IMMUNOGENETIC CHARACTERISTIC THE ANTIGENS OF SYSTEM HLA-ALLELES I AND II CLASS PATIENTS WITH MULTIPLE MYELOMA

Baratova D.A.[1-2], Baratova M.A.[3]

[1] NMU "Eurasian center oncohematology, immunology and therapy"
Saint Petersburg, Russia (195197, St. Petersburg, prospect Polyustrovsky 59, literU),e-mail: baratova@list.ru
[2]:"National Register of hematopoietic stem cells Kirghizia in Saint Petersburg " Saint Petersburg, Russia (195197, St. Petersburg, prospect Polyustrovsky 59, literU), e-mail: baratova@list.ru,
[3] AO " Republican Scientific Center of Emergency Medical Assistance "
Astana, Kazakhstan.(010000, Astana, street-Kerey-Janibek handar, 3), e-mail:maksat-7brt@list.ru

Summary: The article presents the results of the research immunogenetic characteristics antigens system HLA-alleles I and II class the patients of multiple myeloma кirghiz nationalities and in the comparison with patients multiple myeloma of the residents North-West region of the Russian Federation.

Since, how in the system HLA , antigens are significantly different among ethnic peoples and nationalities. Given work was undertaken for the first time among patients with MM and in the complex by laboratory methods revealed important markers HLA characteristic for patients multiple myeloma кirghiz nationalities. In comparing the of genes HLA - alleles loci(A and B), results of analyzes showed, that the have characteristic for multiple myeloma кirghiz ethnic patients: antigens HLA-A28, HLA- B13, HLA- B17 and are less common antigens HLA- A24, HLA-A25, HLA- B35 and at the HLA-locus DRB1* are identified the widespread antigens HLA-DRB1* 0901, HLA-DRB1* 1001, HLA-DRB1*1601 and less encountered gene - HLA -DRB1*1401and has enough statistically authentically differences.

Thus, the polymorphism of HLA system makes it possible for, for unique population-based studies, and the conducted in order to study of genetic differences between populations, interpopulation and establishing the degree of their isolation. The system HLA is one of complex systems in the immunogenetic areas of the human natures. Consequently, it requires about the necessity and the importance of early of its study and addressing the issue in the fight with malignant onco-hematological and autoimmune diseases. Revealed differences in comparing with control groups of the ethnic population at the present stage will allow timely choice treatment and consider in the selection donors when planning with closely related, unrelated transplantation hematopoietic stem cell.

Key words: HLA, multiple myeloma, kirghiz nationality.

Introduction:

At present the problem morbidity with multiple myeloma continues to remain rather actual and contemporary. For today early identification the disease, which is of importance the searching immunogenetic criteria of predispositions to various diseases and detection of factors influencing the course of the disease and prognosis.

It is known, that the system HLA is one the of complex immunogenetics of humans system and it requires about the necessity and the importance of early his examining and resolving issues in combating with malignant, onco hematological and autoimmune diseases. From main and important methods of studying linkages between system of HLA and with various diseases is more are inherently population and family analys.

Multiple myeloma (MM)- clonal malignant disease the system of blood, occurring at the level of cell germinal center of lymph nodes, main morphological substrate, which the are plasmatic cells in varying degrees of maturity,capable of producing a structurally homogeneous immunoglobulins.

Multiple myeloma occurs in countries far and near abroad countries and among different races and considered as "disease of the elderly." The average age of patients is 68 years, however, in recent years there has a trend towards rejuvenation of the disease.

Most often detected MM on late terms because, no markers of malignant disease, especially at an early stage of its development,

The emergence and development of MM - is a complex process associated with the combined effect of diverse exogenous and endogenous factors. Study of diagnostic value and role of the system «human leukocyte antigen» (HLA) with hematological malignancies and lymphoproliferative diseases the has is enough a long history, but previously obtained data of distribution alleles of HLA- antigens in patients with onco hematological pathology rather scarce and contradictory.

The system itself HLA is the most of the from polymorphic systems of human. The each of sublocus has a series allelic genes with varying degrees of the frequencies distribution in human populations

Currently, practically absent works directed on studying the of polymorphism HLA-of genes loci (A and B) and a locus DRB1* among the ethnic peoples and nationalities with multiple myeloma.

However, some researchers noted an increase frequencies of the following antigens among patients with multiple myeloma among the mixed population of the peoples of the USSR-A28, B27 [5], HLA-A11, HLA-B7 [2].

In [3], the author showed, that in patients MM by residents North-Western region of the Russian Federation the antigen HLA-B7 is a genetic marker of MM with low to proliferative activity in women and is associated with dysregulation in the system IL-6, and antigen HLA-A11-characterizes MM with isolated of light product chains of immunoglobulins in men.

The author of [4] showed, that detected and characterized informative attributes had for each immunochemical variants multiple myelomas predisposing HLA-specificity: AI9 and B16 - for myelomas with secretion of Ig G; for option Ig A is HLA-A3, for Bence -Jones HLA-A10, HLA-B18, DRB1 * 09 and DQB 1 * 303. By the disease multiple myeloma at a young age predisposes HLA-A28.

Authors Smith G et al [6] revealed high detectability of frequencies antigen HLA-A9 among white-skinned of US residents suffering from of multiple myeloma with Ig A products.

All specified works were carried out in populations of different ethnic affiliation (on mixed populations).

It is known, that the published data about availability the associations between antigens major histocompatibility complex and multiple myeloma extremely are contradictory.

This may be due to earlier perceptions, about what multiple myeloma is not is a disease associated with by various genes, or antigens of HLA, and because of little known of the disease

among the ethnic of peoples, of nationalities, as well as with technical difficulties and high cost conducting research methods.

However, the to collate data results has led us to use the results of this work [3], devoted to the study of patients with multiple myeloma living in the same geographic area of the North-West of Russia and conduct their own research major histocompatibility of antigens HLA-alleles locus DRB1* class II in patients with of multiple myeloma among кirghiz nationality.

The aim of our research is to study immunogenetic characteristics in distribution on system HLA- alleles 1 and II class in patients with multiple myeloma кirghiz nationality.

MATERIALS AND STUDY METHODS:

The total number of patients with multiple myeloma 44 man, were observed at the National Hospital Ministry of Health of the Kirghiz Republic, in Eurasian center onco hematology, immunology and therapy, patients from citizens of the Kirghiz Republic, of them 22 patients Russian-speaking population of Kirghizia(mixed populations) and 20 patients kirghiz nationality were typing of for studying of immunological and population characteristic on system of HLA-alleles the locus DRB1* class II, with 1995 on 1999 years. in the laboratory of immunology St. Petersburg clinical hospital number 31(Clinical Center of advanced medical technologies) and 2(two) patient kirghiz nationality typing of on system HLA of alleles locus DRB1*class II in the laboratory of "Intermedia"with 2006 on 2012 years (a total of 22 patients kirghiz nationality). Patients with a different stage currents the disease and the age of the patients varies from 33 to 90 years.

Given work was undertaken for the first time among patients with multiple myeloma кirghiz nationality.

Avia transportation and delivery of of analyzes (fresh blood) in conventional containerized for typing of HLA-antigens I class and for typing of HLA-antigens II class was carried out in the mini refrigerated containers at -10 ° C.

These results was compared with the control group - 20 patients with multiple myeloma кirghiz ethnic nationalities typing of from 1995 to 1998 years along system HLA -alleles loci (A and B) class 1 in the laboratory of immunology St. Petersburg clinical hospital of number 31 (Clinical Center of advanced medical technologies) [1] and 98 patients with multiple myeloma residents of the North-West region of the Russian Federation. [3].

TYPING OF HLA-ANTIGENS I CLASS:

Determination of HLA- loci (A and B) phenotypes of surveyed persons was carried out serological method using a panel of sera gistotipiruyuschih antileykotsitarnyh Russian Research

Institute of Hematology and Transfusiology, allowing to define the locus of antigens 17 “A” and 27 “B” the locus of antigens.

TYPING OF HLA-ANTIGENS II CLASS:

Molecular typing of HLA- genes a locus DRB1* was performed by the polymerase chain reaction using a set of primers domestic firm "DNA-technology"(Moscow) allows to select 13 groups of alleles HLA-DRB*1 (basic resolution). Genomic DNA was isolated of peripheral blood mononuclear cell (fresh or frozen at -20°C), stabilized with sodium citrate or EDTA(final concentration 0.5% of anticoagulant), by using a set reagents "NPF DNA technology"(Moscow),or using immunomagnetic method recruiting firm «Dynal»All stages of amplification were conducted on a thermocycler "TERTSIR"DNA technology ", Moscow).The product obtained in the amplification determined by the method horizontal electrophoresis in 3.2% of agrarian gel with visualization under ultraviolet light L= 320 nm.The specificity of the amplification product were compared with standard marker of DNA lengths PUC-09.

Statistical processing of the results obtained included the analysis of standard criteria. X^2-square was used to estimation of reliability differences in the detection certain limitations between control group and the patients MM. Determination of the value "p", corresponding value found. X^2-square was carried out considering of one degree of freedom.

All mathematical calculations and statistical analysis of the overall study was performed using a personal computer using the package application programs for spreadsheets -"Microsoft – Excel M version 7.0, for Windows 95, for Windows-based 2010, Statistica-5.

RESULTS AND DISCUSSION:

Conducted studies among patients with multiple myeloma кirghiz nationalities for the Study of the major histocompatibility complex on system HLA-of alleles 1 -II classes showed that in the results of analyzes are available by the incidence of the most common features of population and been quite distinct series of of alleles in comparison with patients with multiple myeloma residents of the North-West region of the Russian Federation [3].

Table 1

Frequency of occurrence of antigens H LA-class I of alleles of patients with multiple myeloma кirghiz nationalities and residents of the North-West region of the Russian Federation.

HLA (A и B)	The patients multiple myeloma North-West of Russia п=98	The patients multiple myeloma кirghiz nationalities п=22
A24	30% p< 0,05	10%
A 25	42,4% p < 0,05	20%
A28	8,1%	20% p < 0,05
B13	6,9%	30% p < 0,001
B17	7,6%	20% p < 0,05

B35	23,3% $p < 0,05$	10%

According to the results of our study (Table 1) in the comparative analysis of frequency of occurrence of HLA- of alleles at loci (A and B), more likely to occur antigens HLA- A28, HLA-B13, HLA- B17 and less common the antigens HLA- A24, HLA-A25, HLA- B35 in patients with multiple myeloma kirghiz nationality compared with patients with multiple myeloma North-West region of the Russian Federation and have a highly a statistically significant difference.

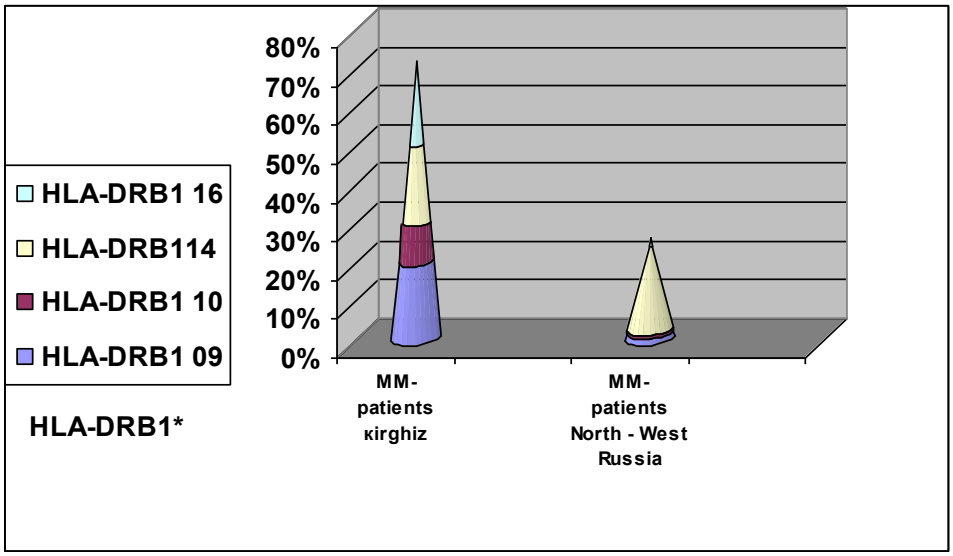

Figure1.Comparison characteristic of system HLA- antigens the locus DRB1*class II at patients with multiple myeloma кirghiz nationality and patients MM residents of the North-West region of the Russian Federation.

In comparative evaluation of results analysis the frequency of occurrence on system HLA- alleles loci DRB1* II class, shown that at patients multiple myeloma kirghiz nationality most often detected genes HLA-DRB1* 0901 in 20% of cases, HLA-DRB1* 1001 in 10%, HLA -DRB1* 1601 in 20% of cases, and several less common gene HLA- DRB1* 1401 in 20% of cases, compared with patients MM residents North-West region of the Russian Federation, the datas which reflected to figure 1.

By importance of differences between patients of multiple myeloma kirghiz nationality and control group is gene HLA-DRB1*1601, that statistically highly authentically $p<0,005$.

CONCLUSION:

Thus, the on results studies in the distribution of HLA alleles 1 and II classes at MM patients, revealed, that the most characteristic for patients with multiple myeloma кirghiz national ethnic identity in the distribution along system HLA -alleles of loci (A and B) class1 genes in the locus A-A28, in the locus B - HLA-B13, B17 and in the HLA- locus DRB1* II class, it appeared that in patients with multiple myeloma кirghiz nationality, more often are found genes HLA-DRB1 * 0901, HLA- DRB1 * 1001, HLA-DRB1 * 1601 and several less frequently HLA-DRB1 * 1401.

Given, that populations of feature HLA- systems is its extraordinary polymorphism.

And the revealed differences in the distribution of HLA-alleles should consider when planning the closely related and unrelated bone marrow transplantation in patients with multiple myeloma and at other of onco hematological malignancies.

REFERENCES:

1.Baratova D.A. Sravnitelnaya characteristic distribution of HLA- alleles I and II class in multiple myeloma patients and healthy individuals кirghiz nationality and residents North-western region of the Russian Federation: Author. dis. cand. honey. sciences. St. Petersburg – 2000. – P.10 –11.

2. Golenkov A., Nazarov I., Petrochenkov A. Serov, L., Sibiryakova L.Raspredelenie HLA- class I antigens in patients with multiple myeloma // Hematology and Blood Transfusion. – 1993. – Vol..2, № 38 –.P.10 –12.

3. Petrochenkov A.N. Major histocompatibility complex antigens in patients with different clinical variants of multiple myeloma: Author. dis. Doc. honey. sciences. St. Petersburg – 1994. – P.4–5.

4. Senkina E.A clinical significance of polymorphism in HLA-specificities of classes I, II and immune disorders in multiple myeloma: Author. dis. cand. honey. sciences. St. Petersburg – 2010. – P. 9 –11.

5. Tananov A.T. Leukocyte antigen (HLA) and human disease // Problems of Hematology and Blood Transfusion —1978. – Vol.21(3) –.P.45–49.

6. Smith G., Walford R.L., Fishkin B. et al. Tissue Antigens. – 1974. – Vol. 4.(4). – P.374-377.