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BASIC BIOCHEMICAL PARAMETERS SIGNIFICANT IN DIAGNOSIS OF MYELOPROLIFERATIVE DISEASES

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Summary. *Myeloproliferative diseases (MPD) belong to the group of clonic malignant diseases of parent cell hematopoiesis, characterized by abnormal increase of one or several blood lines with normal or nearly normal maturing of those cells, both in bone marrow and in extramedullary hemotopoietic organs. The aim of the paper is to determine the basic biochemical parameters that are significant in diagnosis of myeloproliferative diseases. The examination comprised of 219 patients of both sexes, the age between 17 and 83 with the diagnosis MPD. The Values lactate-dehydrogenase (LDH), urates, fibrinogen, erythrocyte sedimentation rate, prothromibnic time (PT) and activated partial thromboplastic time (aPT.T) were monitored among biochemical parameters. Chronic myeloid leukemia is statistically the most spreading form of MPD in comparison with all other types of MPD. The most prominently expressed laboratory finding in subjects with MPD is elevated LDH value, which is statistically more frequent than all others individually. Lactate-dehydrogenesis has been elevated in different levels in all forms of MPD, which is especially obvious in patients with CML and IMF.*

Key words: *Chronic myeloproliferative diseases, urates, fibrinogen, prothrombinic time, lactate-dehydrogenase*

Introduction

Myeloproliferative diseases (MPD) belong to the group of clonic malignant diseases of parent cell hematopoiesis, characterized by abnormal increase of one or several blood lines with normal or nearly normal maturing of those cells, both in bone marrow and in extramedullary hemotopoietic organs (1, 2, 3).

Depending on the dominating cell type, morphological and clinical characteristics, the following diseases are classified into these groups:

A. Chronic myeloid leukemia (Leukemia myeloidea chronica, CML);
B. Polycythaemia vera (PV);
C. Agnogenic myeloid megtaplasia with myelofibrosis or Idiopathic myelofibrosis, IMF;
D. Essential thrombocythaemia (Thrombocythaemia essentialis, ET);
E. Myeloproliferative disease that cannot be classified.

These groups of diseases show similar clinic properties and laboratory changes, especially in the initial phase of disease. It seems that it is a consequence of a similar pathogenesis. Each disease is characterized by distinct proliferation of blood line, although it has been most frequently obvious that the other blood lines were included into proliferation. Overlapping is frequent, so it is not quite simple to differentiate them from each other (1, 2, 4, 5, 6, 7).

In addition to similarities between those diseases, there are also significant clinic and laboratory differences, probably caused by differences in genus changes, which control the response of hematopoietic cells or regulatory factors that determine the type of blood cells that would increase abnormally in some of those syndromes (3, 8, 9, 10).

Aim of the paper

The aim of the paper is to determine the basic biochemical parameters that are significant in diagnosis of myeloproliferative diseases.

Patients and methods

Patients

The examination comprised of 219 patients of both sexes, the age between 17 and 83 with the diagnosis MPD, made at the Clinic for Hematology and Clinic Immunology of Clinic Center in Niš within the period from 1999 to 2005, and of 38 patients of both sexes, of the age from 38 to 78 with normal blood count and myelograph.

Patients with the diagnosis of MPD have been classified into following groups:

I Group – patients with diagnosis CML, comprising 102 patients, of both sexes, the age between 17 and 83.

II Group – patients with diagnosis PV, comprising of 46 patients, of both sexes, the age between 30 and 78.

III Group – patients with diagnosis IMF, comprising of 26 patients, of both sexes, the age between 42 and 78.

IV Group – patients with diagnosis ET, comprising of 19 patients, of both sexes, the age between 38 and 79.

V Group – patients with the disease that has not differentiated yet into the direction of some myeloproliferative diseases up to the end of research, and having been registered under the diagnosis of Syndrome myeloproliferativum (MPS), comprising of 26 patients, of both sexes, the age between 37 and 76.

Control group of patients comprised of 38 subjects, of both sexes, of the age between 38 and 78, with normal blood count and myelograph.

Methods

For the realization of the given objectives, the following methods were used:

1. Determination of essential biochemical parameters in blood:
   - LDH (normal values 230 – 460 IU/L);
   - Uric acid (normal values: 180 – 420 μmol/L for men and 150 – 360 μmol/L for women);
   - Fibrinogen (normal values 2 – 6 g/L);
   - Erythrocyte sedimentation rate (maximum values are 10 mm/h for men and 20 mm/h for women, by method according to Westergren);
   - Prothrombinic time (determined by the method according to Quick, with normal values 70 – 140%);
   - Activated partial thromboplastic time – aPTT (determined by the method according to Rappaport, with normal values 25 – 35s).

2. Statistical methods for processing and analysing the obtained results. The obtained data are presented graphically and in tabular form. The usual parameters of descriptive analysis were used: incidence (n), percentage (%), mean value ($\overline{X}$), standard deviation (SD), minimum (Min), maximum (Max) and coefficient of variation (Cv). ANOVA's analysis and Student's t-test were used as parametric statistical tests for statistical analysis of parameters with continual values. Pearson's χ^2 was used as a non-parametric test, for comparing the incidence of modalities of other parameters. Statistical validity of tests has been expressed according to the standard probability levels (p) of 0.05, 0.01 and 0.001. Statistical analysis and graphic presentation of the obtained results were carried out by MS Excel and SPSS 8.0 of programme package.

Results

As it can be seen from tables 1, 2 and 3, 219 patients were included in this investigation. Among them, 102 (46.58%) patients were female, while 117 (53.42%) were male. The average age of patients was 61.25 ± 11.90 /from 17 to 83 years.

Table 1. Presence of patients according to the type of MPD

Group	n	%
CML	102	46.57
PV	46	21.00
IMF	26	11.88
ET	19	8.67
MPS	26	11.88
Totally	219	100.00

Chronic myeloid leukemia is statistically the most spreading form of MPD in comparison with all other types of MPD ($p<0.001$).

Table 2. Sex structure of patients with MPD and control groups

Group	Male		Female		Totally	
	n	%	n	%	n	%
CML	52	50.98	50	49.02	102	100.00
PV	25	54.35	21	45.65	46	100.00
IMF	19	73.08	7	26.92	26	100.00
ET	9	47.37	10	52.63	19	100.00
MPS	12	46.15	14	53.85	26	100.00
Control	19	50.00	19	50.00	38	100.00
Totally	136	52.92	121	47.08	257	100.00

Table 3. Basic descriptive parameters of age according to groups of subjects

Group	$\overline{X}$	±	SD	Cv	N	Min	Max
CML	59.08	±	13.49	22.83	102	17	83
PV	62.22	±	10.33	16.60	46	30	78
IMF	62.42	±	8.23	13.18	26	42	78
ET	66.11	±	10.98	16.61	19	38	79
MPS	63.35	±	10.42	16.45	26	37	76
Control	59.84	±	11.01	18.40	38	38	78
Totally	61.25	±	11.90	19.27	257	17	83

Pearson's χ^2 test among examined groups did not succeed in establishing significant differences related to the presence of sexes. Anova's analysis did not statistically determine significant differences among average age among the examined groups ($F=1.74$, $p=0.1261$). Comparing the groups individually, with each other reciprocally, statistically significant difference was determined between CML and ET group ($p<0.05$).

Lactate-dehydrogenase (LDH)

185 (84.74%) patients with MPD had elevated values of LDH. The largest number of subjects with elevated values of LDH was in the group with CML (93.14%). It is statistically significantly larger in comparison to all other groups individually ($p<0.01$), except in the group with IMF in which the percentage of those subjects is the second in value (88.46%) (figure 1). In control group there were no subjects with elevated values of LDH. Statistically significant connection of a

number of subjects with elevated values of LDH and the type MPD (χ^2 =14.93, p<0.01) was obtained by Person's χ^2 test.

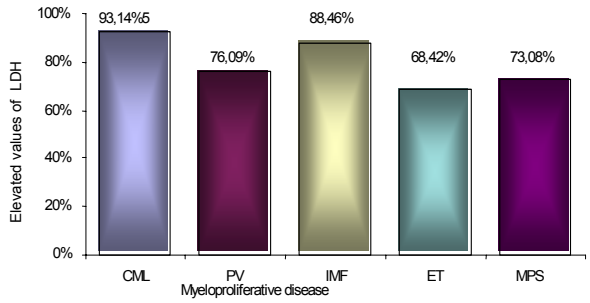

Fig. 1. Elevated values of LDH according to type of MPD

Urates

Among all subjects with MPD, 117 (53.42%) had elevated urate values. Value of χ^2 test of 4,34 together with the number of levels of freedom 4, points out that the percentage of subjects with elevated urate values is not in statistically significant relation to the type of MPD. There is a statistically significant difference in frequency of elevated urate values among some types of MPD. Virtually, statistically significant higher presence of subjects with elevated urate values (χ^2 =3.56, p=0.0591) in patients with MPD in relation to the control group was obtained by Person's χ^2 test (table 4).

Table 4. Value of urates in subjects with MPD according to the type of MPD

Group	Elevated values of urates		Normal values of urates		Totally	
	n	%	n	%	n	%
CML	54	52.94	48	47.06	102	100.00
PV	28	60.87	16	39.13	46	100.00
IMF	16	61.54	10	38.46	26	100.00
ET	9	47.37	10	52.63	19	100.00
MPS	10	38.46	16	61.54	26	100.00
MPD totally	117	53.42	102	53.42	219	100.00

Fibrinogen

One third of the subjects with MPD (33,33% or 73 patients) had elevated fibrinogen values (figure 2). Statistical dependence of a number of subjects with elevated fibrinogen values and the type of MPD was established (χ^2 =13.69, p<0.01). The largest number of subjects with elevated fibrinogen values was in a group with IMF, statistically significantly larger in comparison with MPS (p<0.05) and PV (p<0.01). It can be said that there is a significantly larger number of subjects with elevated fibrinogen values (p<0.001) in the group with MPD. There were no subjects with fibrinogen outside reference values within control group.

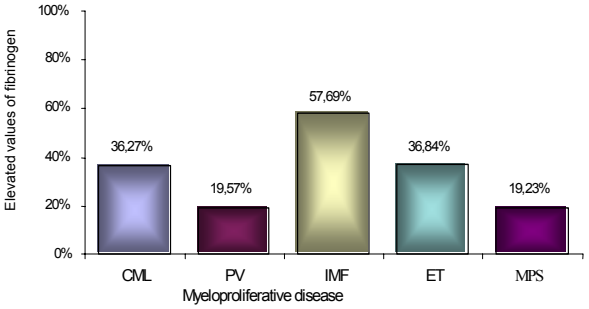

Fig. 2. Elevated values of fibrinogen according to the type of MPD

Erythrocytes sedimentation rate

Rapid sedimentation was determined in 67.58% of patients with MPD, and in 31.58% in control group, that is statistically significant difference (p<0.001).

The largest number of subjects with rapid sedimentation was noticed in patients with CML (over 90%), which is statistically significant in comparison with patients ET (p<0.01), MPS and PV group (p<0.001) (figure 3).

It has been distinctive that in a group with PV only 3 patients (6.52%) are noticed to have rapid sedimentation, which is even 5 times less in percentage, in regard to the control group.

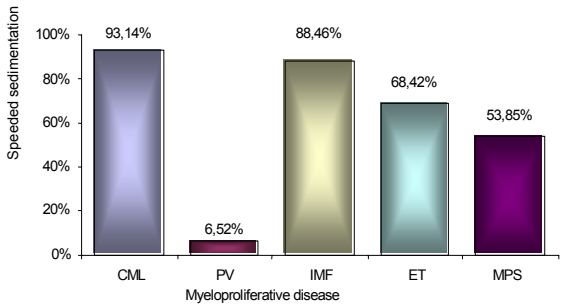

Fig. 3. Rapid sedimentation in patients with MPD according to the type of MPD

Prothrombinic time

Over 40% of subjects with MPD (88) had extended prothrombinic time (PT). The largest percentage of subjects with extended values PT is found in the group with CML (about 52%), that is statistically significantly higher in comparison to IMF and MPS (p<0.05). All subjects in the control group had normal prothrombinic time (figure 4). Statistically more significant presence of extended PT in subjects with MPD in comparison with the control group (p<0.001) was determined. From the table of the contingency 5 x 2, statistical dependence of subjects with elevated values of PT and the group MPD (χ^2 =13.75, p<0.01) was established.

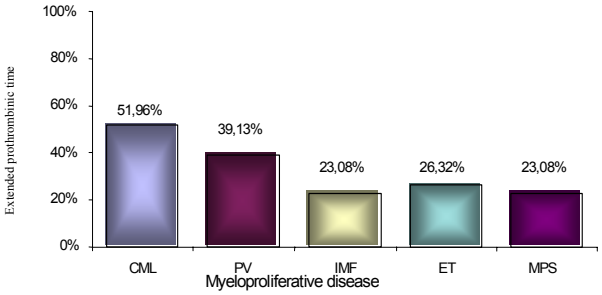

Fig. 4. Extended prothrombinic time in patients with MPB according to the type MPD

Activated partial prothrombinic time (aPTT)

Slightly more than the fourth of subjects with MPD (27,40%) had extended aPTT and the number of subjects showed significant difference in comparison with the type MPD (χ^2 =13.31, p<0.01). The largest number of subjects with values aPTT that are extended is found in the group with IMF (50%), which is statistically significantly higher in comparison with CML (p<0.05) and PV (p<0.001) (table 5). The presence of extended aPTT is statistically significantly more frequent in the group of subjects with MPD in comparison with the control group (p<0.001). In the control group none of the subjects had aPTT outside reference values.

Table 5. Activated partial thromboplastic time (aPTT) in subjects with MPD

Group	Extended aPTT		Normal aPTT		Totally	
	n	%	n	%	n	%
CML	28	27.45	74	72.55	102	100.00
PV	5	10.87	41	89.13	46	100.00
IMF	13	50.00	13	50.00	26	100.00
ET	6	31.58	13	68.42	19	100.00
MPS	8	30.77	18	69.23	26	100.00
MPD totally	60	27.40	159	72.60	219	100.00

The most prominently expressed laboratory finding in subjects with MPD is elevated LDH value, which is statistically more frequent than all others individually (p<0,001) (figure 5).

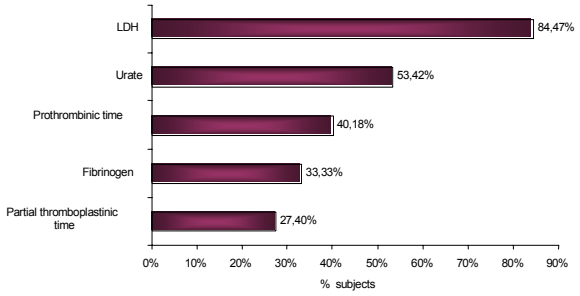

Fig. 5. Laboratory findings in subjects with MPD

Discussion

The Values lactate-dehydrogenase, urates, fibrinogen, erythrocyte sedimentation rate, prothromibnic time and activated partial thromboplastic time were monitored among biochemical parameters. LDH values in blood of patients with CML are elevated due to disintegration of increased number of leukocytes (11, 12, 13). Some literature data show that the LDH value in those patients was elevated in more than 90% (41). There were elevated LDH values found in 95% patients (14, 15,16) having IMF. Also in patients with ET and PV the LDH values in serum are usually elevated, although we have not come across the percentage of such increase in literature (14, 17, 18, 19).

In our investigations the explicitly high percentage of subjects (about 85%) with elevated LDH values is found. This increase is the largest in the group with CML and then in the group with IMF. Our results are mainly in accordance with the available data from literature (11, 12, 14, 17, 18, 20, 21, 22).

The elevated serum level of urates in MPD originates from the increased rate of generation and disintegration of leukocytes, having as a result an increased generation of purine derivatives. In literature we can find that 60% of patients with IMF have elevated urate values (15, 20, 23). In patients with PV the quantity of uric acid in serum may be normal or elevated. Hyperuricemia was present in 70% among 127 examined patients in the study written by Parkins et al. in 1964. In 55% among 325 of examined patients in the study written by Berlin et al. in 1975, the elevated values of urates were found (18, 24, 25). Also in patients with ET concentration of urates in serum may be elevated, but in literature there is no data on the percentage of such patients (22, 26, 27).

In our investigations among all subjects with MPD, a half of them had elevated urate values, which is statistically significant in comparison with the control group. The percentage of subjects with elevated urate values is not in statistically significant connection with the type of MPD. Also, there was no significant difference among certain types of MPD in comparison with the incidence of subjects with elevated urate values. The obtained results are in accordance with the literature data (15, 18, 20, 28).

In literature, there are scanty data on the presence of elevated fibrinogen values in MPD. The available data do not show some especially significant discrepancies (19,29). In our investigation, one third of subjects with MPD are found with elevated fibrinogen values. There is a statistical dependence of a number of subjects with elevated fibrinogen values and type of MPD. Elevated fibrinogen values are mostly represented in the group with IMF.

Rapid sedimentation is determined in about two thirds of patients with MPD, which has been statistically significant. The largest number of subjects with rapid sedimentation was found in patients with CML (over 90%). It is distinctive that only 3 patients were found

with rapid sedimentation in the group with PV, which is even 5 times less in percentage in comparison with the control group. This characteristic of patients with PV makes them different from other types of MPD. These results are in accordance with the literature data (11, 14, 17).

We can see from literature that in patients with CML the values of prothrombinic and activated partial thromboplastic time are mainly normal, although sometimes aPTT can be extended (24, 30). In patients with PV, prothrombinic and partial thromboplastic times are mainly normal (21, 30). In 75% of patients with IMF, prothrombinic time has been extended due to decreased V factor of coagulation (30).

In our investigation, over 40% of patients with MPD had extended prothrombinic time. This has been most prominent in the group with CML, where about 52% of patients were found with extended prothrombinic time.

Slightly more than one fourth of the subjects with MPD had aPTT extended. The highest rate in percentage of the subjects with extended values of aPTT was found in the group with IMF (50%), and it has statistically been much more in comparison with CML and PV.

Conclusion

Chronic myeloid leukemia is statistically the most spreading form of MPD in comparison with all other types of MPD.

The most prominently expressed laboratory finding in subjects with MPD is elevated LDH value, which is more frequent than all others individually. Lactate-dehydrogenesis has been elevated in different levels in all forms of MPD, what is especially obvious in patients with CML and IMF.

A half of patients with MPD them had elevated urate values. The percentage of subjects with elevated urate values is not in connection with the type of MPD.

One third of subjects with MPD are found with elevated fibrinogen values. Elevated fibrinogen values are mostly represented in the group with IMF.

About two thirds of patients with myeloproliferative diseases have rapid erythrocyte sedimentation rate, which has been most prominent in patients with CML.

Over 40% of patients with MPD had extended prothrombinic time. This has been most prominent in the group with CML

One fourth of the subjects with MPD had aPTT extended, which is especially obvious in patients with IMF.

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OSNOVNI BIOHEMIJSKI PARAMETRI ZNAČAJNI ZA DIJAGNOZU MIJELOPROLIFERATIVNIH BOLESTI

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Kratak sadržaj: *Mijeloproliferativne bolesti (MPB) predstavljaju grupu klonskih malignih bolesti matične ćelije hematopoeze, koje karakteriše bujanje jedne ili više krvnih loza sa normalnim ili približno normalnim sazrevanjem tih ćelija u kostnoj srži i u ekstramedulskim hematopoeznim organima. Cilj rada je da se utvrdi koji su to osnovni biohemijski parametri značajni u dijagnozi mijeloproliferativnih bolesti. Ispitivanje je obuhvatilo 219 bolesnika oba pola, uzrasta između 17 i 83 godine sa dijagnozom MPB. Pacijenti sa dijagnozom MPB su razvrstani u pet grupa. Od biohemijskih parametara praćene su vrednosti laktat-dehidrogenaze (LDH), urata, fibrinogena , brzina sedimentacije eritrocita, protrombinsko vreme (PT) i aktivisano parcijalno tromboplastinsko vreme (a PTT). Hronična mijeloidna leukemija je bila statistički najzastupljeniji oblik MPB u odnosu na sve ostale tipove MPB. Najizraženiji laboratorijski nalaz kod ispitanika sa MPB je povišena vrednost LDH, statistički učestaliji od svih ostalih ponaosob. Laktat-dehidrogenaza je bila povišena u različitom stepenu u svim oblicima MPB, što je posebno uočljivo kod bolesnika sa CML i IMF.*

Ključne reči: *Hronične mijeloproliferativne bolesti, urati, fibrinogen, protrombinsko vreme, laktat-dehidrogenaza.*