

## MIGRATION OF PERIPHERAL BLOOD LYMPHOCYTE POPULATIONS AND SUBPOPULATIONS

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Spontaneous migration of peripheral blood leukocytes is an active and dynamic process that is modified by various factors both *in vitro* and *in vivo*. Changes in migration mobility of leukocytes is found in various diseases: rheumatic fever, primary immunodeficiency states, surgical diseases, skin diseases, and tumors [1, 2, 5, 8].

However, the leukocytes are a group of cells with heterogeneous composition, and it therefore was considered useful to make a more detailed study of the migrating power of both polynuclear and mononuclear cells together constituting this large cell population.

The object of the present investigation was to study spontaneous migration of B and T lymphocytes as well as of subpopulations of T lymphocytes in healthy human peripheral blood.

### EXPERIMENTAL METHOD

Heparinized blood from 15 healthy subjects was used. The mobility of the neutrophils and lymphocytes, isolated in a Ficoll-Isopaque density gradient [3], and also of peripheral blood leukocytes was estimated by the direct capillary test [12]. The results were read after culture for 24 h at 37°C and expressed as the migration index (MI), in per cent; the migration index of leukocytes was taken as 100%:

$$MI = \frac{\text{migration of lymphocytes (or neutrophils)}}{\text{migration of leukocytes}} \cdot 100 \%$$

Adherent cells were removed from the suspension of lymphocytes by Bryant's method [4]. To determine the numbers of T and B cells in the migration zone and in the capillary tube, methods of determination of E-RFC and EAC-RFC were used [6]. The subpopulations of T lymphocytes ( $T_\gamma$  and  $T_\mu$ ) were identified by Moretta's method [9]. The results were subjected to statistical analysis by Student's and Fisher's tests.

### EXPERIMENTAL RESULTS

Analysis of the results (Table 1) shows that peripheral blood lymphocytes have marked migrating power. There is an extensive literature on the study of spontaneous lymphocyte migration *in vivo*, but reports indicating that this type of cell migrates spontaneously *in vitro* also have only recently begun to appear [10]. We found that peripheral blood lymphocytes migrate more actively than neutrophils ( $P < 0.05$ ). Removal of adherent cells from the lymphocyte suspension led to a marked ( $P < 0.01$ ) increase in migration mobility of the remaining lymphocytes. Addition of phytohemagglutinin in a dose of 2  $\mu\text{g/ml}$  caused no significant inhibition of lymphocyte migration ( $P > 0.05$ ), whereas the case of the same dose of mitogen during culture of the total peripheral blood leukocyte population led to marked inhibition ( $MI = 62.6 \pm 5.7\%$ ).

Experiments with enriched suspensions of T and B lymphocytes [7, 13] showed that T cells have stronger migrating ability than B cells. Having used a simplified and physiological method, we also found that in a whole suspension of lymphocytes the number of E-RFC, i.e., the number of T lymphocytes in the migration zone, was 50%, whereas the number of EAC-RFC, i.e., of B lymphocytes, was only 5%. Meanwhile, among the nonmigrating cells, there were 24% of E-RFC and 14% of EAC-RFC. The relative number of T cells forming "active" rosettes ( $E_a$ -RFC) was unchanged during migration and amounted to 76-78% of the total number of E-RFC (Table 2).

We know from the literature that T helpers ( $T_\mu$ ) exhibit greater chemotaxic mobility than T suppressor ( $T_\nu$ ) [11]. The possibility cannot be ruled out that this is not only due to the special features of their response of the chemotaxic factor, but is also directly connected with spontaneous migration of these cells. We showed that  $T_\gamma$  for practical purposes do

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TABLE 1. Spontaneous Migration of Peripheral Blood Leukocytes, Lymphocytes, and Neutrophils (n = 15)

Type of cell	Migration index (M ± m), %
Leukocytes	100,0 ± 3,2
Neutrophils	71,1 ± 6,5*
Lymphocytes	91,4 ± 3,1

\*Difference significant (P < 0.05) compared with migration of leukocytes.

TABLE 2. Spontaneous Migration of Peripheral Blood Lymphocyte Populations and Subpopulations (M ± m)

Type of cells	Number of cells, %	
	among migrating cells	among non-migrating cells
E-RFC	49,8 ± 5,2	25,7 ± 3,1
EAC-RFC	4,7 ± 1,0	15,7 ± 1,1
E <sub>a</sub> -RFC	31,0 ± 1,8	14,0 ± 1,0
T <sub>v</sub> -RFC	0,5 ± 0,1	7,5 ± 0,6
T <sub>μ</sub> -RFC	18,7 ± 3,3	26,3 ± 3,1

not migrate, whereas T<sub>μ</sub> are actively migrating cells.

Peripheral blood lymphocytes thus possess marked and varied migrating activity *in vitro* depending on the population and subpopulation of cells to which they belong and also on interaction between various factors. The study of lymphocyte migration and its possible disturbances may be of fundamental importance in the elucidation of the mechanisms of the pathogenesis of diseases of the immune system.

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