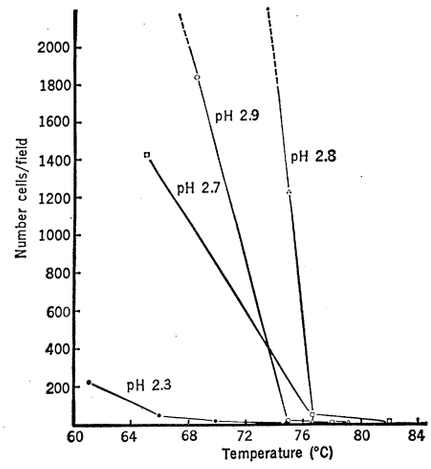

Fig. 2. Quantification of bacterial numbers at different temperatures along the thermal gradients of four acid springs. Microscope slides were immersed at different temperatures for a week; then they were removed and bacterial numbers per microscope field (3.7×10^4 μm^2) were counted. The dotted lines extend toward approximate values for slides that had too many bacteria for precise counting.

vironment was unfavorable for growth.

For the acid high-temperature springs, from which bacteria were always absent, it is important to ascertain that the waters would be favorable for bacterial growth upon cooling. This could most easily be done by observing the presence or absence of bacteria at various temperatures along the outflow channels of such acid springs, since this would provide a series of temperatures all in water of the same chemical composition. (There is no change in *p*H of the water upon cooling.) When such studies were made, it was found that bacteria did develop in these cooler waters, an indication that the water was indeed favorable for bacterial growth, provided that the temperature was not too high. The upper temperature limits observed along these thermal gradients were 70° to 75°C, which is the same as the upper temperature limit defined by studies of the whole group of springs (Fig. 1).

Quantitative studies on the rate of bacterial growth were performed at different temperatures along the thermal gradients of a few springs by immersing slides in the water for defined periods and then counting the number of bacteria per unit area of slide. These data (Fig. 2) also reveal that the upper temperature limit for bacterial growth in waters of *p*H 2 to 3 is 70° to 75°C. A further indication that low *p*H is not inherently limiting for bacterial growth is the fact that a number of obligately acidophilic bacteria which are also moderate thermophiles have been obtained in laboratory culture. Kaplan (*5*) and Schwartz and Schwartz (*6*) obtained cultures of acidophilic thiobacilli able to grow at temperatures up to 55°C, and Brierley (*7*) has obtained ferrobacilli of similar character. Uchino and Doi (*8*) isolated several acidophilic, thermophilic spore-forming bacilli, and we have confirmed and extended these observations (*9*).

High acidity also limits the upper temperature limit at which algae are found. Thus, although blue-green algae are found in neutral and alkaline hot springs at temperatures up to 73° to 75°C (*1*, *10*), in acid waters the upper temperature limit at which an alga (*Cyanidium caldarium*) is found is 55° to 56°C (*2*, *11*).

It thus seems as if high acidity may add an additional environmental stress that makes microbial growth at very high temperatures impossible. These results are of considerable interest in defining the physicochemical limits within which life is possible. It would now be of considerable interest to examine the stability to acid and high temperature of various kinds of cellular constituents to see which types are most likely to be labile. It is most likely that resistance to high acidity is associated with the impermeability of the cell to hydrogen ions (*2*), thus focusing attention on the cell membrane.

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Nuclear Sexing in a Population of Congolese Metropolitan Newborns

Abstract. *In a study of 5631 Congolese newborn children, the incidence of numerical X-chromosomal anomalies was the same as was previously reported in Europe and in North America.*

An extensive survey on the Congolese newborn was carried out between March and June 1969 at the Hôpital Général des Congolais in Kinshasa, in which hospital 80 to 120 births are registered per day (about 55 percent of all births in Kinshasa).

The survey was intended to provide a broad insight into problems presently encountered in newborn children in the large metropolitan city of Kinshasa (nearly 1 million inhabitants). Substantial information about various aspects of the mothers' and children's health, including some genetic and social aspects, has been obtained.

Nuclear sexing of buccal mucosa cells was performed on all liveborn children and all stillborn fetuses of at least 1000 g with the exception of the children and fetuses born every 6th day. The preparations were stained with aceto-orcein (*1*) and interpreted by two different investigators. All doubtful or abnormal smears were repeated. In every child, 500 cells were counted on two different slides. The total number of newborns examined was 5,631. Eight of the 2,834 males, all liveborn, had chromatin-positive buccal smears (2.8 per thousand). Chromosome analysis of peripheral blood

lymphocytes showed 47,XXY karyotypes in each case. Clinically, all the newborn chromatin-positive boys appeared normal. One of 2,797 females was chromatin negative (0.37 per thousand) and one had two chromatin bodies in buccal mucosal cells (0.37 per thousand). The chromatin-negative child had a 45,X karyotype and had typical clinical manifestations of Turner's syndrome. The child with two chromatin bodies was clinically normal and her karyotype was 47,XXX.

In this study of a newborn Congolese metropolitan population, which is comprised in one single socioeconomic class, the incidence of chromatin-positive males with 47,XXY karyotype, chromatin-negative females with 45,X–Turner's syndrome, and 47,XXX females is in good agreement with the results of similar studies on other populations (*2–9*) and with chromosome studies on newborn children (*10–12*) in other parts of the world (Table 1). The factors which predispose a woman to having a fetus with sex chromosomal abnormalities have not been conclusively demonstrated, but among the possibilities are maternal virus infections (*13*), endocrine alterations including those associated with the cessation of oral contraceptive therapy (*14*), and thyroid autoimmunity (*15*). In addition, it has been suggested that exposure of the mother to abdominal irradiation prior to conception may constitute an important predisposition to trisomy-21 in the offspring (*16*). In man, no similar information is available with regard to numerical anomalies of the X chromosome.

With only a very few exceptions, none of the mothers in this survey, and specifically none of the mothers of the children with abnormal buccal smears, had received either diagnostic or therapeutic radiation, and none had ever taken contraceptive hormonal drugs. It is thus quite likely that neither ionizing radiation nor hormonal contraceptives can be put forward as etiological factors for numerical aberrations of the X chromosome in this population. Moreover, the similarity of the incidence of sex chromosome abnormalities detected by screening for nuclear sex in the buccal mucosa in this population, as compared to other populations in Europe and North America, suggests that the incidence of sex chromosome aneuploidy may not be as dependent upon socioeconomic class as some pre-

Table 1. Sex chromatin surveys on newborn children.

Reference	Author (first)	Year and country	Total No. surveyed	Males				Females					Remarks
				Total No.	Chromatin-positive	Per thousand	Two chromatins	Total No.	Chromatin-negative	Per thousand	Two chromatins	Per thousand	
(*2*)	Moore	1959 Canada	3,715	1,911	5	2.6	None	1,804	None		None		
(*3*)	Bergemann	1961 Switzerland	3,728	1,890	4	2.1	None	1,838	1	0.54	None		
(*4*)	Wiesli	1962 Switzerland	3,029	1,563	1	0.6	None	1,466	None		None		Performed on amnion cells
(*5*)	Subray	1962 India	3,890	2,058	None		None	1,832	None		None		
(*6*)	Maclean	1966 Scotland	20,725	10,725	18	1.67	None	10,000	4	0.4	12	1.2	
(*7*)	Marden	1966 United States	4,412	2,206	7	3.1	None	2,206	None		1	0.45	
(*8*)	Taylor	1967 England	9,688	4,934	11	2.2	1	4,754	5	1.0	2	0.42	Two chromatin-negative females were 46,XY
(*9*)	Marquez-Monter	1968 Mexico	3,000	1,484	4	2.6	None	1,516	3	2.3	None		
(*10*)	Walzer	1969 United States	2,400	1,332	4	3.0	None	1,068	None		None		Chromosome study of all babies
(*11*)	Stewart	1969 Scotland	2,500	1,266	4	3.2	None	1,236	1	0.8		0.8	Chromosome study of babies with congenital anomalies
(*12*)	Sergovich	1969 Canada	2,081	1,066	1	1.1	None	1,015	None		None		Chromosome study of all babies
	This study	1969 Congo	5,631	2,834	8	2.8	None	2,797	1	0.37	1	0.37	

vious studies have suggested (*17*). The possible role of virus infections and of thyroid autoimmunity in this population should be investigated.

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Prebiotic Synthesis of Propiolaldehyde and Nicotinamide

Abstract. *We have identified propiololdehyde as a product of the action of an electric discharge on mixtures of methane and water or methane, nitrogen, and water. The aldehyde reacts with cyanoacetaldehyde and ammonia (other "prebiological molecules") to yield nicotinonitrile. This substance can be hydrolyzed to nicotinamide and nicotinic acid.*

Highly unsaturated molecules that can readily be obtained from simple gas mixtures by strong heating or by the action of an electric discharge are the starting materials in many potentially prebiotic syntheses (*1*). Recently we have shown that cyanoacetylene is formed in an electric discharge and that it reacts with cyanogen or cyanamide to give cytosine. We noted that cyanoacetylene also reacts with ammonia to give 6-aminonicotinonitrile. This led us to consider cyanoacetylene as a possible precursor of the vitamin nicotinamide (*2*).

Although 6-aminonicotinonitrile is formed in high yields from cyanoacetylene and ammonia, we have been unable to convert it to nicotinonitrile under potentially prebiotic conditions. Direct syntheses of nicotinonitrile from one molecule of cyanoacetylene and a three-carbon source at a more reduced level were therefore attempted.

Nicotinonitrile is not formed under mild conditions from cyanoacetylene, ammonia, and acrylonitrile nor from cyanoacetylene, ammonia, and compounds such as glyceraldehyde or malonaldehyde. Attempts to synthesize dihydronicotinonitrile—for example, from acrylonitrile and ammonia—were also unsuccessful. This led us to investigate propiolaldehyde as a three-carbon source. Here we describe the identification of propiolaldehyde as a product of the action of a discharge on certain simple gas mixtures, and its utilization in a plausible prebiotic synthesis of nicotinonitrile, nicotinamide, nicotinic acid, and corresponding *N*-alkyl derivatives.

Triatomic carbon (C_3) reacts with alcohols at low temperatures to give the diacetals of propiolaldehyde (*3*).

$$:C=C=C: + ROH \rightarrow HC \equiv C - CH(OR)_2$$

The corresponding reaction with water should give propiolaldehyde.

$$C_3 + 2\,H_2O \rightarrow HC \equiv C - CH(OH)_2 \rightleftharpoons HC \equiv C - CHO + H_2O$$

Since C_3 is formed by the action of an electric discharge on hydrocarbons such as methane (*4*) we investigated the products accumulating in liquid water when an electric discharge is passed through an atmosphere of pure methane above it (*5*).

Propiolaldehyde was first detected in the aqueous mixture by its color reaction with thiobarbituric acid (TBA), which is a specific test for propiolaldehyde, malonaldehyde, and certain of their derivatives (*3*, *6*), and by the formation of an adduct with HS^- with maximum absorption at 328 nm at *p*H's 7 to 9 (not formed by malonaldehyde).

In further experiments the propiolaldehyde was concentrated, first by distillation at reduced pressure and then by careful freezing of the distillate to crystallize out most of the water. The concentrate contained a complex mixture of organic compounds. We could not resolve propiolaldehyde completely by gas chromatography even though a number of different columns were used. The identification was therefore confirmed as follows.

1) A standard solution of authentic propiolaldehyde (*7*) was injected at 150°C onto a copper column (6 m by 0.3 cm) packed with Porapak Q, and the aldehyde peak was collected. A portion of the reaction product was then injected, and the material with the retention time of propiolaldehyde was collected. Both eluates gave identical TBA color reactions (maximum absorption at 532 nm; shoulder at 500 nm). By comparing the color intensities given in the TBA test by the solutions before injection and by the fractions collected from the column, we estimated a 10 percent recovery both for the standard and for the reaction product.

2) Chromatography at 95°C through a copper column (6 m by 0.9 cm) packed with 5 percent Carbowax 1500 on Chromosorb P showed the authentic aldehyde to have a retention time corresponding to a shoulder on the emerging side of a major peak of the product mixture (probably toluene). The area with the same retention time as propiolaldehyde was collected. This eluate gave an intense TBA test identical to that given by authentic propiolaldehyde. A collection was then made of the total eluate excluding the propiolaldehyde region. Although this eluate gave a TBA test, its intensity was only 5 percent of that given by the propiolaldehyde fraction.

The maximum yield of propiolaldehyde obtained in these experiments was about 0.1 percent based on the methane destroyed. When a mixture of nitrogen and methane (90:10) was used the yield of propiolaldehyde (as judged by the TBA test carried out on the total aqueous phase) was increased up to 0.37 percent.

The formation of pyridines by the condensation of enamines with acetylenic ketones or aldehydes and subsequent cyclization of the adducts has