http://dx.doi.org/10.35630/2199-885X/2020/10/2.6

INTERRELATION OF TRACE ELEMENTS AND THE STRUCTURAL ORGANIZATION OF LYMPH NODES AT YOUNG AND SENILE AGE

Received 02 April 2020;
Received in revised form 19 May 2020;
Accepted 29 May 2020

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ABSTRACT — The article deals with the study of interrelation of trace elements and morphology of lymph nodes at different localization and age-related changes. Lymph nodes of different localization at young and old animals were analyzed by a morphological method with definition of trace elements by roentgen fluorescent method and the use of synchrotron radiation. There are data on forming of the lymphoid-microelement association important for integrative assessment of the structural organization of lymph nodes. Structural modification of lymph nodes has certain patterns, proceeding from features of a microelement profile. The region-dependent morphological variant of lymph nodes corresponds to a certain content of trace elements and their correlation with compartments of lymph nodes. Trace elements deficiency and reduction of compartments of lymph nodes are considered as predictors of aging of lymphoid tissue. The results have practical significance to justify a higher trace-element content in anti-aging programs.

KEYWORDS — lymph nodes, trace elements, gerontology.

INTRODUCTION

Lymph nodes are peripheral organs of immune and lymphatic system representing difficult organized structures reacting to different exo- and endogenous factors [1, 2]. Lymph nodes functions together with trace elements. Many trace elements play an important role in maintenance of the optimum immune response, showing regulatory, structural and stabilizing functions [3, 4]. Trace elements take an active part in cofactors or catalysts of enzymes of free radical oxidation in cells of the immune system [4, 5]. Activity of enzymes and the immune system are closely related with change of a trace element homeostasis and, finally, with change of the structure of peripheral lymphoid organs [1, 3]. Aging is connected with an immunogenesis problem that is defined by features of the structural organization of peripheral lymphoid organs according to the concept of the lymphatic region [1]. It does relevant studying morphological equivalents of the immune response of lymph nodes depending on a microelement profile and age.

The aim of the study is assessment the structural organization and trace elements of lymph nodes at young and senile age.

MATERIALS AND METHODS

The experiment was conducted on white Wistar rats with the natural rate of aging at the age of the 3rd month (young animals) and 1.5 years (old animals). Mesenteric, inguinal and tracheobronchial lymph nodes were analyzed by a morphological method.

Lymph nodes were fixed in 10% neutral formalin. After fixing we resorted to the classical scheme of washing, dehydration, imbibition with a xylol, paraffin and preparation histologic sections on the microtome. Histologic sections of lymph nodes painted hematoxylin and eosine, azure-II-eosine, trichromatic paint on Masson. The cross-section area of lymph nodes was standardized and taken for 100% that allowed to estimate and compare the structural organization of lymph nodes.

The content of trace elements (Se, Mn, Fe, Cu, Zn) in lymph nodes was defined by the roentgenfluorescent method with use of synchrotron radiation. The work was done at the shared research center SSTRC on the basis of the Novosibirsk FEL/VEPP-4 – VEPP-2000 complex at BINP SB RAS, using equipment supported by project RFMEFI62119X0022 [6].

The morphometric analysis of structures of a lymph node was carried out by means of a morphometric grid and the Image-Pro Plus 4.1 program. Statistical data processing was performed with licensed statistical software package StatPlus Pro 2009, AnalystSoft Inc. Data were expressed as average arithmetic with definition of a standard (mean square) error. Belonging to normal distribution was defined when calculating criterion of Kolmogorov-Smirnov and the

accompanying indicators. A P-value < 0.05 was considered statistically significant.

RESULTS

Lymph nodes reach the maximum development to three-months age of young rats of Wistar. It precedes involution of lymphoid tissue at the natural rate of aging. The structural and functional answer of a lymph node is optimum at this age and has rather high degree of correlation with the content of trace elements. Structural and functional zones occupy the different area in the standardized lymph nodes. Parameters of structures define a row: mesenteric — inguinal — tracheobronchial lymph nodes from the smallest to the greatest value. The structure and a microelement profile of lymph nodes differs in each lymphatic region.

The mesenteric lymph node has the cortical and medullary ratio equal 1.33±0.14 in comparison with other lymph nodes. Such value of a cortical and medullary ratio determines the smaller area occupied by medullary cords (18.25±0.82%) and lymphoid small follicles (8.60±0.21%) and vice versa, high index of T- and B-zones ratio (1.24±0.17) and also the area occupied by a sinus system (11.89±0.41%). Construction features of the mesenteric lymph node are combined with the maximum content of Cu, Se, an average content of Fe, Zn, the minimum content of Mn (Table 1).

The tracheobronchial lymph node distinguishes the high cortical and medullary index (2.20±0.13) and the lymph node has the big B-zone consisting of medullary cords (24.26±0.54%) and lymphoid follicles (11.46±0.28%). Low ratio of T- and B-zones (1.02±0.09) and the small area, occupied by a medullary lymphatic sinus (5.25±0.26%) in a lymph node is observed. Morphological variant of the tracheobronchial lymph node are combined with the smallest content of Fe, Cu, Zn, an average content of Mn, Se (Table 1).

The inguinal lymph node is characterized by average parameters of compartments at the size of cortical and medullary index equal 1.97±0.10 and a ratio of T- and B-zones — 1.08±0.13 in comparison with mesenteric and tracheobronchial lymph nodes. The inguinal lymph node has the maximum comtent of Mn, Fe, Zn, average content of Cu, minimum content of Se (Table 1)

It is obvious that trace elements are necessary for support of lymph nodes activity in young age. Microelements participate in a differentiation and proliferation of the immunocompetent cells forming compartments in lymph nodes [1–3]. This fact will be significant for the relationship between trace elements and compartments of a lymph node. Morphofunctional the analysis showed prevalence of the immune response on cellular type in all lymph nodes at young age. The structure and a trace element profile of lymph nodes carry the region-dependent character, reflecting features of each lymphatic region.

There is a minimization of structurally functional compartments of lymph nodes to reduction of maintenance of microelements in process of aging by 1.5 years of rat life. The structure of lymph nodes changes when aging depending on features of the lymphatic region.

The mesenteric lymph node is characterized by low indexes of structure. The cortical and medullary ratio is 0.73±0.07, the ratio of T- and B-zones is 0.58±0.11. These values of indexes determine the smallest area of lymphoid structures of cortical substance at increase in the area of medullary substance (medullary cords are increased up to 41.68±1.08% and lymphatic sinus are increased up to 9.46±0.43%). Change of structure of the mesenteric lymph node is combined with the maximum content of Se, an average content of Cu, Zn and the minimum content of Fe, Mn (Table 2).

The tracheobronchial lymph node distinguishes high value of structure indexes. The cortical and medullary ratio is 1.79±0.12, the ratio of T/B-zones is 1.28±0.10. These indexes define development of structures of cortical substance (the cortical plateau reaches 15.95±0.34%, the paracortex — 25.53±0.57%, lymphoid follicles — 3.22±0.19%) at the smallest area occupied by medullary cords (24.48±0.58%) and a lymphatic sinus (6.71±0.39%). Morphological variant of the tracheobronchial lymph node are combined with the maximum content of Cu, an average content of Se, Mn, Fe, the minimum content of Zn (Table 2).

The inguinal lymph node is characterized by average parameters (the cortical and medullary ratio is 1.11±0.10, a ratio of T- and B-zones is 0.91±0.12) at the increased lymphatic sinus system (12.24±0.44%) in comparison with mesenteric and tracheobronchial lymph nodes. The ingual lymph node distinguishes the maximum comtent of Mn, Fe, Zn and minimum content of Se, Cu (Table 2).

There is forming of regional specifics of the structure and a microelement profile of lymph nodes when aging. according to the drained lymphatic region. The morphofunctional analysis showed prevalence of the immune response on humoral type in mesenteric and inguinal lymph nodes and saving the immune response on cellular type in a tracheobronchial lymph node at senile age.

We carried out the correlation analysis for confirmation of relationship between microelements and adenoid tissue. Microelements are necessary for proliferation and functioning of lymphoid (immune) cells

Table 1. *The content of trace elements in lymph nodes of young animals, mkg/g*

Trace elements	Young rats of Wistar (three-months age), lymph nodes		
	inguinal	mesenteric	tracheobronchial
	1	2	3
Mn	4,12 ± 0,32	2,15 ± 0,13	2,54±0,15
Fe	672,5 ± 54,22	254,8 ± 20,66*	221,4±12,12*
Cu	6,45 ± 0,35	6,48 ± 0,47	5,27±0,17*°
Zn	75,6 ± 2,81	68,71 ± 2,52*	58,26±2,30*°
Se	0,96 ± 0,05	1,38 ± 0,05*	1,25±0,06*

Table 2. *The content of trace elements in lymph nodes of old animals, mkg/g*

Trace elements	Old rats of Wistar (age 1.5 years), lymph nodes		
	inguinal	mesenteric	tracheobronchial
	1	2	3
Mn	4,40 ± 0,63	2,71 ± 0,14*	3,34±0,25*°
Fe	523,8 ± 71,91	182,5 ± 14,33*	226,4±14,64*
Cu	4,68 ± 0,27	5,29 ± 0,35	5,37±0,14*
Zn	61,5 ± 2,06	57,27 ± 1,72*	47,36±2,83*°
Se	0,73 ± 0,08	1,14 ± 0,06*	0,81±0,04°

The note to tables 1-2: $*P_{1-2,3} < 0,05$; $°P_{2-3} < 0,05$ — the level of statistical significance of distinctions

[1–4, 7, 8]. Lymphoid follicles are the most important among other functional compartments of a lymph node. Lymphoid follicles are responsible for proliferation of lymphoid cells (lymphopoiesis).

Trace elements show medium-to-high correlation with lymphoid nodules in lymph nodes of young animals. Trace elements differ on structure in lymph nodes of different localization. There is a positive correlation of lymphoid follicles concerning Mn ($r=0.37$, $p < 0.05$), Cu ($r=0.70$, $p < 0.001$) in a tracheobronchial lymph node; negative correlation concerning Zn ($r=-0.51$, $p < 0.01$) in an inguinal lymph node; positive correlation concerning Cu ($r=0.52$, $p < 0.01$), Mn ($r=0.43$, $p < 0.05$), Fe ($r=0.69$, $p < 0.01$) and negative correlation concerning Zn ($r=-0.38$, $p < 0.05$) in a mesenteric lymph node. It is possible to assume emergence of lymphoid-microelement association for ensuring lymphopoietic function at young age. We obtained other data at old animals. The lack of microelements reduces activity of enzymes that, in turn, initsirut decrease in proliferation of cells and a reduction of lymphoid follicles [1, 7–9]. There is a loss of lymphoid-microelement association because of easing or lack of correlation between trace elements and lymphoid follicles. It leads to decrease of function of lymph nodes at senile age.

DISCUSSION

There are several reasons of close attention to a problem of interrelation of microelements and structures of lymph nodes. First, immune function is connected with development of compartments of a lymph node in the process of proliferation and a differentiation of immunocompetent cells [2–4, 7]; secondly, existence of direct or indirect connection between trace elements and enzymes participating in proliferation of lymphoid cells [8], thirdly, purposeful uses of trace elements for correction of the immune response [7, 9]. Trace elements have the modification properties concerning structure of lymph nodes, defining degree of cell-mediated and humoral immunity. Influence of trace elements on the immune response has ambiguous character and depends on localization of lymph nodes. Level of content of trace elements leads to structural reorganization in a lymph node.

We consider that sign of effectively operating compartments is their interaction with trace elements during the different periods of life. Forming of lymphoid-microelement association carries the region-dependent character and leads to forming of a certain morphological type of the structure of a lymph node. Trace elements deficiency causes disturbances in the immune system. [1, 4, 7, 8]. Aging is accompanied by disorganization of internal structure of lymph nodes depending on concentration of trace elements. The imbalance of structure and trace elements in a lymph node is the reason of decrease immune and drainage functions of lymph nodes at senile age. Trace elements prevent oxidant-dependent damage of tissue when aging, positively influencing an immunological homeostasis [4, 8]. It is possible to conclude that different concentration of microelements changes structures of a lymph node and forms cellular or humoral type of the immune response.

CONCLUSION

The obtained results show patterns of structural modification of lymph nodes, proceeding from age and features of a microelement profile. Forming of lymphoid-microelement association defines development of lymphoid tissue at young age. The indispensable condition of functioning of lymph nodes is optimum concentration of trace elements. Trace elements enter cofactors of enzymes and possess the regulatory and structural stabilizing function in lymph nodes. Easing and loss of lymphoid-microelement association is a predictor of aging of peripheral lymphoid organs. Destabilization of structure and a microelement pro-

file of lymph nodes deteriorates drainage and immune functions in the lymphatic region. It is supposed that prevention of microelements deficiency will allow to improve the structural organization and to increase function of lymph nodes that will slow down process of aging of lymphoid tissue.

REFERENCES

1. **Borodin Yu.I, Gorchakova O.V., Suhovershin A.V., Gorchakov V.N., Fartukov A.V., Kolmogorov Yu.P., Demchenko G.A.** The concept of lymphatic region in preventive lymphology. – LAP LAMBERT Academic Publishing. 2018. – 74p.
2. **Gorchakova O.V., Gorchakov V.N.** Increase in drainage and immune functions of a lymph node as a factor of endoecological wellbeing at advanced and senile age // Advances in gerontology. – 2015. – Vol. 28. – No. 3. – P.521–526.
3. **Gorchakova O., Kolosova N., Gorchakov V., Starkova E., Demchenko G.** Premature aging and structural organization of the mesenteric lymph node // Archiv Euromedica. – 2019. – Vol. 9. – Num. 3. – P.22–24. https://doi.Org/10.35630/2199-885X/2019/9/3.7
4. **Intorre F., Polito A., Andriollo-Sanchez M.** Effect of zinc supplementation on vitamin status of middle-aged and older European adults: the ZENITH study // Eur. J. Clin. Nutr. – 2008. – V.62. – P.1215–1223.
5. **Nomura A., Sugiura Y.** Contribution of individual zinc ligands to metal binding and peptide folding of zinc finger peptides // Inorg. Chem. – 2002. – Vol. 41. – P.3693–3698.
6. **Piminov P.A.** Synchrotron Radiation Research and Application at VEPP-4 // Physics Procedia. – 2016. – Vol. 84. – P.19–26. DOI:10.1016/j.phpro.2016.11.005
7. **Petrov I.M., Gagina T.A., Troshina I.A., Medvedeva I.V.** Modern features of nutrition and immune system // Siberian medical journal. – 2006. – No 6. – P.10–14. [in Rus.]
8. **Kudrin A.V, Gromova O.A.** Trace elements in immunology and oncology. – M.: GEOTAR-Media. 2007. – 548p. [in Rus.]
9. **Zaitseva I.P., Skalny A.A., Tinkov A.A., Berezkina E.S., Grabeklis A.R., Skalny A.V.** The influence of physical activity on hair toxic and essential trace element content in male and female students // Biol. Trace Element Res. – 2015. – V. 163. – No. 1–2. – P.58–66.