Immunity Parameters in Mice of Different Strains

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Quantitative composition and functional activity of immunocompetent cells differ in mice of different strains. The counts of T cells in the bone marrow and spleen, proliferative activity of T cells in the spleen, levels of IL-2 and IL-10 production by splenic T cells, number of antigen-specific T cells and their functional activity are low in C57Bl/6, BALB/c, and CC57W mice and high in CBA/CaLac, DBA/2, and C3H animals. Low phagocytic activity of peritoneal macrophages was detected in BALB/c and CC57W mice and high activity in C3H animals. The content of antibody-producing cells in the spleens of C57Bl/6, BALB/c, and CC57W mice is higher than in CBA/CaLac, DBA/2, C3H, A/SN, and AKR/JY mice. Functional activity of B cells is lower in BALB/c and CC57W compared to CBA/CaLac and DBA/2 mice.

Key Words: *mice of different strains; nonspecific resistance parameters; cellular and humoral immunity parameters*

Individual sensitivity of animals to similar exposure is one of the main problems of biomedical research. The use of genetically homogenous animals in experimental studies provides the identity of data and improves the efficiency and reliability of biological findings. Inbreeding fixed rare and valuable signs in laboratory animals, which allowed the use of inbred mouse strains as experimental models in medicine and biology [1]. One of the main indicators of homeostasis is the immune system [3,9,10]. Differences in the parameters of immune reactions in mice of different strains [1,4] suggest quantitative and functional differences in the pool of immunocompetent cells.

We studied some immunity values in intact 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), C3H (n=10), A/SN (n=10), and AKR/JY (n=10) mice weighing 18 g (collection fund of Laboratory for Experimental Biomedical Simulation of Institute of Pharmacology).

Phagocytic activity of peritoneal macrophages was evaluated by measuring optical density of the solution after destruction of phagocytes loaded with neutral red particles [13]. The levels of total T cells [12], theophylline-resistant cells (T helpers) [2], and B cells [6] in the bone marrow were evaluated by the method of rosette formation. Lymphocyte subpopulations ($CD4^+$, $CD8^+$ cells) in the spleen were evaluated by indirect immunofluorescence [5] with monoclonal antibodies (Caltag Lab.). Proliferative activity of lymphoid (T and B) cells was evaluated in the lymphocyte blast transformation test using 2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-5-[(phenylamino)carbonyl]-2H-tetrazolium hydroxide (XTT, soluble tetrazolium salt) [11]. The results were evaluated on AIFR-01 enzyme immunoassay analyzer (Uniplan) at λ=450-500 nm.

Enzyme immunoassay for measuring IL-2 and IL-10 was carried out with supernatants of Con A-stimulated splenic lymphocytes (ICN Biomedicals Inc.) using Amersham Pharmacia Biotech kits. Cellular immune response was evaluated in the delayed-type hypersensitivity test [7]. For sensitization the animals were subcutaneously injected with 1×10^7 sheep erythrocytes (100 μl). The resolving dose of the antigen (1×10^8

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sheep erythrocytes, 20 μl) was injected on day 5 after sensitization under the hind paw aponeurosis. The same volume of normal saline was injected into the contralateral paw (control). The intensity of inflammatory reaction was evaluated 24 h after injection of the resolving dose of the antigen. The animals were sacrificed, both paws were cut at the level of the talocrural joint, and weighed on torsion scales. The inflammation index was evaluated by the difference in the weights of experimental and control paws. The absolute (10^6/organ) count of antibody-producing cells in the spleen was evaluated by the method of local hemolysis [8]. The data were processed by variation statistics method using Statistica for Windows software.

RESULTS

The phagocytic activity of peritoneal macrophages was minimum in CBA/CaLac, BALB/c, and CC57W mice and maximum in C3H mice (Table 1).

The total content of T cells and count of T helpers were higher in DBA/2 mice than in C57Bl/6 ones. The count of B cells was higher in the bone marrow of C57Bl/6 mice (Table 2).

The content of $CD4^+$ cells in the spleen was maximum in C3H mice (37.67%) and minimum in BALB/c mice (27.33%). The content of $CD8^+$ cells was higher in C3H mice and lower in BALB/c and CC57W mice. The $CD4^+/CD8^+$ ratio was minimum in C3H mice and maximum in BALB/c ones (Table 3).

The highest index of T and B cell stimulation was detected in C57Bl/6 (1.74±0.02 and 1.53±0.06) and the lowest in BALB/c (1.07±0.06 and 1.06±0.04) and CC57W mice (1.07±0.01 and 1.10±0.06, respectively).

Analysis of the capacity of splenic T cells of CBA/CaLac and C57Bl/6 mice to produce IL-2 and IL-10 revealed higher values in CBA/CaLac mice compared to C57Bl/6 mice. In CBA/CaLac mice the level of IL-2 production by splenocytes was 790.51±3.31 pg/ml, of IL-10 273.57±36.11 pg/ml, *vs.* 679.16±30.77 and 141.60±12.54 pg/ml, respectively, in C57Bl/6 mice.

Delayed-type hypersensitivity test showed that the production of antigen-specific T cells was higher in CBA/CaLac mice and functional activity of these cells (production of proinflammatory cytokines) was higher than in C57Bl/6 mice.

The content of antibody-producing cells in the spleens in CBA/CaLac, DBA/2, C3H, A/SN, and AKR/JY mice (from 2.80±0.97 to 4.40±0.45 million cells/organ) was lower than in C57Bl/6, BALB/c, and CC57W animals (from 9.55±0.88 to 10.12±1.80 million cells/organ).

Hence, the parameters of cellular immunity (T cell counts in the bone marrow and spleen, including counts of $CD4^+$ and $CD8^+$ cells), proliferative activity of T cells, levels of IL-2 and IL-10 production by splenic T cells, content of antigen-specific T cells and their functional activity) were lower in C57Bl/6, BALB/c, and CC57W mice than in CBA/CaLac, DBA/2, and C3H animals. Nonspecific resistance (phagocytic activity of peritoneal macrophages) was low in BALB/c and CC57W mice and maximum in C3H animals. Humoral immunity parameters (count of antibody-producing cells in the spleen) were high in C57Bl/6, BALB/c, and CC57W mice and low in CBA/CaLac, DBA/2, C3H, A/SN, and AKR/JY animals. Functional

TABLE 1. Phagocytic Activity of Peritoneal Macrophages in Mice of Different Strains (Arb. Units; $X \pm m$)

Mouse strain	Value
CBA/CaLac	32.70±6.29
C57Bl/6	92.60±14.78
DBA/2	84.00±10.61
BALB/c	33.00±5.94
CC57W	28.60±5.94
C3H	186.60±12.56
A/SN	51.60±7.44
AKR/JY	57.00±12.49

TABLE 2. Counts of Common T (Tc), B cells, and T Helpers (Th) in the Bone Marrow and Spleen of DBA/2 and C57Bl/6 Mice (%; $X \pm m$)

Parameter	DBA/2	C57Bl/6
Bone marrow		
Tc cells	54.40±8.55	45.00±2.51
Th cells	11.60±2.16	2.0±0.5
B cells	14.60±1.91	25.20±1.53
Spleen		
Tc cells	45.0±2.5	45.00±2.51
Th cells		4.5±0.5
B cells	25.20±1.54	25.20±1.53

TABLE 3. Content of $CD4^+$, $CD8^+$ Lymphocytes and Their Ratio in the Spleens of Mice of Different Strains (%; $X \pm m$)

Mouse strain	$CD4^+$ lymphocytes	$CD8^+$ lymphocytes	$CD4^+/CD8^+$
CBA/CaLac	31.00±1.29	18.00±2.31	1.71±0.19
C57Bl/6	34.80±1.80	28.25±3.09	1.27±0.19
DBA/2	33.00±1.00	26.00±0.58	1.05±0.08
BALB/c	27.33±1.76	15.00±0.58	2.21±0.21
CC57W	28.33±0.88	16.67±2.03	1.83±0.48
C3H	37.67±1.33	39.67±2.33	0.96±0.09

activity of B cells was lower in BALB/c and CC57W mice than in CBA/CaLac and DBA/2 animals.

REFERENCES

1. Z. K. Blandova, V. A. Dushkin, A. M. Malashenko, and E. F. Shmidt, *Laboratory Animal Strains for Biomedical Research* [in Russian], Moscow (1983).
2. E. D. Gol'dberg, A. M. Dygai, and V. P. Shakhov, *Tissue Culture Methods in Hematology* [in Russian], Tomsk (1992).
3. V. I. Konenkov, *Medical and Ecological Immunogenetics* [in Russian], Siberian Division of Russian Academy of Medical Sciences, Novosibirsk (1999).
4. N. V. Masnaya, A. A. Churin, O. S. Borsuk, and E. Yu. Sherstoboev, *Byull. Eksp. Biol. Med.*, **134**, No. 10, 437-439 (2002).
5. I. Lefkowits and B. Pernis, Eds., *Methods of Investigation in Immunology* [in Russian], Moscow (1981).
6. E. U. Paster, V. V. Ovod, V. K. Pozur, and N. E. Vikhot', *Immunology: Practicum* [in Russian], Kiev (1989).
7. R. V. Khaitov, I. S. Gushchin, B. V. Pinegin, and A. I. Zebrev, *Vedomosti Farmakologicheskogo Komiteta*, No. 1, 34 (1999).
8. A. J. Cunningham, *Nature*, **207**, 1106 (1965).
9. Y. Ichiki, M. Tekenoyama, M. Mizukami, *et al.*, *J. Immunol.*, **8**, No. 172, 4844-4850 (2004).
10. J. S. Isenberg, *Ann. Plast. Surg.*, **5**, No. 52, 523-530 (2004).
11. D. A. Scudiero, R. H. Shoemaker, K. D. Paull, *et al.*, *Cancer Res.*, **48**, 4827-4833 (1988).
12. M. Jondal, G. Holm, and H. J. Wigrell, *Exp. Med.*, **136**, 207-222 (1972).
13. B. Vrav, J. Hoebeke, M. Saint-Guillain, *et al.*, *Scand. J. Immunol.*, **11**, 147-153 (1980).