

Morphological and Functional Peculiarities of Lymphoid Cells in Mice of Different Strains

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We revealed differences in quantitative composition and functional activity of lymphoid cells in intact mice of different strains. Cellularity and counts of lymphoid elements in hemopoietic and lymphoid organs, proliferative activity of T and B lymphocytes, and counts of CD4⁺ and CD8⁺ lymphocytes in the spleen were minimum in CC57W and Balb/c mice and maximum in CBA/CaLac and DBA/2 mice. The highest absolute content of lymphoid elements in the spleen was detected in Balb/c mice, while CC57W mice had the highest content of these elements in the thymus.

Key Words: mice of different strains; lymphoid cells

The use of genetically identical animals in experiments provides high reproducibility of results and improves the efficiency and reliability of biological studies. Selection and inbreeding of laboratory animals led to fixation of some rare signs and permitted the use of inbred mouse strains as biological models [3].

The reactions of the hemopoietic and immune systems as indicators of homeostasis in animals with different genotypes attract special interest. The detected differences in immune reactions of different mouse strains [4] suggest quantitative and functional differences in the basal pool of immunocompetent cells. We studied the morphology and functions of lymphoid cells in mice of different strains.

MATERIALS AND METHODS

Experiments were carried out on CBA/CaLac ($n=20$), C57Bl/6 ($n=20$), DBA/2 ($n=20$), Balb/c ($n=20$), CC57W ($n=20$), and C3H ($n=10$) mice (body weight 18 g). The animals (conventional mouse strains) were obtained from the Laboratory for Experimental Biomedical Simulation, Institute of Pharmacology.

Standard hematological methods for evaluation of the total cellularity of the bone marrow, peripheral blood, thymus, and spleen. Cytograms were expressed in absolute values [1].

The count of T lymphocytes [7], theophylline-resistant cells (T helpers), B lymphocytes [2] in the bone marrow and spleen was determined out by rosette formation; lymphocyte subpopulations (CD4⁺, CD8⁺ cells) in the spleen were evaluated by indirect immunofluorescence [5] using monoclonal antibodies (Caltag Laboratories) in accordance with manufacturer's instructions. Proliferative activity of T and B cells was studied in the lymphocyte blastogenesis test using XTT tetrazolium (2,3-bis[2-methoxy-4-nitro-5-sulfophenyl] 2H-tetrasolium-5-carboxanilide inner salt) [6]. The results were evaluated on AIFR-01 Uniplan enzyme immunoassay analyzer at 450-500 nm. Enzyme immunoassay for measurements of IL-2 and IL-10 was carried out in supernatants of ConA-stimulated (ICN Biomedicals Inc.) splenocytes using Amersham Pharmacia Biotech kits according to the manufacturer's instructions.

The data were processed using Statistica software.

RESULTS

Studies of the bone marrow hemopoiesis in mice of different strains showed that the content of karyocytes

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TABLE 1. Total Cellularity and Absolute Counts of Lymphoid Elements in the Bone Marrow, Spleen, Thymus ($10^6/\text{Organ}$), and Peripheral Blood ($10^9/\text{liter}$) in Mice of Different Strains ($X \pm m$)

Parameters for organs	DBA/2	C57Bl/6	Balb/c	CC57W	CBA/CaLac
Bone marrow					
total karyocyte count	18.90 \pm 0.61	18.75 \pm 0.59	16.16 \pm 1.60	17.41 \pm 0.36	19.2 \pm 0.57
lymphoid cells	7.31 \pm 0.87	6.49 \pm 0.25	5.39 \pm 0.58	4.86 \pm 0.27	6.43 \pm 0.37
Peripheral blood					
total leukocyte count	12.65 \pm 0.89	8.20 \pm 0.77	8.95 \pm 0.35	5.75 \pm 0.31	10.78 \pm 0.73
lymphocytes	7.61 \pm 0.40	6.42 \pm 0.70	5.98 \pm 0.30	4.55 \pm 0.21	6.41 \pm 0.71
Spleen					
total splenocyte count	100.80 \pm 3.43	143.40 \pm 6.56	172.61 \pm 25.63	96.80 \pm 1.28	116.00 \pm 4.19
lymphoid cells	72.48 \pm 3.01	112.89 \pm 4.04	139.54 \pm 20.87	76.68 \pm 1.35	104.20 \pm 4.06
Thymus					
total thymocyte count	54.00 \pm 6.16	106.40 \pm 5.30	36.81 \pm 6.05	106.20 \pm 2.22	90.58 \pm 4.91
lymphoblasts	2.86 \pm 0.43	3.42 \pm 0.27	2.11 \pm 0.58	4.78 \pm 0.74	2.16 \pm 0.29
medium lymphocytes	7.11 \pm 0.92	11.07 \pm 0.72	3.31 \pm 0.83	11.33 \pm 1.50	12.77 \pm 1.04
minor lymphocytes	40.53 \pm 4.60	85.79 \pm 4.49	29.74 \pm 4.30	81.87 \pm 3.45	73.69 \pm 4.02

was minimum in Balb/c mice and maximum in CBA/CaLac mice. The absolute count of lymphoid elements was minimum in CC57W and maximum in DBA/2 mice (Table 1). Studies of the peripheral blood characteristics revealed minimum leukocyte count in CC57W and maximum leukocyte count in DBA/2 mice. The same trend was observed for the absolute count of lymphocytes in the peripheral blood: the lowest level in CC57W and the highest level in DBA/2 mice (Table 1). The minimum splenocyte count was detected in CC57W and the maximum in Balb/c mice. Consequently, the content of lymphoid cells in the spleens was low in CC57W and DBA/2 mice and high in Balb/c mice. The study of the thymus showed the minimum total count of thymocytes in Balb/c mice, which was paralleled by the lowest counts of lymphoblasts, medium and minor lymphocytes in this mouse strain. CC57W and C57Bl/6 mice had the highest counts of cells in the thymus, CC57W mice having the maximum number of lymphoblasts in the thymus, CBA/CaLac mice the greatest number of medium lymphocytes, and C57Bl/6 mice the highest number of minor lymphocytes (Table 1).

Studies of the quantitative parameters of lymphocyte subpopulations in the bone marrow and spleen of C57Bl/6 and DBA/2 mice showed that the counts of B and total T lymphocytes in these organs was higher in C57Bl/6 mice. The count of T helpers in the bone marrow was higher in DBA/2 mice (Table 2).

The content of CD4⁺ cells in the spleens varied from 27.33% (Balb/c mice) to 37.67% (C3H) and the count of CD8⁺ cells from 15% (Balb/c) to 39.67% (C3H; Table 3). The CD4⁺/CD8⁺ ratio was minimum in C3H and maximum in Balb/c mice (Table 3).

The study of proliferative activity of T and B lymphocytes in the spleen showed the highest index of T lymphocyte stimulation in C57Bl/6 mice and the lowest in Balb/c and CC57W mice. The index of B lymphocyte stimulation varied from 1.06 (Balb/c) to 1.57 (CBA/CaLac; Table 3).

Production of IL-2 and IL-10 by splenic T lymphocytes was more intensive in CBA/CaLac mice (790.51 \pm 3.31 and 273.57 \pm 36.11 pg/ml, respectively) compared to C57Bl/6 mice (679.16 \pm 30.77 and 141.60 \pm 12.53 pg/ml, respectively).

Hence, differences in quantitative composition and functional activity of lymphoid cells in intact mice of different strains were revealed. The total cellularity and the number of lymphoid elements in the hemopoietic and lymphoid organs, proliferative activities of T and B lymphocytes, counts of CD4⁺ and CD8⁺ lymphocytes in the spleen were minimum in CC57W and

TABLE 2. Absolute Counts ($10^6/\text{Organ}$) of B, Total T Lymphocytes, and T Helpers in the Bone Marrow and Spleen of Mice of Different Strains ($X \pm m$)

Parameter	C57Bl/6	DBA/2
Bone marrow		
B lymphocytes	4.75 \pm 0.43	2.13 \pm 0.26
total T lymphocytes	8.48 \pm 0.69	7.96 \pm 1.30
T helpers	0	1.69 \pm 0.33
Spleen		
B lymphocytes	36.37 \pm 3.43	33.21 \pm 1.82
total T lymphocytes	64.92 \pm 5.88	59.33 \pm 3.52
T helpers	0	0

TABLE 3. Counts of CD4⁺ and CD8⁺ Lymphocytes, Their Ratio, and Index of Stimulation of Proliferative Activity of Splenic T and B Lymphocytes in Mice of Different Strains ($\bar{X} \pm m$)

Mouse strains	Percentage		CD4 ⁺ /CD8 ⁺ , %	Stimulation index	
	CD4 ⁺ lymphocytes	CD8 ⁺ lymphocytes		T lymphocytes	B lymphocytes
CBA/CaLac	31.00±1.29	18.00±2.31	1.71±0.19	1.46±0.13	1.57±0.24
C57Bl/6	34.80±1.80	28.25±3.09	1.27±0.19	1.74±0.02	1.53±0.06
DBA/2	33.00±1.00	26.00±0.58	1.05±0.08	0.96±0.07	1.52±0.12
Balb/c	27.33±1.76	15.00±0.58	2.21±0.21	1.07±0.06	1.06±0.04
CC57W	28.33±0.88	16.67±2.03	1.83±0.48	1.07±0.01	1.10±0.06
C3H	37.67±1.33	39.67±2.33	0.96±0.09	1.10±0.01	1.13±0.03

Balb/c mice. The maximum values were observed in CBA/CaLac and DBA/2 mice. Balb/c mice had the highest absolute count of lymphoid elements in the spleen and CC57W mice in the thymus.

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