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Importance of Iron Status Monitoring During Erythropoietin Treatment in Uremic Predialysis Patients

Znaczenie monitorowania gospodarki żelazowej u przeddializacyjnych pacjentów z mocznicą

Since recombinant human erythropoietin (r-Hu EPO) has been introduced to the treatment of anemia in uremic patients the issue of optimal therapy appeared (1-4). For proper erythropoiesis not only erythropoietin but also iron, folic acid and B_{12} vitamin are needed (5-9). Iron deficiency is one of the most common factors causing resistance to r-Hu EPO in uremic patients, so its recognition and eventual supplementation is required for optimal hemopoietic response (10, 11).

The aim of presented study, besides monitoring hematological changes, was to measure iron status parameters such as iron, transferrin, ferritin and percentage of hypochromic erythrocytes and estimation of their usefulness in monitoring iron deficiency during r-Hu EPO treatment.

MATERIALS AND METHODS

Studies were carried out on 14 anemic (duration: 2-6, mean 3.5 years) predialysis patients (11W, 3M) with CRF (duration 2-11, mean 5 years) aged 32-65 (mean 50.5 $\pm$10.5) years. Mean hemoglobin (HGB) and creatinine (Cr) concentration were, respectively, 8.26 $\pm$0,6 g/dl and 572 $\pm$143 µmol/l (6.56 $\pm$1.5 mg%). Before r-Hu EPO anemia was treated with iron, folic acid and blood transfusion (mean 989 ml packed red cells). r-Hu EPO (Eprex-Cilag) was given during 24 weeks subcutaneously with initial dose 3 × 50 u/kg b.w. and dosage was changed accordingly to HGB concentration to keep it between 10-12 g/dl (mean dose during 24 weeks 27 $\pm$10 u/kg b.w., 3 × weekly). From the begining of r-Hu EPO therapy iron supplemetation was started. Eight patients were given iron p.o. (Hemofer 1 tabl. = 105 mg, twice a day, from 1 to 24 week), another six after 4 weeks of oral iron were supplied i.v. till 16 week (Ferrum Lek 100 mg Fe^{++} 3 × week) when oral form was reintroduced. 11 patients during first 12 weeks received also folic acid in dose 5-10 mg daily. Hematological parameters were monitored weekly using hematological autoanalyser Technicon H 1. Following parameters were measured: erythrocytes count (RBC), hemoglobin concentration (HGB), mean red cell volume (MCV), hematocrit (HT), mean HGB concentration in erythrocytes (MCHC). The type of

erythropoietic response to r-Hu EPO was determined by counting the percentage of hypo-and hyperchromic erythrocytes together with percentage of micro- and macrocytes. An iron status parameters such as: iron level (Goodwin method) (12), transferrin (Human Transfferin Test Kit Lis No. 085-411, Hyland) (13) and ferritin level (Ferrizime, Abbot) (14) were measured monthly. Simultaneously a kidney function indicators (Cr, BUN, electrolyte, acid base status) were determined. Statistical analysis was performed by paired T-Student test.

RESULTS

Advanced normocytic and normochromic anemia was observed before r-Hu EPO therapy. A gradual increase of HGB concentration, RBC and HT was observed in all patients during r-Hu EPO administration (Table 1). The analysis of parameters characterizing erythrocytes volume (MCV, % macrocytes, % microcytes) and hemoglobin concetration (MCHC, % hypo- and hyperchromic cells) suggests appearance of homogeneous, typical of iron deficiency type of erythropoiesis (Table 2) during r-Hu EPO treatment. Statistically significant increase of the mean percentage of hypochromic cells was observed. That phenomenon appeared during first 2-8 weeks in all patients. We also observed statistically significant decrease of mean percentage of hyperchromic cells and MCHC at the beginning of treatment and increase of mean macrocytosis and MCV (Table 1 and 2). A transient increase of percentage of hypochromic erythrocytes was also noted during infectious diseases.

In iron status parameters following changes were observed (Table 3): a) statistically significant decrease of iron level was observed in all patients particularly during the first two months; b) statistically significant decrease of mean ferritin level was noted in the third month of treatment and then subsequent increase of this parameter; c) no change in mean transferrin level.

In treated group of patients statistically significant increase of mean Cr concentration was observed (572 $\pm$143 μmol/l to 835 $\pm$278 μmol/l) 6.5 $\pm$1.7 mg% to 9.5 $\pm$3.3 mg%). Maintenance dialysis was started in two patients due to infectious diseases (pneumonia, urinary tract infection) (12).

DISCUSSION

Subcutaneous r-Hu EPO therapy appeared an effective and convenient method of treatment of anemia in predialysis patients, which is in agreement with other data (1-4).

Using of hematological autoanalyser Technicon H1 allowed to determine the type of erythropoietic response to r-Hu EPO administration. As we have published previously three different kinds of answer are possible: 1) macrocytic type; 2) hypochromic type; 3) non-hypochromic, without lasting macrocytosis (3, 4). In our group of patients a homogeneous response to r-Hu EPO treatment was observed.

The appearance of high percentage of hypochromic red cells and transient

Table 1. Changes of hemoglobin concentration, hematocrite value (HT), mean cell volume (MCV) and mean cell hemoglobine concentration (MCHC) in treated group of patients during r-Hu EPO administration (X ± SD)

	Duration of treatment in months				
	0	1	2	4	6
HGB (g/dl)	8.2 ±0.6	10.2 ±1.2**	10.5 ±1.4**	10.6 ±1.4**	9.8 ±1.3*
RBC (T/l) * $p < 0.01$ ** $p < 0.001$	2.68 ±0.3	3.25 ±0.4	3.5 ±0.5*	3.5 ±0.4*	3.24 ±0.4*
MCV (fl)	89.9 ±4.1	93.8 ±3.1*	94.5 ±4.9*	90.7 ±6.6	88.9 ±8.8
MCHC (g/dl) * $p < 0.03$	34.1 ±1.9	32.5 ±1.6*	32.2 ±1.5	33.3 ±2.3	34.4 ±4.2

Table 2. Changes in percentage of hypo- and hyperchromic cells, micro- and macrocytes in treated group of patients during r-Hu EPO administration (X ± SD)

	Duration of treatment in months				
	0	1	2	4	6
% hipo	2.2 ±2.9	8.0 ±5.3	12.7 ±9.9*	18.2 ±15.8	2.9 ±2.7
% hiper	3.6 ±3.4	2.2 ±2.6	1.2 ±1.1	0.9 ± 0.7*	1.5 ±0.9*
% micro	1.3 ±0.8	2.1 ±3.8	1.7 ±2.2	1.5 ± 1.2	1.4 ±0.8
% macro	1.7 ±1.5	4.9 ±5.1*	4.6 ±4.8*	3.0 ± 4.7	1.5 ±1.3

Table 3. Changes in the levels of iron, transferrin and ferritin in treated patients during r-Hu EPO administration (X ± SD)

	Duration of treatment in months				
	0	1	2	4	6
iron (μg/dl)	76 ± 23	72 ± 39	53 ±20*	64 ± 27	74 ± 19
ferritin (ng/ml)	151 ±208	132 ±203	73 ±95*	217 ±252	314 ±195
transferrin (mg/dl) * $p < 0.02$	211 ± 87	216 ±106	191 ±99	154 ± 80	147 ± 68*

inconsiderable macrocytosis were typical reactions to r-Hu EPO administration. Macrocytosis was rather caused by the increased reticulocytosis than by vitamin B_{12} or folic acid deficiency. A gradual decrease of percentage of macrocytes without supplementation of these factors additionally confirmed the above consideration. The analysis of hemopoietic response together with iron status

revealed importance of iron deficiency for bone marrow answer to r-Hu EPO. Our observations suggest that hypochromic type of erythropoietic response is typical of predialysis patients.

During r-Hu EPO treatment lowering of the body iron stores occurs due to utilization of that agent in HGB synthesis. Iron deficiency in laboratory tests manifests usually as decreased levels of iron and ferritin and increased transferrin concentration. As showed above the percentage of hypochromic erythrocytes, which increases very fast could be the earliest test for iron deficiency. In presented study a transient decrease of iron and ferritin was observed. The most pronounced decrease was accompanied by the fastes HGB, RBC and HT increase. In some patients slight transferrin increase was noticed simultaneously with low iron and ferritin levels, which was the additional confirmation of iron deficiency.

Previously described ferritin increase during the last three months was due to intensive iron supplementation and appeared probably as a result of severe bacterial infections (3 patients) and chronic liver damage (1 patient). In patients suffering from those infections a low iron concentration and high percentage of hypochromic red cells were observed. Also in this status the last one appeared as the first one.

As it was shown above, iron status influenced the erythropoietic response to r-Hu EPO. In all patients with hypochromic type of reaction the laboratory features of iron deficiency were observed. Monitoring of iron status parameters is an important element of r-Hu EPO treatment, because due to HGB synthesis the body stores of iron draws out (15, 16). Once again we have to underline that iron deficiency is the most frequent cause of inadequate response to r-Hu EPO therapy. According to our data the increase of percentage of hypochromic erythrocytes appeared to be the earliest parameter of iron deficiency indicating the necessity of this factor supplementation.

CONCLUSIONS

1. Subcutaneously given r-Hu EPO is an effective and convenient method of the treatment of anemia in predialysis patients.
2. The typical hemopoietic response of predialysis patients to r-Hu EPO is hypochromic type probably due to low body stores of that substance.
3. The increase of percentage of hypochromic erythrocytes is the most sensitive marker of iron deficiency.

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STRESZCZENIE

14 pacjentów przeddializacyjnych z przewlekłą niewydolnością nerek oraz anemią leczono przez 6 miesięcy erytropoetyną podawaną podskórnie. Parametry morfologiczne krwi były oceniane przy użyciu analizatora hematologicznego Technicon H1.

Zaobserwowano wzrost średniego poziomu hemoglobiny od wartości 8.2 $\pm$0,6 do 9,8 $\pm$1,3 g/dl. Pojawienie się dużego odsetka hipochromicznych erytrocytów, składające się na hipochromiczny typ odpowiedzi, jest przypuszczalnie najbardziej charakterystyczną reakcją na leczenie r-Hu EPO w tej grupie pacjentów.

To zjawisko zostało spowodowane przez niedobór żelaza, co zostało potwierdzone przez ocenę parametrów gospodarki żelazowej.

Wzrost odsetka hipochromicznych erytrocytów wydaje się być najbardziej czułym, wczesnym markerem niedoboru żelaza.