assess. However, differences did appear depending on the length of time before immersing in the TTC solution after thawing (Table 2). The more badly damaged tissue (−13° C) can be seen to show a nearly negative test sooner than the less damaged tissue (−10°).

In seed testing TTC has been widely and successfully used. Evaluating the intensity and extent of staining so as to predict the germinability of a sampled lot of seed is the main difficulty. Sometimes the test seems to overrate the germinability percentage in certain conifer seed which have a period of dormancy that must be overcome for relatively rapid germination. This discrepancy may be partly the fault of the germination tests which are carried out with samples from the same lot of seed (*3*). We obtained the best predictions by making the TTC test on a sample of seed as they arrived from the seed house before stratification, and comparing these with percentages of germinating seed after stratification in moist cloth at 40° F for 2 months. Evidently there is not only a decrease in time of reacitvity of the seed to TTC during stratification, but there is an increase in the total percentage of seed showing a positive TTC test (Table 3).

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The Influence of a Plasma Factor on *in vitro* Leucocyte Migration[1]

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While the intrinsic mobility of leucocytes is a well-known phenomenon, the influence of the plasma on this mobility has not yet been made clear. Using an *in vitro* method we have been able to show that the rate of migration of leucocytes is strongly influenced by a factor in the plasma.

In this technique, heparinized venous blood is first divided into cells and plasma. The cells are then washed three times by a sequence of suspension and centrifugation in Hanks' solution (*1*), and finally resuspended in autologous or homologous plasma. This suspension is then drawn into thin-walled capillary tubes of the type commonly used for melting point determinations. Each tube is sealed and then centrifuged to subdivide the system into three components; namely, a basal layer of packed red cells, an intermediate buffy coat of leucocytes, and an uppermost layer of plasma (Fig. 1). The initial concentration of heparin is adjusted so that the plasma clots after centrifugation, thus providing a matrix into which the leucocytes can migrate. Tubes are incubated in an upright position for 16 hr at 37° C, and the distance which the leucocytes have migrated into the plasma is then measured with an ocular micrometer. This measurement is therefore an indication of the distance which the fastest-moving leucocytes have traveled from the buffy coat. For each combination of cells and plasma, ten tubes were measured, and an average

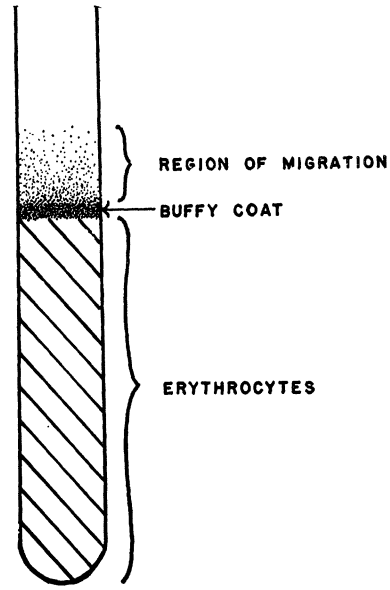

FIG. 1. Diagrammatic representation of the capillary tube method of measuring leucocyte migration.

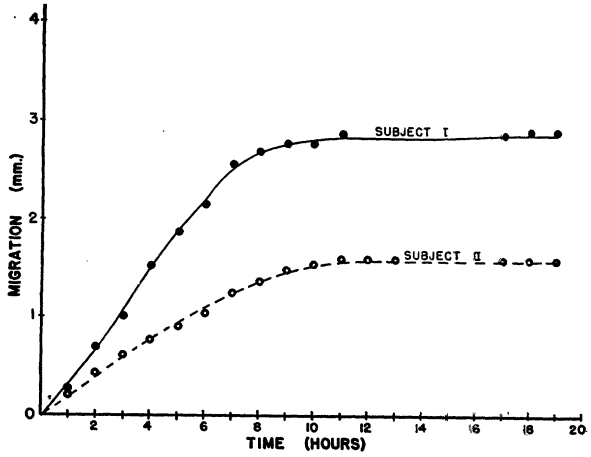

FIG. 2. Rate of leucocyte migration in two subjects with different migration distances. The difference in migration distance is the result of a difference in the rate of migration during the early hours of the experiment.

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value taken. The migrations are not free movements into a liquid phase. If a clot is present, the cells move along the fibrin micellae. But if no clot is present, the cells will migrate along the walls of the capillaries, and the results are very similar.

For the experiments, it was most convenient to make a single measurement after 16 hr. However, as shown in Fig. 2, rate studies indicate that when the migration distance between two subjects varies, this is a result of the difference in the rate of migration during the early hours of the experiment.

The normal subjects used for these experiments were laboratory personnel. At the time of the experiment each subject was asymptomatic. It was discovered that symptoms from slight infections were accompanied by a marked effect on the leucocyte migration.

TABLE 1

AVERAGE MIGRATION DISTANCE OF LEUCOCYTES FROM NORMAL SUBJECTS

Subject	Sex	Blood type	No. expt	Mean (mm)	Standard deviation (mm)
KE	F	A, Rh +	5	0.47	0.05
JH	F	A, Rh +	6	0.56	0.04
MK	M	A, Rh +	18	0.71	0.10
JL	M	A, Rh +	13	2.16	0.38
EAS	F	O, Rh +	5	2.47	0.61
CBF	M	O, Rh +	5	2.64	0.26

Table 1 records the data on experiments designed to evaluate the technique. The cells of normal persons were here suspended in the individual's own plasma. Though the cell migration of a given individual was relatively constant on different days, it is clear that a great deal of variation exists between individuals.

TABLE 2

AVERAGE MIGRATION DISTANCE OF LEUCOCYTES IN AUTOLOGOUS AND HOMOLOGOUS PLASMA

Plasma	MK cells (mm)	JL cells (mm)
MK	0.54	0.78
JL	1.63	1.93

Table 2 summarizes the results of six experiments designed to ascertain the importance of cells and plasma in the migration of the leucocytes. Two normal males having widely different average migrations were chosen. Both had type A, Rh positive blood. The migration of the cells of each individual was measured both in his own plasma and in the plasma of the other individual. MK cells in MK plasma migrated an average distance of 0.54 mm, and JL cells in JL plasma migrated an average distance of 1.93 mm. However, when MK cells were tested in JL plasma, their migration increased to 1.63 mm, and when JL cells were tested in MK plasma, their migration decreased to 0.78 mm. While there is still a difference in the migratory ability of the cells of the two subjects, it is evident that the plasma dictates to a large degree the rate of cell migration.

Similar experiments were repeated on types A, O, and B individuals. The results were consistent with the findings on the two initial subjects. In another series of experiments, the blood groups were mixed. Under such circumstances, if migration was not prevented by agglutination of the erythrocytes during the preparation of capillary tubes, the results were also consistent with those shown in Table 2.

The substance or substances contained in the plasma which condition the migration of leucocytes are heatlabile and nondialyzable. At present we are attempting to characterize the substances further by fractionation of the plasma proteins according to the small volume cold alcohol technique (*2*). Preliminary data indicate that when Fraction 3 of a slow plasma is added to a fast plasma, the migration of leucocytes in that plasma is markedly suppressed. And, when Fraction 2 of a fast plasma is added to a slow plasma, the migration of leucocytes in that plasma is increased.

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Hepatic Tumors Due to Prolonged Ethionine Feeding[1]

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Ethionine (α-amino-γ-ethylthiobutyric acid) is an analogue of methionine and one of its biologic antagonists (*1, 2*). It produces, within a few days, fatty metamorphosis of the liver in female but not in male rats (*3, 4*). The lesion was considered the result of interference with amino acid or protein metabolism (*5*). Feeding synthetic diets containing 0.5% ethionine to male and female rats for 4 weeks produces a reduction of the fat content and severe liver cell damage (preceded by diffuse fatty metamorphosis and central necrosis). The liver shows diffuse infiltration by inflammatory cells as well as intralobular fibrosis and bile duct proliferation (*6*). Large liver cells with huge, sometimes multiple, nuclei and very large eosinophilic nucleoli are noted as well as irregularity in the appearance of the bile ducts (*7*). These pictures, interpreted as irregular regeneration, raised the question whether tumor formation may eventually result

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