INDUCTION OF THE LUPUS ERYTHEMATOSUS ("L.E.") CELL IN VITRO IN PERIPHERAL BLOOD*

ROBERT N. HAMBURGER

In 1948, Hargraves, Richmond, and Morton[2] described a new type of cell observed in bone marrow preparations of patients with acute disseminated lupus erythematosus. This cell appears to be a mature polymorphonuclear leukocyte which has within its cytoplasm a large homogeneous mass of blue staining material which displaces the nucleus to the periphery of the cell. Since the cell has been observed only in patients with acute lupus erythematosus, these investigators have called it the "L.E." cell. In these same preparations they described other cellular forms which they thought might be precursors in the formation of the L.E. cell. In a subsequent article[1] the same investigators described the production *in vitro* of the L.E. cell phenomenon by incubating normal bone marrow with the plasma from patients with acute disseminated lupus erythematosus. Haserick and Bortz[3] confirmed the *in vitro* production of the L.E. cell and suggested that this be used as a diagnostic test. Following the report by Sundberg and Lick[4] that they had observed the L.E. cell in the peripheral blood of patients with acute lupus erythematosus, Hargraves[1] also noted the presence of these cells in peripheral blood preparations in very small numbers. However, it was necessary to centrifuge the heparinized or citrated venous blood in a specially designed flask in order to have an adequate "buffy coat" from which to make the smear.

The present investigation was carried out in an attempt to produce the L.E. cell in the peripheral blood by incubating the cellular elements of normal whole blood with plasma from patients with acute disseminated lupus erythematosus. The leukopenia usually seen in acute lupus erythematosus presents a problem because of the small number of leukocytes available for examination during the search for the L.E. cell. By the method of using the "buffy coat" from the peripheral blood of the normal subject which had been incubated with plasma from a case of acute disseminated lupus erythematosus the probability of finding the L.E. cell seemed more favorable.

---

* From the Department of Internal Medicine.
Received for publication February 10, 1950.

Method

Plasma from a case of acute disseminated lupus erythematosus was incubated with cells from normal subjects and the "buffy coat" was examined for L.E. cells. Control studies consisted of the incubation of normal cells with normal plasma in place of the plasma from the patient with lupus. A detailed description of the method follows.

Five cc. of venous blood from a patient with acute disseminated lupus erythematosus were collected in bottles containing balanced oxalate crystals (4 mg. potassium oxalate plus 6 mg. ammonium oxalate). The whole blood was centrifuged for ten minutes at 2000 RPM, and the plasma was pipetted off and saved.

Five cc. of venous blood from each of five normal subjects were collected in a similar way and each five-cc. specimen was treated in the following manner. One cc. of blood was placed in a Wintrobe hematocrit tube. Two cc. of the blood were placed in each of two five-cc. test tubes, and these two tubes were centrifuged for five minutes at 2000 RPM. The plasma was carefully pipetted off from each test tube and saved. To one test tube was added one cc. of the plasma previously prepared from the blood of the patient with acute lupus erythematosus. To the other test tube, which was used as a control, was added one cc. of plasma from another normal subject. These two test tubes were agitated gently to mix thoroughly the cellular elements and plasma and then incubated at a temperature ranging from 34° C. to 37° C. for twenty-five minutes. The tubes were agitated once during incubation and again at the end of twenty-five minutes. After incubation two Wintrobe hematocrit tubes were filled with the cells and plasma preparations. These two hematocrit tubes, along with the normal subject's control hematocrit tube, were centrifuged for ten minutes at 2000 RPM. The plasma was carefully pipetted off from each and discarded, and a single drop of the "buffy coat" was placed on a clean glass slide. A thin smear was made in the usual manner and stained with Wright's stain. Each slide was searched for thirty minutes, using an oil immersion lens.

For each of the five normal subjects in this series three slides were prepared: slide No. 1 contained the untreated "buffy coat"; slide No. 2 contained the "buffy coat" which had been incubated with the plasma from the patient with acute disseminated lupus erythematosus; and slide No. 3 contained the "buffy coat" which had been exposed to the plasma from another normal subject.

In addition to the plasma from the case of lupus erythematosus, plasma from three other patients was incubated with the normal cellular mixture and smears made as described above. Clinically these three patients had acute rheumatic fever, rheumatoid arthritis, and an acute sensitivity reaction to penicillin, respectively.

Results

In the untreated "buffy coat" smear from a patient with acute lupus erythematosus, L.E. cells were consistently observed in small numbers. In every slide prepared with normal "buffy coat" treated with plasma from

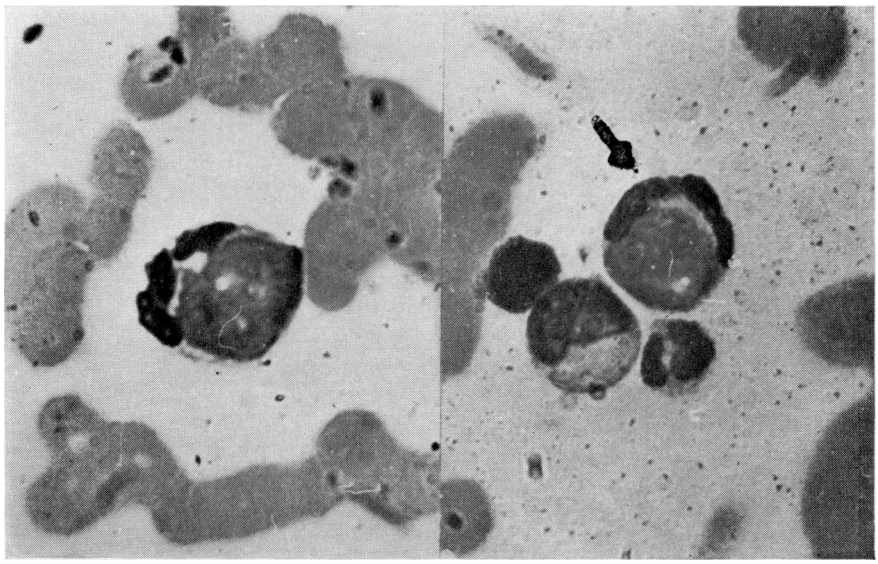

FIG. 1. Photomicrographs of L.E. cells from slides prepared with normal "buffy coat" treated with plasma from patient with acute disseminated lupus erythematosus.

this same patient with acute disseminated lupus erythematosus one or more L.E. cells were seen (Fig. 1). No L.E. cells were demonstrable in any of the control slides.

Discussion

The presence of the L.E. cell in the peripheral blood of patients with disseminated lupus erythematosus has been confirmed in these studies. In addition, these observations demonstrate that the polymorphonuclear leukocyte in the peripheral blood of a normal individual will assume the characteristic appearance of the L.E. cell after incubation with plasma from a patient with disseminated lupus erythematosus. The mechanism of the production of the L.E. cell is obscure. It may be that the polymorphonuclear leukocyte has been acted upon by the lupus plasma to cause it to phagocytize cellular debris. Or possibly the plasma from the patient with lupus erythematosus causes nucleolysis of certain of the normal cellular elements, which, in turn, are then phagocytized by polymorphonuclear leukocytes.

Hargraves[1] noted the difficulty of isolating the L.E. cell in the peripheral blood of patients with acute disseminated lupus erythematosus because of the paucity of leukocytes concomitant with this illness. The use of normal peripheral blood containing adequate numbers of leukocytes treated with plasma from a patient with lupus erythematosus would alleviate this aspect of the problem. This procedure technically simplifies to a great extent the induction of the L.E. cell for diagnostic purposes in suspected cases of acute disseminated lupus erythematosus.

Summary

1. The L.E. cell has been found in the peripheral blood of patients with acute disseminated lupus erythematosus.

2. The L.E. cell can be induced by cross incubation of cellular elements from normal peripheral blood with the plasma from a patient with lupus erythematosus.

3. The induction of the L.E. cell using the method described in this study simplifies the diagnostic test for acute disseminated lupus erythematosus.

The author wishes to thank Dr. F. G. Blake and Dr. F. Douglas Lawrason for their help and suggestions in the preparation of the manuscript.

References

1 Hargraves, M. M.: Production in vitro of the L.E. cell phenomena; use of normal bone marrow elements and blood plasma from patients with acute disseminated lupus erythematosus. Proc. Mayo Clin., 1949, *24*, 234.

2 Hargraves, M. M., Richmond, H., and Morton, R. J.: Presentation of two bone marrow elements, "Tart" cell and "L.E." cell. Proc. Mayo Clin., 1948, *23,* 25.

3 Haserick, J. R. and Bortz, D. W.: A new diagnostic test for acute disseminated lupus erythematosus. Cleveland Clin. Q., 1949, *16,* 158.

4 Sundberg, R. D. and Lick, N. B.: "L.E." cells in the blood in acute disseminated lupus erythematosus. J. Invest. Derm., 1949, *12,* 83.