creased pressure on the epicardial surface and due to a decrease in pressure.

Summary. Dogs were subjected to various procedures in an attempt to clarify several existing theories about the electrocardiographic findings in pericarditis or cardiac tamponade. In this study changes in P- and R-wave amplitude are related to decreased arterial pressure and increased central venous pressure. Problems related to volume conduction are not involved in the decreased R-wave amplitude. The presence of additional fluid in the pericardial space results in S-T segment shift and T-wave inversion. Changes in the S-T segment require the additional factor of a systemic pressure change whereas the latter is independent of pressure changes. The shift in anatomical axis of the heart in a direction which impinges on the venous return to the right atrium will cause P- and R-wave changes and may be an important factor in the etiology of the electrocardiographic changes. Coronary insufficiency, on the basis of these studies, is not thought to be contributory to early electrocardiographic changes but may be of significance in later stages of pericarditis.

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Effect of Thymectomy in Newborn Rats Infected with Moloney Virus.* (30853)

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Thymectomy in mice as late as 31-71 days of age reduces the incidence of spontaneous leukemia in a high-leukemia stock of mice (1). Similarly, thymectomy reduces the incidence of induced leukemia in mice injected with a leukemogenic virus(2,3). Only a small percentage of mice thymectomized at 3-4 weeks of age and inoculated with a leukemogenic virus either as newborns or after thymectomy develop leukemia, and then only after a long latent period(4-6). Gross(7) reported that rats injected as newborns with Gross virus, passage A, and thymectomized at 10 days of age, are somewhat resistant to induction of leukemia but to a much lesser extent than are thymectomized mice. The present study was initiated to determine if thymectomy of rats at 24 hours of age, and prior to virus inoculation, would increase

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their resistance to a greater degree than that found by Gross in rats thymectomized at 10 days of age. Before completion of this present study, Kunii *et al*(8) reported on similar studies using the Gross virus in Sprague-Dawley rats. This report therefore confirms their findings using a different virus-host system, *i.e.*, Moloney virus and inbred Osborne-Mendel rats. In addition, serum protein patterns of thymectomized and intact rats were studied and an examination made for presence of viremia in the thymectomized group of animals.

Materials and methods. Animals. Inbred Osborne-Mendel rats (O/M) and inbred Balb/c mice, originally obtained from Dr. John Moloney and Dr. H. B. Andervont respectively, and propagated in our laboratory by brother-sister mating, were used in this study.

Thymectomy. Thirty-three rats were thymectomized using the method described by Jankovic *et al*(9) when less than 24 hours of age; 22 intact rats were kept as virus inoculated controls, and 10 intact rats as normal uninoculated controls. All rats were weighed weekly.

Virus injection. Moloney virus, 0.05 ml of undiluted passage RT1, was injected intra-abdominally into thymectomized rats and into intact virus control rats at 36-48 hours of age. Passage RT1 was the first rat tissue passage, prepared according to the method of Moloney(10) from a rat plasma passage (LTTR(RV)TR38) obtained from Dr. John Moloney.

Test for viremia. Plasma from each thymectomized rat either at time of autopsy or at 6 months of age (whichever occurred first) was injected intra-abdominally (0.05 ml) into newborn Balb/c mice, one litter for each rat plasma. Induction of malignant lymphoma in the mice as observed grossly by enlarged spleen, thymus and lymph nodes (5-20 times normal size) was taken as evidence of viremia in the rats.

Serum protein patterns. Immunoelectrophoresis (IEP) was done according to the method of Scheidegger(11), 5 ma/slide, barbital buffer, pH 8.2, μ 0.05, 2 hours at 5°C. Patterns were developed in the cold, using rabbit gamma globulin (obtained from rabbit antiserum by three $(NH_4)_2SO_4$ precipitation at $\frac{1}{3}^d$ saturation) prepared against inbred O/M rat serum.

Pathology. All rats were euthanized when moribund or at termination of the experiment, and complete autopsies were performed. Spleen and thymus weights were recorded; all enlarged lymph nodes, the thymus, and the spleen were fixed in Carnoy solution. All fixed tissues were sectioned, stained and examined microscopically. Histologic diagnoses of malignant lymphoma were based on the following criteria: anaplasia of lymphoid cells with an increase in mitotic index, partial to complete obliteration of the normal architecture of the organ involved and invasion of the stroma and capsule.

Careful dissection and gross examination were made of the posterior cervical and anterior mediastinal regions of thymectomized rats prior to fixation of the tissue. Thin blocks of tissue from the entire anterior mediastinal region of thymectomized rats were sectioned at various levels to insure detection of residual thymic tissue.

Results. Induction of leukemia in intact and thymectomized rats. In the group of intact rats, consisting of 22 animals, 19 died over a period of 2½ to 7 months, with average age at death of 4½ months. On microscopic examination all 19 rats had obvious changes of malignant lymphoma in the thymus, 17 of these also had involvement of the spleen, and 18 involvement of lymph nodes. The 3 remaining rats were autopsied at 7½ months of age. Microscopically 2 had malignant lymphomatous changes in the thymus, while the organs of the third rat showed no significant lesions. Thus, 21/22 or 95% developed malignant lymphoma in the thymus; 17/22 or 77% of which also had splenic involvement, and 18/22 or 82% lymph node involvement. Of 33 rats thymectomized as newborns, 17 rats died within 2-5 days either from maternal neglect or from the surgery. Of the 16 surviving rats 2 were incompletely thymectomized as evidenced by lymphomatous thymic tissue microscopically observed in autopsy tissue. This report, therefore, is concerned with the 14 completely thymecto-

TABLE I. Effect of Injection of Moloney Virus* into Intact and Thymectomized Rats.

Treatment	Pathology†			Positive‡ mouse test	Immunoelectrophoresis IgA line			IgG
	Thymus	Spleen	LN		Present	Faint	Absent	Abnormal§
Thymectomy virus injection	—	‖3/14	1/14	13/13	2/13	5/13	6/13	8/13
Intact virus injection	21/22	17/22	18/22	n.d.	2/15	3/15	10/15	10/15
Intact, no injection	0/10	0/10	0/10	n.d.	10/10			

* 0.05 ml of Moloney virus, passage RT1, was injected intra-abdominally into 48-hr-old rats.
† No. developing malignant lymphoma/total No. inoculated.
‡ Rat plasma when injected into newborn mice induced lymphoma.
§ An IgG line 2 mm or less shorter than that of the normal serum electrophoresed on the same slide was scored normal. Those scored as abnormal were3 to 11 mm shorter than the normal IgG line.
‖ Two of 3 positives in this group were diagnosed as reticulum cell sarcomas.

mized rats. Six of these rats died, one at 2½ months (a runt which never grew normally); one at 3½ months (pneumonia); one at 5 months (middle ear infection); 2 at 6½ months (one diagnosed as subacute focal interstitial pneumonitis and subacute splenitis), and one at 7 months. On microscopic examination of spleens and lymph nodes, one rat had reticulum cell sarcoma in the spleen (rat dying at 7 months); the rest showed no malignant lymphomatous changes. The 8 remaining rats were killed at 7½ months. One had malignant lymphomatous changes in the spleen and lymph nodes; one had reticulum cell sarcoma of the spleen. The other rats showed no significant lesions in the organs examined. Thus, over the period of the study, 3/14 or approximately 21% developed splenic neoplasms, one lymphoma and 2 reticulum cell sarcomas. If only the 12 rats surviving longer than the average length of time for the intact rats (4½ months) are considered, then the percentage of neoplasm is increased with 3/12 or 25% showing neoplasms of the spleen, *i.e.*, 1/12 or 8% with malignant lymphoma and 2/12 or 17% with reticulum cell sarcoma.

Test for viremia. All the plasma from the thymectomized rats upon injection into mice induced lymphoma, including plasma from those rats diagnosed as reticulum cell sarcoma. The diagnosis of malignant lymphoma in the latter group of mice was determined microscopically. Since 95% of the intact rats died of lymphoma, it was not considered necessary to check for viremia in this group.

Serum protein patterns. On IEP analyses, the rabbit anti-rat serum gamma globulin formed with normal adult rat serum, an albumin line, 2 fast moving α_1 lines, 4-5 α_2 lines, 2 β lines (one probably transferrin), one IgG line and one IgA line, the latter a line lying inside the curvature of the IgG line (12). No line corresponding to an IgM line was observed with this gamma globulin preparation. The majority of the rats inoculated with virus in the thymectomized and intact group had an abnormal IgG line when compared to the normal pattern and a deficiency in the IgA line. The abnormality in the IgG line consisted of a 3-11 mm reduction in the length of the line when measured using ruled graph paper. Each test rat serum was evaluated against the normal rat serum pattern developed on the same slide (Table I). Some of the virus-inoculated intact rats (6/15) also had a deficiency in the fast moving α_1 globulins. Instead of 2 well-defined lines in this region, some had only one ill-defined line. This same deficiency was seen in 1/10 normal rats and 1/14 thymectomized rats. Since it was observed previously (12) that the serum protein patterns of immature rats are not fully developed in regard to the IgG and IgA globulins, only the patterns of those rats living longer than 3 months are charted in Table I.

Weight measurements. The weights of both males and females in the 3 groups of rats were approximately the same at 3 weeks of age, the virus-inoculated groups (thymectomized 6 males, 8 females and intact 9 males,

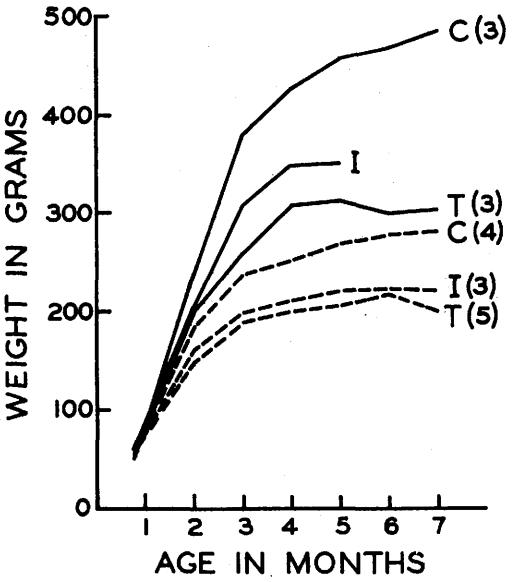

FIG. 1. Correlation of average weight gain with age in the thymectomized virus-inoculated group (T), intact virus-inoculated group (I) and control uninoculated group (C). Broken lines indicate females and solid line, males. Numbers in parentheses at end of each line indicate number of rats surviving at 7 months.

13 females) weighing slightly more on the average than the control uninoculated group (5 males, 5 females). Thereafter, the control uninoculated group gained weight more rapidly and reached a higher level than the other two groups, Fig. 1. Each point on the figure is the average weight of the animals surviving in that age group.

Discussion. In this study thymectomy within the first 24 hours of life reduced the incidence of leukemia from 95% to 25% in rats injected with Moloney virus at 48 hours after birth. This finding is in agreement with that of Kunii *et al*(8) where in one experiment the incidence of leukemia was reduced from 74% to 13% and in another from 93% to 19% in rats thymectomized 1-3 days after birth and then injected with Gross virus, passage A. The pronounced protection afforded by thymectomy in 1-3-day-old rats is not observed when thymus removal is delayed. Thus, in Gross' studies(7) where virus was injected into newborns and the thymus removed at 10 days, 93% of the thymectomized rats developed leukemia at an average age of 4 months, while in the control group 100% died of leukemia at an average age of 2.8 months. He interpreted these findings as indicating a slight delay in the development of leukemia when thymectomy was performed at 10 days of age. This age-thymectomy relationship was not observed in mice. In Gross' studies(4) mice were injected with Gross virus, passage A, when less than 9 days old and thymectomized at 1 month of age. Only 2% of the thymectomized group developed leukemia as compared to 87% in the control group. Similar results are reported by Miller(6). This disparity in results between rats and mice may be due to differences in the functional capacity of the thymus at the different ages in the two species of animals.

In the present study the thymectomized rats produced virus, but the proliferation of virus in the majority of these rats was in the absence of detectable transformation of lymphoid cells to malignancy. All newborn mice injected with plasma from the thymectomized rats died with symptoms of lymphoma over a period of 3 to 4 months, postinjection. The only deviation from this was the mice injected with plasma from a rat dying with reticulum cell sarcoma. These animals died over a period of 3 to 4 weeks and on microscopic examination had malignant lymphoma.

There was no significant difference in serum protein patterns between the thymectomized and intact virus-infected rats. Some of the rats in both groups showed deficiencies in the IgG and IgA classes of globulins as compared to normal control rats. The deficiency in the IgG class of globulins therefore appears to be a result, at least initially, of viral multiplication in lymphoid cells rather than due solely to transformation of such cells to malignancy. Thymectomized normal rats have been shown to have normal production of IgG and IgM globulins and a deficiency in IgA globulins(12). Thus, the deficiency in the IgA class of globulins appears to be correlated with loss of a functioning thymus, whether the loss be due to thymectomy(12) or to infection with Moloney virus.

Summary. Twenty-five per cent of a group of Osborne-Mendel rats thymectomized within 24 hours of birth and injected with Moloney virus at 48 hours of age developed neoplasms, 8% malignant lymphoma, 17% reticulum cell sarcoma, while 95% of the intact group developed malignant lymphoma over the period of the study. All of the thymectomized rats tested, however, were producing virus. Similar deviations from normal were found in the serum protein patterns of both thymectomized and intact virus-inoculated rats.

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Regulation of Erythropoiesis XIX. Effect of Hypoxia on Erythropoiesis in the Newborn Animal.* (30854)

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During late fetal life erythropoiesis in the rat is almost entirely hepatic and splenic. Erythropoiesis is first seen in the bone marrow at birth. In the next few days of life there is an explosive increase in red cell production so that by the 7th day of life, approximately 60% of the marrow elements are erythroid precursors(1). The peripheral blood at birth is characterized by the presence of hypochromic macrocytes which are short-lived. During the first 7-10 days of life, the cell production continues to be hypochromic and macrocytic in character; but the degree of macrocytosis is constantly decreasing so that there is a gradual shift in the indices toward those seen in adult rats. By the 40th to 60th days after birth, normocytic-normochromic cells are present in the peripheral blood and macrocytes are no longer seen.

The hepatic and early myeloid phase of erythropoiesis in the rat is characterized by the presence of large cells(1) frequently appearing in syncytial network with a leptochromatic nucleus with a diameter of about 15-20 μ; the nucleus stains a light pink and has a rather spongy appearance. The cytoplasm is slightly basophilic with fine vacuolization. When the cell membranes are discrete the cell diameter measures about 30-40 μ. This type of cell comprises about 5-10% of the nucleated elements of the bone marrow at birth. During the first 10-20 days of neonatal life, the frequency of this cell begins to decrease and after the 30th to 40th day

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