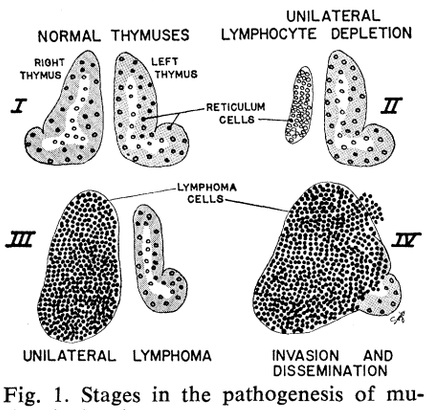

Fig. 1. Stages in the pathogenesis of murine leukemia.

and mitoses, architectural disorganization and cellular proliferation resulted in significant enlargement of the affected organ.

4) Generalized lymphoma: Animals in which the proliferating lymphoma had invaded the opposite thymus and disseminated to spleen, liver, and peripheral nodes.

In chromosome counts, a significant alteration was considered to occur when at least 10 percent of the cells of that organ exhibited a chromosome number different from the normal diploid mode of 40. Chromosome counts were carried out independently and without prior knowledge of the pathologic diagnosis.

The nonproliferative "preleukemic" changes described for group 2 invariably preceded the development of overt lymphoma. This was observed consistently in the murine leukemias induced by a variety of viruses (*9, 11*). If the change in chromosome number is part of the sequence leading to or causing the neoplastic transformation, it should be observed prior to the histologic appearance of tumor. Table 1 shows that this is not the case. Significant numbers of aneuploid cells were not observed in the thymuses of animals with nonproliferative preleukemic thymic changes (group 2) and only rarely even in mice with early thymic lymphoma (group 3). This is in contrast to animals exhibiting late proliferating lymphoma (group 4), in which the thymus cells of six out of ten animals showed alterations in chromosome number.

Table 1. Incidence of aneuploidy during various stages of leukemogenesis. Group 1, no pathologic changes; group 2, unilateral thymus depletion (pre-lymphoma); group 3, unilateral lymphoma; group 4, generalized lymphoma.

Incidence of aneuploidy	Group			
	1	2	3	4
	Right thymus			
In animals*	0/15	0/6	0/7	4/10
In cells†	2/181	0/73	2/87	64/283
	Left thymus			
In animals*	0/15	0/6	1/7	3/10
In cells†	2/159	3/65	52/149‡	54/209

* Number of animals whose thymus cells showed 10 percent or more aneuploidy/number of animals examined. † Number of aneuploid cells/number of cells examined. ‡ Fifty-one aneuploid cells were observed in one animal with late, though unilateral, lymphoma (51/53).

The sparsity of aneuploidy in pre- and early leukemia compared with the high incidence in late disseminated lymphoma suggests that the chromosome changes which are associated with the "thymic" group of murine leukemias are not causal but rather represent one of the consequences of the neoplastic transformation.

At present, the noncausal role of observed chromosome aberrations may be properly applied only to the murine viral leukemias. The clinical similarity of murine leukemia to that in man may offer insight into the role of chromosome aberrations in human leukemia.

The results of studies with chemically induced neoplasia are equivocal (*13*). In a study of aneuploidy in murine leukemia induced by irradiation, Nadler (*14*) suggested that these leukemias were not suitable for evaluation of the significance of aneuploidy.

In chromosome studies of the Shope virus papilloma-carcinoma system during the transition from benign to malignant tumors, no consistent abnormalities were observed (*15*). Hellström *et al.* (*16*), studying primary polyoma tumors, suggested that the observed aneuploidy was secondary to the neoplastic transformation.

The data presented here do not bear upon the possible role of undiscernible chromosome aberrations as discussed by Nichols (*17*), or the role of chromosome aberrations in enhancing the progression of tumor (as opposed to its initiation). They do, however, add experimental proof to the hypothesis that chromosome aberrations do not play a primary role in the etiology of the virus-induced leukemias.

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Serotonin Rhythm in the Pineal Organ: Control by the Sympathetic Nervous System

Abstract. *The serotonin content of the pineal organ of the rat varies diurnally in relation to the photoperiod. When the sympathetic nerves to the pineal are interrupted by the removal of the superior cervical ganglia, no such fluctuation in the serotonin content of the pineal occurs.*

On the basis of careful anatomical studies, Kappers (*1*) has proposed that the pineal organ is innervated chiefly, if not completely, by fibers of the autonomic nervous system. Of these, the nerves leaving the superior cervical ganglia, passing in the tentorium over the dorsal surface of the cerebellum as the nervi conarii, and penetrating the pineal organ, constitute the primary supply.

There is some biochemical evidence supporting the view that such a neural

Table 1. Serotonin content of the pineal organ (nanograms per gland) at different times in the photoperiod in male Sprague-Dawley rats 8 to 9 weeks old. The experimental rats were ganglionectomized; the controls were sham-operated or intact.

Series	Control rats		Experimental rats	
	Light*	Dark†	Light*	Dark†
I	88	48		
II	117	42	67	53
	97	52	72	70
III	96	33	46	54
IV	134	22	48	
		76		
		76		
V	104	20	51	44
	99		44	84
			40	
VI	112	41	40	41
		Means		
	106	42	53	58
		Range		
	134–88	76–20	72–40	84–41

* Animals killed approximately 8 hours after the onset of the light period. † Animals killed approximately 4 hours after the onset of the dark period.

pathway is important in pineal metabolism. After the removal of the superior cervical ganglia the amount of serotonin in the pineal organ is reduced as measured by spectrofluorometric assays, or as seen in a study of fluorescent sections (*2*). Furthermore, an enzyme which is essential in the transformation of serotonin to melatonin, hydroxyindole-*O*-methyl transferase does not vary in response to continuous light or darkness in ganglionectomized rats (*3*) as it does in intact animals. An interesting aspect of pineal physiology has been demonstrated by Quay (*4*) who has shown that the amount of serotonin in the pineal organ fluctuates diurnally in rats in relation to the photoperiod, the largest amount being found during the 8th hour of the light period and minimum amounts being detected 4 hours after the onset of the dark period. The study reported here was designed to determine whether or not superior cervical ganglionectomy would affect this diurnal rise and fall of serotonin in the pineal organ.

Fifty-eight Sprague-Dawley male rats were placed under controlled environmental conditions when 22 days of age. Throughout the experimental period all animals were exposed to 14 hours of light followed by 10 hours of darkness. The temperature was maintained at 24° ± 1°C, and the humidity at 50 percent. Purina Chow and tap water were available all the time.

Of these animals, 26 were subjected to superior cervical ganglionectomy under ether anesthesia while 30 served as sham-operated or unoperated controls. All operations were performed when the rats were 24 to 28 days old. The completeness of this procedure was evident in the ptosis of the eyelids which developed. This was marked immediately after the operation but with time was partly overcome as the voluntary nerves corrected for the tonus normally maintained by the sympathetic nerve supply. Five to six weeks later the animals were killed quickly by cervical dislocation at a given point in the photoperiod, that is, within 7½ to 8½ hours after the onset of the light phase or 3½ to 4½ hours after the beginning of the dark period. During the dark period the animals were killed in almost total darkness, a substage lamp equipped with a green filter serving as the light source.

Each pineal organ was removed within 1 minute after death and immediately homogenized in a 5-ml centrifuge tube as described by Quay (*4, 5*). To insure that adequate amounts of serotonin were present for accurate measurement, two pineal organs were combined in each tube. All pools were assayed within 24 hours and prior to assay they were maintained at −5°C. The extraction procedure used has been fully described elsewhere (*5*). While this method is not entirely specific for serotonin it does insure the nearly complete removal of interfering indoles known to occur in the pineal body. The fluorescence was measured with a Farrand spectrofluorometer equipped with a 0.3-ml quartz cuvette and adaptor. The preparation of the pools and the extraction procedure were repeated on six different occasions.

As shown in Table 1, pineal organs removed from intact or sham-operated rats approximately 8 hours after the onset of the light period always contained large amounts of serotonin; pineal organs from control rats, removed about 4 hours after the beginning of the dark period, gave significantly lower serotonin values. After the removal of the superior cervical ganglia, there was no such variation in serotonin content in relation to the photoperiod.

These results confirm Quay's observation that the amount of serotonin in the pineal organ of the rat varies with the photoperiod. In addition they indicate that such diurnal fluctuations do not occur if the sympathetic pathways from the superior cervical ganglia to the pineal organ are interrupted.

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Electron Microscopy of Single-Stranded DNA: Circularity of DNA of Bacteriophage ϕX174

Abstract. *The single-stranded DNA of coliphage ϕX174 has been examined with the electron microscope by a modification of the protein-monolayer-adsorption technique. The molecules were found to be circular with a total length of 1.77 ± 0.13 microns.*

The DNA of coliphage ϕX174 is single-stranded (*1*) and also circular (*2*). The latter conclusion, which has been based upon the resistance of the DNA to digestion by exonucleases and upon an analysis of the change in the ultracentrifugal pattern after the introduction of chain scissions by deoxyribonuclease, is confirmed by electron microscopy.

Direct observation of double-stranded DNA molecules by electron microscopy is easily performed if the DNA is adsorbed onto protein monolayers (*3*). However, making this observation has been complicated by the entanglement of a variable fraction of the molecules, although it is usually possible to find a sufficient number of extended, untangled filaments to obtain a length distribution.

When ϕX174 viral DNA was examined, exceedingly severe tangling eliminated any possible conclusion concerning length or configuration, although the success with the double-stranded replicative form (*4*), that is, the clear