On the Etiology and Pathogenesis of the Leukemias: A Review*

HENRY S. KAPLAN

(Department of Radiology, Stanford University School of Medicine, San Francisco, Calif.)

This paper presents an analytical review of the recent literature on the etiology and pathogenesis of human and experimental leukemia, and a hypothetical interpretation of the mechanism of induction of the experimental disease. Emphasis has been placed on the gaps in our existing knowledge and on the relationship, if any, between what we have learned in the mouse and the corresponding situation in man. The term “leukemia” has been used rather broadly in the title to include evidence pertaining to lymphosarcoma and Hodgkin’s disease, but an attempt will be made to be more specific regarding terminology in the text. The older literature is thoroughly covered in earlier reviews (39, 40, 79).

There has been an astounding increase in the incidence of leukemia and the related malignant lymphoid tumors, which has received little recognition in comparison with the considerable furor about the increasing incidence of carcinoma of the lung. Yet in the year 1950, 8,845 people died of leukemia in the United States, a crude death rate of 5.9 per 100,000 population, as compared with a rate of only 5.1 for primary malignant tumors of the stomach, colon, breast, rectum, cervix uteri, and lung, and bronchus (119). The leukemias and lymphomas rank about fourth or fifth among the causes of death from malignant neoplasms (Table 1) and deserve a commensurate degree of our interest and effort (131).

WHAT IS KNOWN REGARDING THE ETIOLOGY AND PATHOGENESIS OF HUMAN LEUKEMIA?

The Leukemias as an Etiological Entity

When we refer to the “etiology of leukemia” we must carefully avoid the inference that all the leukemias necessarily have the same etiology. The leukemias have in common certain morphological similarities and share the same fatal prognosis, though at different rates. There is as yet no conclusive evidence that the various types of leukemia represent an entity from the etiologic point of view.

Despite certain transitional situations, such as the preterminal transformation of many chronic leukemias to the morphology ordinarily associated with acute leukemia, there are many clinical reasons to question the view that acute and chronic leukemias are merely different manifestations of the same disease. For example, chronic leukemias are rare in childhood, are seldom ushered in by serious infection, tend to show few hemorrhagic phenomena until late, are usually highly radio-responsive over the major part of their course, and are gen-

* This is one of a series of papers presented in a symposium on leukemia at the annual meeting of the American Association for Cancer Research, Inc., in Atlantic City, April 10, 1954. The other two papers, by Burchenal and by Law, will be published in the October and November issues. Acknowledgment is made with respect to previously unpublished work cited of grant-in-aid support from the National Cancer Institute, National Institutes of Health, Public Health Service, and the Jane Coffin Childs Memorial Fund for Medical Research.

Received for publication May 21, 1954.
erally associated with gross tumefaction of liver, spleen, and lymph nodes. In contrast, acute leukemias are very prevalent in infancy and childhood, with a peak incidence in the 4th year of life, and tend not to occur with any great frequency again until the other extreme of life. As might be expected from the early onset of pancytopenia in acute leukemia, significant infection is often the first manifestation of the disease; hemorrhagic phenomena tend to dominate the clinical picture and are the major cause of death (25, 48, 122).

Acute leukemias are notoriously refractory to radiation and may indeed be adversely affected by it. The chemotherapeutic responses of the acute and chronic leukemias are also significantly different (11). Aminopterin and other folic acid antagonists have been successful in producing clinical remissions in the acute leukemias of childhood but generally ineffectual in both the acute and chronic leukemias of adult life. Conversely, such agents as urethan, radioactive phosphorus, and the mustards, which have been quite useful in the treatment of chronic leukemias and the malignant lymphoid tumors, have been entirely without effect in the acute leukemias. It is also worth noting that, with the exception of two dubious reports (62, 85), no authentic instance of the transformation of acute to chronic leukemia either spontaneously or in response to chemotherapy has ever been recorded. We must therefore guard against drawing sweeping conclusions concerning the etiology of all leukemias from evidence pertaining to any one variety.

Leukemogenic Agents in Man

Radiation.—Ionizing radiation is the only external agent that has been conclusively shown to be leukemogenic for man. The first line of evidence is that furnished by case reports (92) and statistical studies (108, 104, 197) which reveal a significantly greater incidence of leukemia in radiologists and in others occupationally exposed to radiation than in the general population.

More recent evidence has come from the studies of the Atomic Bomb Casualty Commission in Japan. Folley, Borges, and Yamawaki (38) reported that, as early as 1950, there was a highly significant increase in leukemia incidence among those exposed to relatively high doses of radiation (less than 2,000 meters from the bomb hypo-center), as compared with others at a greater distance who received negligible amounts of radiation. The distribution of leukemia by type suggested a disproportionate increase in subacute or chronic myelogenous leukemia among the irradiated population. More recent studies continue to reveal a very high incidence of chronic myelogenous leukemia; there has been little chronic lymphatic leukemia, however, and the evidence that exposure to radiation has enhanced the incidence of acute leukemia is still equivocal.

Another line of evidence implicating radiation as a leukemogenic agent is the suggestion by Schwartz and Ehrlich (126), Merskey (111), and others that the relatively high incidence of leukemia as a terminal manifestation in polycythemia vera is not an expression of the natural history of the disease but the result of repeated intermittent radiation therapy generally given to polycythemic patients. Though plausible, this suggestion is not yet susceptible of statistical verification.

Benzol and other chemicals.—Much of the evidence with regard to other leukemogenic agents in man is of circumstantial nature. The evidence that chronic benzol poisoning may lead to myelogenous leukemia is perhaps the most convincing (15, 60, 61, 102). A number of other industrial and medicinal chemicals have been incriminated in sporadic cases of human leukemia (60, 105), but, with the possible exception of the arsenicals and sulfonamides, no one class of drug or chemical recurs with sufficient regularity in these reports to give substance to the charge that it caused the disease.

Genetic constitution.—The evidence just cited represents all that is really known to date regarding exogenous agents in human leukemia. A number of other factors may contribute to the leukemogenic process, however. That genetic constitution may condition susceptibility to leukemia is indicated by the genealogical studies of Videbaek (139) and others (2, 19, 49), as well as by isolated reports of a high familial incidence (53, 140). Most striking of these is the family reported by Anderson (1), in which five of eight siblings died of acute leukemia at about the same age. It is of course difficult to prove in such instances that the causative factor is indeed genetic, and not associated with some obscure, unique aspect of the family environment.

Congenital leukemia is exceedingly rare; Bernhard, Gore, and Kilby (5) were able to find only fourteen acceptable cases in the literature before 1951, to which they added four cases. In none of the reported cases was the mother leukemic. Conversely, it has long been known that pregnant women with leukemia almost invariably give birth to nonleukemic offspring. Cooke (27) has reported the development of acute leukemias in fraternal
male twins, at 17 and 22 months of age, respectively; of possible significance is the fact that the mother had a toxemia with edema and albuminuria during this pregnancy.

Infection.—In about 50 per cent of all cases, the acute leukemias of childhood are associated with a history of severe, usually nonspecific infection antedating the apparent onset of leukemia by a few weeks or months (25, 26, 122). Some observers have suggested that these children suffer from an abnormal host response reflected in their inability to recover promptly from such infections (34, 192). While there is little disagreement that infection is a frequent clinical manifestation in acute childhood leukemia, there are at least two prevailing interpretations of this fact. Cooke (26), and others (16, 78) have regarded infection as a major etiologic factor in the acute leukemias of childhood and have pointed to the similarity of the age distribution of miscellaneous pediatric infectious diseases and that of acute leukemia. Furth, Ferris and Reznikoff (41) and others (48) have considered that leukemia is already present in occult form in such cases and that the infection is merely its first clinical manifestation, as a sequel to a lowering of systemic resistance. Furth has emphasized the essentially neoplastic nature of leukemia in both man and experimental animals. It is of course possible to reconcile the infectious and neoplastic concepts, since there is no reason why an infection cannot play a causal role in the induction of a neoplasm.

Apart from the possibility that nonspecific infection may conceivably be causally related to acute childhood leukemia, there have been a number of recent claims that human leukemia is caused by a virus (47, 106, 186). Typical of the evidence adduced for this view is the report by Magrassi and his associates (99, 100) that the inoculation of human leukemic cells into guinea pigs regularly elicits a severe, usually fatal anemia after a variable incubation period. At autopsy, the animals exhibit severe degenerative lesions in the parenchyma of the liver, kidney, adrenals, and cutaneous epithelium, together with marked proliferation of histiocytes and of the reticuloendothelial system. This disease is transmissible after intracerebral passage through cats and with liver and spleen cell suspensions from affected guinea pigs. Similar evidence for the presence of a viral agent in patients with Hodgkin's disease has been reported in recent years by Bostick (18) and others (47). Such evidence of course cannot really establish the viral etiology of human leukemia or Hodgkin's disease, since individuals with these conditions are notoriously susceptible to secondary infection, and viruses detected in this way may justifiably be regarded as mere contaminants, at least until it can be shown that one particular virus is regularly recoverable from any given variety of leukemia.

Antibiotics and chemotherapeutic medications.—The widespread use of chemotherapeutic and antibiotic agents in the treatment of infectious disease has excited suspicion that some of these agents may have leukemogenic activity, particularly since they have been responsible for a significant number of cases of severe pancytopenia with generalized marrow injury (29, 98, 105). Since they are now prescribed almost universally, and their administration is only rarely followed by untoward sequelae, some other factor, such as individual or hereditary predisposition, must also be invoked to explain the occasional cases of pancytopenia or leukemia associated with their use.

The Preleukemic State

Finally, it seems worth while to inquire into the pathogenesis of the leukemias in the hope that some clue to their etiology may come forth. Here again, the evidence is discouragingly meager and perhaps open to the criticism that some cases have been selected for special attention because of the existence of some other blood disorder prior to the onset of leukemia. Block, Jacobson, and Bethard (12) studied twelve patients with heterogeneous blood disorders characterized by anemia, neutropenia, and/or purpura. Ten of these developed acute myelogenous or stem-cell leukemia, the earliest signs of which appeared from 3 to 17 months after they were first seen. All had one or more bone marrow studies during the preleukemic phase of their disease. Two presented an aplastic marrow, two had marrows of normal cellularity but with a maturation arrest of the granulocytic series, and six had a hypercellular marrow, usually with a disturbance in cellular differentials. A feature of several cases was a distinct monocytosis in the peripheral blood for an appreciable period before leukemia became manifest. Three of the patients had a definite history of medication which may have precipitated their initial blood disorder, and three others exhibited the marrow findings often seen in toxic agranulocytosis due to drugs.

A number of other observers (34, 55, 101, 105, 110, 147) have reported the development of acute myelogenous leukemia in patients with an initial picture of pancytopenia, anemia, or neutropenia, and evidence of either gross hypoplasia (7) or hyperplasia of the marrow with maturation ar-
rest. One of the best documented of these reports is that of Black and Meynell (10), concerning a young girl who developed a fulminating pancytopenia with marrow hypoplasia following oral sulfadiazine treatment for sore throat. Following a complete return of the blood to normal 2 months later, she remained in apparent good health for 5 months, and then developed a rapidly fatal acute myelogenous leukemia, 7.5 months after the onset of her illness. That nonspecific hypersensitivity reactions may cause severe marrow injury leading to leukemia is suggested by the case reported by Strauss (133), in which this sequence of events abruptly followed a second vaccination against typhoid and paratyphoid.

In recent studies for the Atomic Bomb Casualty Commission, Moloney and Lange (112) studied four patients for periods of 5–37 months prior to the development of myelogenous leukemia. All four cases had been within 1,500 meters of the bomb hypo-center at Hiroshima. These patients exhibited mild, variable leukocytosis on routine surveys; careful study revealed increased numbers of basophils in the peripheral blood. Platelets were usually increased, sometimes strikingly, but anemia was not a prominent feature. Bone marrow studies were carried out in three of the four cases revealing a tendency to hypercellularity and myeloid immaturity but an essentially nonspecific picture during this prodromal phase. It is noteworthy that basophilocytosis was also a prominent feature in two cases of benzol-induced chronic myelogenous leukemia studied serially by Bousser and Tara (14).

The myelogenous leukemias which develop in patients with chronic benzol poisoning tend to be superimposed upon an aplastic anemia, agranulocytosis, or hemorrhagic syndrome of variable duration, in which the marrow may be either severely hypoplastic or paradoxically hyperplastic but associated with a maturation arrest, particularly in the granulocytic series (15, 35, 60, 102.)

There is clearly a great need for additional clinical studies of the preleukemic state for each type of leukemia. The paucity of information is greatest for lymphatic leukemia and the malignant lymphoid tumors. The evidence cited would suggest that leukemia does not as a rule arise de novo in individuals with a previously normal hematopoietic system. Instead, patients studied prior to the onset of leukemia have exhibited definite disturbances in the peripheral blood and bone marrow, although the type of disturbance is not always the same. It is of course possible that these cases are exceptional and are unduly emphasized because the pre-existence of another blood disorder has made possible their observation prior to the onset of leukemia.

Biochemical characterization of the leukemic and preleukemic hematopoietic tissues is also beginning to take shape from a number of recent clinical and experimental studies (6, 9, 18, 57, 112, 129, 138). These studies reveal that there are interesting aberrations in certain enzyme activities, especially those of the phosphatases, and other biochemical anomalies in these malignant or pre-malignant tissues.

**Epidemiology**

There is very little published information concerning the epidemiology of leukemia (20, 46, 124). A sex differential in incidence appears to exist, with leukemias, lymphosarcomas, and Hodgkin's disease all about 1.3–1.7 times more common in men than in women (46, 128). There is also a distinctly lower incidence in the nonwhite United States population. In 1950, for example, the leukemia death rate per 100,000 in nonwhite males was 3.6, as compared with 7.1 in white males (119). This is merely a crude death rate, however, and may not reflect a real difference in susceptibility to the disease, since there is a shorter average duration of life in the nonwhite population, whereas the leukemia rate rises steeply in the population over 60–70 years of age. There is also an apparently lower rate in the Southern states than in the Northern states (Table 2). Again, this is a crude death rate and may merely reflect differences in accuracy of reporting, in average length of life, in proportions of white and nonwhite population, etc., rather than a real difference conceivably due to climate or other environmental factors. There is clearly a tremendous need for careful epidemiologic and demographic studies with regard to the leukemias, lymphosarcomas, and Hodgkin's disease in the United States and elsewhere. The most recent revision of the international list of Causes of Death provides a much more satisfactory breakdown of this group of conditions than has previously been available and will undoubtedly help to make epidemiological studies more meaningful in the future.

**WHAT IS KNOWN OF THE ETIOLOGY AND PATHOGENESIS OF LEUKEMIA AND THE MALIGNANT LYMPHOID TUMORS IN EXPERIMENTAL ANIMALS?**

This group of diseases has been studied systematically in mice, poultry, and to a limited extent in rats. The disease in birds has long been known to be caused by a virus which is readily

---

1 Dr. William C. Moloney very kindly made this material available for review in manuscript form.
recoverable from leukemic cells and apparently transmissible extra-chromosomally in the egg (28). The fact that mammalian leukemia has for many years frustrated the attempts of investigators to demonstrate a similar viral agent has led to the view that avian leukemia may be a special case with little or no etiologic relationship to the human disease. This has been responsible for a diminution of interest in experimental avian leukemia in the past 20 years which has perhaps been unjustified and unfortunate.

**Nature of Mouse Leukemia**

Most experimental investigations on leukemia have been carried out in the mouse. It seems pertinent, therefore, to inquire critically to what extent the diseases which have been studied in the mouse may be the morphological or clinical counterparts of human leukemias and malignant lymphoid tumors.

Myelogenous leukemia occurs spontaneously in some strains of mice (82), and its incidence may be augmented by x-radiation (42). The majority of leukemias which occur in mice are, however, lymphatic, and most experimental studies in recent years have really been concerned with lymphosarcoma, during the course of which the peripheral blood and bone marrow may be invaded in a variable but generally minor fraction of all instances (64, 82).

Because some transplanted leukemias have acquired a rather fulminating course, mouse leukemias have sometimes been termed acute. Most spontaneous or induced lymphatic leukemias, however, are probably subacute or chronic, as judged by morphology and survival. Moreover, there is no murine counterpart to human acute childhood leukemia. It is imperative to keep in mind these limitations to the direct human applicability of mouse leukemogenesis. Nonetheless, for corresponding forms of leukemia and lymphosarcoma in man and mouse, there are striking morphological and clinical similarities, as was pointed out many years ago by Furth, Ferris, and Reznikoff (41). These similarities encourage the hope that our studies in mice may have some applicability in man as well.

Lymphoid diseases which are morphologically identical with or very closely similar to human reticulum-cell sarcoma and Hodgkin’s disease have also been encountered in mice and have recently been carefully characterized by Dr. Thelma Dunn. However, little experimentation has yet been done with these tumors, and nothing is known about their etiology.

**Mechanism of Leukemogenesis in the Mouse**

The balance of this presentation will be devoted to an analysis of the factors which have thus far been shown to play a part in the very complex induction mechanism of lymphatic leukemia and lymphosarcoma in the mouse. Several aspects of the subject have been reviewed elsewhere in recent years (40, 79) and will not be extensively treated here. The experimental literature to date will be discussed under four headings.

**Non-specific constitutional and environmental factors.**—Genetic constitution is of great importance in relation to leukemia susceptibility in mice. In addition to the spontaneous occurrence of a high incidence of leukemia in certain strains, it has been shown by Kirschbaum and Mixer (80) that strains which exhibit little or no spontaneous leukemia may be highly susceptible to the leukemogenic action of one or more classes of exogenous agents, the pattern of susceptibility or refractoriness to any specific agent being conditioned by genetic constitution.

Age is a factor of importance; animals exposed to whole-body x-radiation when prepubertal develop a far higher incidence than older animals.

### Table 2

<table>
<thead>
<tr>
<th>Region</th>
<th>LEUKEMIA Total deaths</th>
<th>Rate/100,000</th>
<th>LYMPHOSARCOMA Total deaths</th>
<th>Rate/100,000</th>
<th>Hodgkin’s disease Total deaths</th>
<th>Rate/100,000</th>
</tr>
</thead>
<tbody>
<tr>
<td>New England (6)*</td>
<td>608</td>
<td>6.5</td>
<td>255</td>
<td>2.7</td>
<td>217</td>
<td>2.3</td>
</tr>
<tr>
<td>Middle Atlantic (3)</td>
<td>1,852</td>
<td>6.4</td>
<td>689</td>
<td>3.0</td>
<td>556</td>
<td>1.9</td>
</tr>
<tr>
<td>East North Central (6)</td>
<td>1,905</td>
<td>6.2</td>
<td>737</td>
<td>2.4</td>
<td>538</td>
<td>1.8</td>
</tr>
<tr>
<td>West North Central (7)</td>
<td>1,045</td>
<td>7.4</td>
<td>509</td>
<td>2.2</td>
<td>264</td>
<td>1.9</td>
</tr>
<tr>
<td>South Atlantic (9)</td>
<td>985</td>
<td>4.4</td>
<td>539</td>
<td>1.6</td>
<td>277</td>
<td>1.3</td>
</tr>
<tr>
<td>East South Central (4)</td>
<td>506</td>
<td>4.4</td>
<td>178</td>
<td>1.5</td>
<td>145</td>
<td>1.3</td>
</tr>
<tr>
<td>West South Central (4)</td>
<td>725</td>
<td>5.0</td>
<td>945</td>
<td>1.7</td>
<td>167</td>
<td>1.1</td>
</tr>
<tr>
<td>Mountain (8)</td>
<td>280</td>
<td>5.5</td>
<td>96</td>
<td>1.9</td>
<td>75</td>
<td>1.5</td>
</tr>
<tr>
<td>Pacific (5)</td>
<td>809</td>
<td>6.2</td>
<td>409</td>
<td>2.8</td>
<td>268</td>
<td>1.9</td>
</tr>
<tr>
<td></td>
<td>8,845</td>
<td>5.9</td>
<td>3,467</td>
<td>2.3</td>
<td>2,537</td>
<td>1.7</td>
</tr>
</tbody>
</table>

* The numbers in parentheses are literature citations.
need of careful repetition in several strains of mice, for careful experimental studies with a number of drugs and chemicals to which humans are occupationally or environmentally exposed and which have at least occasionally been responsible for the appearance many months after the animal has been exposed to x-radiation, methylcholanthrene, or estrogen? It is in this area that most rapid progress has been made in recent years. The thymus, the bone marrow, and the hormonal factors which affect lymphoid tissue growth and involution appear to be interrelated in a complex manner which has a decisive influence upon the leukemia induction process.

**Exogenous leukemogenic agents.**—It is now well established that x-rays and other ionizing radiations (42, 64), methylcholanthrene and other hydrocarbons (86, 116), and estrogens (43, 45, 84) will induce lymphatic leukemia and lymphosarcoma in susceptible strains of mice. Shay and his associates (197) have shown that methylcholanthrene is also an effective leukemogenic agent in rats. That benzol may also be leukemogenic for mice was suggested by the work of Lignac (94). This has not been fully substantiated and is in need of careful repetition in several strains of mice, since it may provide a method for the selective study of myelogenous leukemia.

Not much is known about other classes of agents. Nitrogen mustard has not caused leukemia or lymphosarcoma in C57BL mice in two experiments to date. There appears to be a need for careful experimental studies with a number of drugs and chemicals to which humans are occupationally or environmentally exposed and which have at least occasionally been responsible for serious hematopoietic injury. Dameshek (29) suggests the sulfonamides, aniline dyes, and arsenicals as deserving of study. Certain newer drugs such as phenylbutazone (58, 132), mesantoin (4, 134), and chloramphenicol (90, 56, 98) may cause aplastic anemia and/or agranulocytosis in man, which would suggest their inclusion in such an experimental program.

In a series of recent papers, Gross (51, 52) has reported that the apparently spontaneous occurrence of a very high incidence of lymphatic leukemia in AK mice is in fact due to a filtrable agent, presumably a virus, which is transmissible heterologously to young mice of the CSH strain, in which it reportedly causes the appearance many months later of lymphatic leukemias which are apparently derived from CSH tissue and not merely due to delayed growth of AK leukemic cell transplants. There is at present no evidence to indicate whether this observation, which is not yet confirmed, is of general significance or merely a peculiarity of the strains mentioned. There is also no evidence that such a virus-like agent is liberated or activated in low-leukemia strains by treatment with x-rays, estrogens, or methylcholanthrene. This possibility deserves careful experimental study.

Leukemia incidence is related to the dose of the exogenous agent employed, and there is a minimal effective dose below which no detectible effect is exerted by the agent (71). In the case of x-radiation, it appears that multiple irradiations spaced several days apart are appreciably more effective than a single massive exposure (71). The mechanism whereby this periodicity operates has not yet been clarified. That the various exogenous agents operate through a common mechanism is clearly established by the studies of Kirschbaum and his co-workers (81), who have demonstrated synergism of any two external agents to which a given strain of mice is independently susceptible.

**Specific intermediate mechanisms.**—What do we know about the sequence of events that occur after the animal has been exposed to x-radiation, methylcholanthrene, or estrogen? It is in this area that most rapid progress has been made in recent years. The thymus, the bone marrow, and the hormonal factors which affect lymphoid tissue growth and involution appear to be interrelated in a complex manner which has a decisive influence upon the leukemia induction process.

a) The thymus: The thymus has been shown to be the site of origin for the lymphosarcomas and lymphatic leukemias which occur in several mouse strains, although there are strains in which no thymic predilection exists (45, 64, 89). Rask-Nielsen has shown that even the remote implantation of carcinogenic hydrocarbons may cause the appearance of lymphomas in the thymus (123). The relative rarity of the thymic origin of lymphatic leukemia in man is probably associated with the relatively rapid and complete involution of the human thymus at puberty. There are well documented cases (8, 24) of acute lymphatic leukemia developing in children with a pre-existing thymic lymphoid tumor mass. The lymphoid organs in general appear to be more prominent in mice than in men, and this is also reflected in the relatively high lymphocyte counts of mice. The mouse thymus involutes slowly through the 1st year of life. Perhaps these species differences can be invoked to explain the prevalence of lymphatic...
over myelogenous leukemia in the mouse, and the important role of the thymus in its development.

It was first shown by Furth and his associates (109) that thymectomy may virtually abolish lymphatic leukemia in a high leukemia strain of mice, and this finding has been confirmed by other investigators (67, 89, 90). Thymectomy also inhibits leukemogenesis in strains in which the thymus is not the site of origin of the disease (89). Law and Miller have shown (88–90) that re-implantation of the thymus can restore susceptibility to thymectomized animals, and we have reported that susceptibility is at least partially restored when homologous thymus glands are implanted into thymectomized C57BL mice at the completion of their course of total-body irradiation (74). More recently, we have found that in this strain the tumors appear to arise in nonirradiated thymic transplants, by virtue of their residence in the irradiated host, suggesting that the mechanism of lymphoid tumor induction is an indirect process (73). The fact that the thymus need not be directly exposed to the carcinogenic agent is of course difficult to reconcile with the recently revived somatic mutation theory of carcinogenesis (36). Although much remains to be clarified, it would appear that the thymus plays a dual role in relation to lymphosarcoma development: in some strains, it appears to be the site of origin of the disease, and in many and possibly all strains, it appears to furnish a factor which contributes to the induction process, even when the disease arises elsewhere.

b) The bone marrow factor: A few years ago it was shown that, although the lymphosarcomas induced by total-body irradiation of C57BL mice arose in the thymus, local irradiation over the thymic region was ineffective (66). It has since been shown that a striking inhibition of lymphoid tumor induction can be brought about by merely shielding one hind leg (68) and that a similar inhibition can be achieved by injecting normal homologous bone marrow cell suspensions intravenously into unshielded irradiated animals (75). Lorenz and his associates have also shown (97) that spleen shielding inhibits leukemogenesis in this strain. These findings obviously bear a very close relationship to the observation by Jacobson and associates (89) that spleen shielding and the intraperitoneal implantation of spleens following irradiation are highly effective in preventing acute radiation death in mice.

In more recent work, three different end-points have been employed in an attempt to further characterize this factor in mouse bone marrow and spleen. These end points are: acute radiation mortality, regeneration of thymic weight following systemic irradiation, and lymphoid tumor incidence. Reports by Cole, Fishier, and Bond (21), Lorenz and his colleagues (23, 95, 96), Jacobson and his group, and studies from our laboratory have established the following additional facts with regard to the active principle: It is radiosensitive; is destroyed by lysis of marrow cells; is present in ground bone; and has thus far not been extractible in active form from intact marrow cells or their nuclei. The mechanism and site of its action are still entirely obscure, but it appears to be essential for the regeneration of the hematopoietic tissues, including the thymus, following severe irradiation injury (17, 72, 75).

Hormonal factors.—The action of estrogen as a leukemogenic agent in mice is still unexplained. The effects of the other hormones appear, however, to fit into a distinct pattern which parallels their effect on thymic growth and involution. Since the experimental evidence relating to these hormonal influences has been extensively reviewed elsewhere (38, 77), only a summary of the evidence to date will be given here.

With spontaneous or induced lymphoid tumor incidence as a criterion, ovariectomy has usually been inhibitory or without effect (3, 87, 117). Progesterone, which has moderate thymolytic activity in rather high doses, mildly inhibits the tumor process.6 Orchiectomy is followed by a shorter latent period, greater incidence, or both in virtually all strains thus far studied (3, 87, 117), suggesting that endogenous testosterone secretion is inhibitory. This is supported by the fact, noted by several observers (44, 69, 70, 117), that exogenous testosterone administration may dramatically suppress the tumor process. Testosterone is a very active thymolytic agent which differs in its action from the adrenal cortical steroids in affecting only the thymus, with little or no detectible effect on the lymph nodes and spleen (77).

The important role of the adrenal cortex and of pituitary ACTH in regulating lymphoid tissue growth and involution is now well known. Cortisone and hydrocortisone, which are both powerfully thymolytic (39), very significantly depress lymphoid tumor development at suitable dose lev-

6 Some of the conclusions presented are based upon unpublished experimental data.

6 C. S. Nagareda and H. S. Kaplan, unpublished data.
Desoxycorticosterone acetate, which has little or no effect on thymic weight at ordinary dose levels, is without effect on tumor incidence (76). Adrenalectomy is followed by a distinct increase in relative and absolute thymic weight and by a corresponding increase in lymphoid tumor incidence in irradiated C57BL mice (76, 87). Recent unpublished studies indicate that thigh shielding confers protection on adrenalectomized mice just as effectively as on intact mice with respect to lymphoid tumor incidence (Table 3).

The only hormone other than estrogen which has any carcinogenic activity for lymphoid tissue is pituitary growth hormone, the chronic administration of which was shown by Moon and Li (113) to yield localized lymphosarcomas of pulmonary origin in rats. This response was abolished by hypophysectomy (115). Hypophysectomy has also been reported to inhibit the induction of other types of tumors (37, 50, 114), suggesting that the pituitary might be essential for neoplasia in general. That this is not the case for lymphoid tumor induction in irradiated C57BL mice is indicated by the outcome of a recent experiment in our laboratory (118). These data support the conclusion that the pituitary is not essential for the maintenance of relative thymic and lymph node weight, for regeneration of the irradiated thymus, or for the development of lymphosarcoma in this strain.

Little is known about the role of thyroid function in leukemogenesis. In a single experiment, Dr. Sumner Marder failed to influence lymphoma incidence significantly by chronic administration of thyroxine (77).

With the exception of the estrogens, the action of which is unique, it may be seen (Table 4) that any hormonal change which tends to cause significant thymic involution also inhibits lymphoid tumor induction and, conversely, any hormonal change which tends to augment thymic weight also augments lymphoid tumor incidence. Moreover, the intensity of effect on tumor incidence seems directly proportional to the intensity of effect on thymic weight. It seems clear that the growth and involution of the thymus in the mouse are under the control of a rather intricately bal-

![Chart 1. Effect of hydrocortisone on development of radiation-induced lymphoid tumors in C57BL mice](chart_1.png)

**Chart 1.**—Effect of hydrocortisone on development of radiation-induced lymphoid tumors in C57BL mice

**Table 3**

<table>
<thead>
<tr>
<th>EFFECT OF THIGH SHIELDING ON RADIATION-INDUCED LYMPHOID TUMOR INCIDENCE IN ADRENALECTOMIZED AND INTACT C57BL MICE</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>GROUP</strong></td>
</tr>
<tr>
<td>-----------</td>
</tr>
<tr>
<td>I</td>
</tr>
<tr>
<td>II</td>
</tr>
<tr>
<td>III</td>
</tr>
<tr>
<td>IV</td>
</tr>
</tbody>
</table>

*Four doses of 100 r each at eight-day intervals.*
anced homeodynamic equilibrium in which hormones play an important part. The level at which thymic weight will be in equilibrium may be set, much in the same manner in which a thermostat is set, either up or down by such endocrine manipulation.

If we now go back to pick up the lines of evidence previously described, we find that they can be harmonized in the following hypothesis (Chart 2): When the thymus is accidentally injured, as by x-radiation, transplantation, etc., its controlling equilibrium is disturbed and the thymus is subjected to a hypothetical proliferative stimulus whose intensity will be measured by the disparity between normal thymic weight and the weight to which the thymus has been reduced by the injury. If the bone marrow factor which the thymus requires for its regeneration has been destroyed by irradiation or the other exogenous agents, the thymus is unable to respond to this regenerative influence; it is under these circumstances that lymphosarcoma and lymphatic leukemia result. If the bone marrow factor has been uninjured, or is supplied from exogenous sources, the thymus will be enabled to recover, and, if equilibrium is restored with reasonable promptness, few or no lymphoid tumors will occur. Even when the bone marrow factor is unavailable, tumor induction may be prevented by the administration of thymolytic hormones, which have the effect of moving the hypothetical thermostat downward and depressing the intensity of the regenerative stimulus on the thymus. Conversely, the number of tumors may be augmented by endocrine manipula-

![Chart 2: Schematic diagram of lymphosarcoma induction mechanism](chart2.png)

### Table 4

<table>
<thead>
<tr>
<th>Endocrine factor</th>
<th>Physiological effect on thymus</th>
<th>Effect on lymphoma incidence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Testosterone</td>
<td>Marked involution</td>
<td>Marked inhibition</td>
</tr>
<tr>
<td>Hydrocortisone</td>
<td>Marked involution</td>
<td>Marked inhibition</td>
</tr>
<tr>
<td>Cortisone</td>
<td>Moderate involution</td>
<td>Moderate inhibition</td>
</tr>
<tr>
<td>Progesterone</td>
<td>Mild involution</td>
<td>Mild inhibition</td>
</tr>
<tr>
<td>Desoxycorticosterone</td>
<td>No effect</td>
<td>No effect</td>
</tr>
<tr>
<td>Hypophysectomy</td>
<td>No effect (relative weight)</td>
<td>No effect</td>
</tr>
<tr>
<td>Thyroxine</td>
<td>No effect</td>
<td>No effect</td>
</tr>
<tr>
<td>Orchietectomy</td>
<td>Moderate hypertrophy</td>
<td>Moderate augmentation</td>
</tr>
<tr>
<td>Adrenalectomy</td>
<td>Marked hypertrophy</td>
<td>Marked augmentation</td>
</tr>
<tr>
<td>Estrogen</td>
<td>Involution</td>
<td>Tumor induction</td>
</tr>
</tbody>
</table>

The above table lists the effects of various endocrine factors on thymic weight and lymphoma incidence.
tions having a thymotrophic effect, such as adrenalectomy or castration.

This hypothesis assigns a rather limited significance to the exogenous agents, which are thought of as merely producing a special kind of disturbance of growth equilibrium by virtue of their injurious effects upon the thymus and bone marrow. In effect, the hypothesis suggests that the real carcinogenic agent is the normal endogenous growth stimulus for the thymus acting over a prolonged period upon the injured gland under circumstances in which it cannot respond normally. It is tempting to speculate on the possible role of cellular or organ-specific materials akin to the embryonic inducers and organizers in such situations of induced growth imbalance (59, 141, 143).

The neoplastic cellular transformation.—The evidence already presented points clearly to the fact that no sudden irreversible cellular change takes place at the time of exposure to a leukemogenic agent. Instead, these agents, by precipitating certain types of injurious disturbance, initiate a train of events during the course of which an irreversible change does finally take place. The exact time of this change has not been defined, but, since established lymphosarcomas are demonstrable as early as 8 weeks after irradiation or methylcholanthrene treatment (74, 108), it seems clear that the initial malignant change in one or more cells within the thymus must occur appreciably earlier in the most susceptible animals. In recent unpublished studies, we have deferred the injection of intravenous bone marrow for various time intervals after irradiation in an attempt to study this question. Our data to date suggest that the time for 50 per cent loss of the inhibitory activity of the marrow occurs at between 3 and 4 days after exposure to radiation (Chart 3). Essentially complete loss of inhibition by the injected marrow occurs after 8 days. Further data on the average time interval required for the neoplastic transformation would be of tremendous help by making it possible to focus our attention upon a part of the tumor induction process which is its very core.

The irreversible neoplastic cellular change is heritable from one generation of tumor cells to the next. Whether it is a genic or a cytoplasmic change is unknown, although a considerable body of experimental and theoretical evidence (93, 107) would suggest that a cytoplasmic change is more likely. It is perhaps at this point that the current ideas about virus action, plasmagenes, and induced abnormalities in nucleic acid metabolism (32, 120, 121, 144) will all be brought together and harmonized as our knowledge unfolds (31, 54, 88, 107, 142).

**SUMMARY**

1. The incidence of leukemia appears to be increasing rapidly.
2. The leukemias probably do not all have the same etiology, although the mechanisms by which the various forms arise may well have much in common.
3. To account for the bulk of human leukemias, it would seem necessary to widen our horizons beyond such agents as radiation and benzol and...
to look with suspicion as a possible inciting agent on any chemical, drug, or body reaction, such as hypersensitivity, which is capable of causing severe injury to hematopoietic tissue.

4. We are in need of additional careful studies of the preleukemic phase, as well as of the epidemiology of the leukemias; the fragmentary information now available indicates that the leukemias do not spring abruptly from previously normal tissues but burst into flame, as it were, from a smoldering pre-existent hematopoietic disorder of varying origin and morphology.

5. Whereas most human evidence pertains to myelogenous leukemia, experimental work to date has been primarily concerned with lymphatic leukemia and lymphosarcoma. The evidence reviewed established the fact that the induction mechanism is an indirect process reflecting the complex interrelationship of multiple factors. A hypothesis has been suggested which is consistent with this evidence and which implicates a chronic disturbance of normal tissue growth equilibrium as the major factor in leukemogenesis.

REFERENCES


51. GRIFFIN, A. C.; RINFHET, A. P.; and COHSIGLIA, V. F. The Inhibition of Liver Carcinogenesis with 3'-Methyl-4-dimethylaminoazobenzene in Hypophysectomized Rats. Cancer Research, 13:77-78, 1953.


86. Law, L. W. The Induction of Leukemia in Mice Following Percutaneous Applications of 9,10-Dimethyl-1,2-Benzanthracene. Cancer Research, 1:594-71, 1941.


88. ——. Increase in Incidence of Leukemia in Hybrid Mice Bearing Thymic Transplants from a High Leukemia Strain. Ibid., 12:799-807, 1950.


90. ——. The Influence of Thymectomy on the Incidence of Carcinogen-Induced Leukemia in Strain DBA. Ibid., pp. 482-83.


96. Lorenz, E.; Condgon, C. C.; and Uphoff, D. Modification of Acute Irradiation Injury in Mice and Guinea-Pigs by Bone Marrow Injections. Radiology, 58:683-77, 1952.


On the Etiology and Pathogenesis of the Leukemias: A Review

Henry S. Kaplan


Updated version
Access the most recent version of this article at:
http://cancerres.aacrjournals.org/content/14/8/535

E-mail alerts
Sign up to receive free email-alerts related to this article or journal.

Reprints and Subscriptions
To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

Permissions
To request permission to re-use all or part of this article, contact the AACR Publications Department at permissions@aacr.org.