric discharges on mixtures of various hydrocarbons and ammonia or nitrogen. Nicotinamide and nicotinic acid would be obtained by hydrolysis of the nitrile.

A mixture of ethylene (at a pressure of 330 mm) and ammonia (at a pressure of 120 mm) in a 3-liter flask containing tungsten electrodes (2) was sparked with a corona discharge for 4 hours. The compounds volatile at  $-78^{\circ}\text{C}$  were discarded, and the remaining yellow liquid was distilled into a trap cooled to  $+78^{\circ}\text{C}$ . The

yellow distillates from three similar experiments were combined and chromatographed on a 5-foot (152-cm) Porapak Q gas chromatograph column at  $170^{\circ}\text{C}$ . The peaks corresponding to 3-, 2-, and 4-cyanopyridine were collected and rechromatographed on a 5-foot,  $\frac{1}{8}$ -inch (0.29-cm) SE-30 column at  $50^{\circ}\text{C}$ . The cyanopyridine peaks were again combined and rechromatographed on the Porapak Q column. The cyanopyridine peaks were collected separately and their ultraviolet spectra were measured with a spectrophotometer (Cary model 15). The spectra corresponded to known samples of 3-, 2-, and 4-cyanopyridine. The percentage yields of nicotinonitrile, 2-cyanopyridine, and 4-cyanopyridine were  $7 \times 10^{-4}$ ,  $4 \times 10^{-4}$ , and  $1.3 \times 10^{-4}$ , respectively, based on the ethylene carbon.

It seemed likely that the cyanopyridines were formed from pyridine and

cyanogen radicals. Therefore, the yield of pyridine from the reaction of various hydrocarbons and nitrogen or ammonia was investigated (Table 1). The yields of pyridine vary from  $10^{-4}$  to 0.07 percent. Electric discharges are more efficient by a factor of 2 to 20 than pyrolysis (3) for the hydrocarbon-ammonia mixtures. The yield of pyridine is about the same with nitrogen as with ammonia if an electric discharge is used, but the hydrocarbon-nitrogen mixtures require a longer sparking time.

The yield of pyridine from the sparking of a mixture of methane and nitrogen over a water surface was  $4 \times 10^{-2}$  percent, greater by a factor of 7 than the yield in the absence of a water surface. It is not clear why the yield is greater in the presence of a water surface.

Pyridine synthesized in the atmosphere would dissolve in the primitive ocean as pyridine rather than as the pyridinium ion ( $pK_a = 5.4$ ). However, pyridine would not be reactive in the ocean, and, since it is more volatile than water in dilute aqueous solution (4), a substantial fraction would remain in the atmosphere where it would be subject to the effects of electric discharges and other sources of energy. Hydrogen cyanide, which is obtained in high yields from electric discharges, is also more volatile than water and would be present in the atmosphere (5). The reaction of these two compounds was simulated by sparking a mixture of pyridine ( $6.2 \times 10^{-3}$  mole) and hydrogen cyanide ( $1.6 \times 10^{-2}$  mole) for 3 hours; the products were distilled as before. The distillate was injected into a combination gas chromatograph-mass spectrometer, and the products were identified by their retention times and mass spectra. The percentage yields of nicotinonitrile, 2-cyanopyridine, and 4-cyanopyridine were 1.4, 0.85, and 0.23, respectively, based on the added pyridine. The percentage yields based on the pyridine used were 2.8, 1.7, and 0.48, respectively. These results can account for the cyanopyridine yields from the sparking of ethylene and ammonia. The yield of pyridine was  $5 \times 10^{-2}$  percent in this experiment, and the cyanopyridine yields are about 1 percent of the pyridine yield.

Pyrolysis and ultraviolet light are less effective than electric discharges in bringing about the synthesis of cyanopyridines. A mixture of pyridine and hydrogen cyanide was pyrolyzed with a hot wire at  $1200^{\circ}\text{C}$  for 45 minutes. The

Table 1. Formation of pyridine from simple hydrocarbons and ammonia or nitrogen.

| Hydro-carbon                  | Hydro-carbon pressure (mm) | Ammonia pressure (mm) | Nitrogen pressure (mm) | Energy source* | Time (hr) | Yield of pyridine (%)† |
|-------------------------------|----------------------------|-----------------------|------------------------|----------------|-----------|------------------------|
| CH <sub>4</sub>               | 426                        | 51                    |                        | Hot wire       | 2.0       | $0.8 \times 10^{-4}$   |
| CH <sub>4</sub>               | 407                        | 100                   |                        | Spark          | 1.0       | $3 \times 10^{-4}$     |
| CH <sub>4</sub>               | 290                        |                       | 125                    | Spark          | 18        | $6 \times 10^{-3}$     |
| C <sub>2</sub> H <sub>6</sub> | 355                        | 55                    |                        | Hot wire       | 1.3       | $7 \times 10^{-3}$     |
| C <sub>2</sub> H <sub>6</sub> | 340                        | 46                    |                        | Spark          | 1.5       | $1.6 \times 10^{-2}$   |
| C <sub>2</sub> H <sub>6</sub> | 220                        |                       | 105                    | Spark          | 12        | $8 \times 10^{-3}$     |
| C <sub>2</sub> H <sub>4</sub> | 432                        | 45                    |                        | Hot wire       | 1.0       | $2 \times 10^{-3}$     |
| C <sub>2</sub> H <sub>4</sub> | 350                        | 100                   |                        | Spark          | 1.5       | $5 \times 10^{-2}$     |
| C <sub>2</sub> H <sub>4</sub> | 230                        |                       | 90                     | Spark          | 18        | $4 \times 10^{-2}$     |
| C <sub>2</sub> H <sub>2</sub> | 210                        | 46                    |                        | Hot wire       | 1.0       | $7 \times 10^{-3}$     |
| C <sub>2</sub> H <sub>2</sub> | 110                        | 60                    |                        | Spark          | 1.0       | $7 \times 10^{-2}$     |
| C <sub>2</sub> H <sub>2</sub> | 250                        |                       | 85                     | Spark          | 0.5       | $4 \times 10^{-3}$     |

\* See (2) for a description of the hot wire and spark apparatus. † Yield based on carbon added. Over 90 percent of the hydrocarbon reacted in each case.

percentage yields of 3-, 2-, and 4-cyanopyridine were 0.22, 0.23, and 0.08, respectively, based on the initial pyridine. The action of ultraviolet light (Hanovia medium-pressure mercury lamp) on a mixture of pyridine and cyanogen in the gas phase gives only a 0.02 percent yield of nicotinonitrile. A similar experiment with pyridine and hydrogen cyanide gave less than a  $10^{-5}$  percent yield of nicotinonitrile.

We consider this to be a plausible mechanism by which nicotinamide could have been synthesized on the primitive earth. An estimate of the relative yields from this synthesis and from the cyanoacetylene-propionaldehyde synthesis (1) on the primitive earth would require a more detailed knowledge of the conditions than is currently available.

The presence of nicotinamide in the primitive ocean suggests that nicotinamide adenine dinucleotide may also have been synthesized, and that oxidation-reduction reactions analogous to

present day biochemical pathways were developed early in the chemical evolution of the earth.

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5. Solutions  $10^{-2}M$  in pyridine and hydrogen cyanide have partial pressures at 25°C of 0.07 and 0.4 torr, respectively.
6. Supported by NSF grants GB-8056 and GB-5303.

31 July 1970; revised 23 November 1970

## Bilateral Symmetry in Chick Embryo Determination by Gravity

**Abstract.** The determination of bilateral symmetry of chick blastodiscs occurring during the second half of the uterine period is only indirectly caused by the intrauterine rotation of the egg. As a result of this rotation the blastodisc is forced into an oblique position and the force of gravity affects the symmetrization process. The same result may be artificially achieved, without rotation, by forcing the blastodisc into an oblique position.

Von Baer (1) has shown that although the blastoderm of a freshly laid hen's egg is a circular sheet of cells, it is in fact a bilateral symmetric system. He showed that the axis of the future embryo can be predicted, after laying, from the shape of the egg and that in most cases when the egg is surveyed with its pointed edge to the right and the blunt edge to the left, the embryo

will develop perpendicularly to the long axis with its tail toward the observer.

Vintemberger and Clavert (2) studied the determination of the symmetry before laying and showed that it takes place between the 14th and 16th hour of the egg's rotation in the uterus (10 to 15 revolutions per hour), and not in the ovary as had been thought earlier

(3). They demonstrated a clear correlation between the direction of rotation and the orientation of the future embryo. They also showed that most eggs rotate in the uterus with their pointed edge toward the cloaca, in which case the embryo develops according to Von Baer's rule. In the minority of eggs which rotate with their blunt edge toward the cloaca the embryo develops in the direction opposite to the Von Baer direction.

The egg is composed of the shell and shell membranes surrounding the semi-fluid albumen, in which the egg yolk with the blastoderm on top of it float, the yolk being fastened to both edges of the shell by means of the chalazae. Conrad and Phillips (4) deduced from the formation of chalazae in an artificially rotated uterine egg that the yolk remained quiescent during the rotation with the blastoderm situated at the highest point. Vintemberger and Clavert (5) consequently attributed the symmetrization effect to the rotation of the albumen around the blastoderm on top of the stationary yolk.

Here experiments are reported in which an attempt was made to verify whether it is the rotation itself or a possible change of the blastoderm's position caused by this rotation that determines the future symmetry of the embryo.

Eggs were removed from the uterus of the hen by the application of external pressure 16 hours after the previous egg had been laid. Such uterine eggs are known to enter the uterus 6 hours after the previous egg has been laid. Therefore, they had been rotating in the uterus for only 10 hours, and their bilateral symmetry had not yet been determined (6). These uterine eggs were further incubated in different positions at the uterine temperature of  $41^\circ \pm 0.5^\circ C$  for about 10 more hours, and then were transferred (at estimated laying time) to  $38^\circ \pm 0.5^\circ C$ . For each uterine egg a previous egg laid normally by the same hen was used as a control, because from our experience the embryos of the same hen manifest a more or less constant angle to the long axis of the egg. The previous eggs were incubated at routine temperature of  $38^\circ \pm 0.5^\circ C$  immediately after having been laid.

In an extensive series of experiments we confirmed the results of Vintemberger and Clavert. We found that in uterine eggs the direction of rotation around their longitudinal axis determined whether the embryo will form according to Von Baer's rule, or con-