BRIEF REPORT

Presence of Erythropoietin in the Plasma of One Anephric Patient

By J. P. Naets and M. Wittek

The role of the kidney in the production of erythropoietin has been extensively demonstrated experimentally.1-4 In man, no measurable elevation of the erythropoietin plasma level is generally observed in anemic patients with renal insufficiency, as opposed to high titers in anemic patients with normal renal function.5-7 Accordingly, it has been suggested that alteration of the kidney leads to decreased production of erythropoietin, responsible for depressed erythropoiesis in renal disease. However, it must be emphasized that in man, erythropoiesis persists though depressed, after bilateral nephrectomy.8-9 It might thus be inferred that in anephric man, erythropoiesis is either controlled by erythropoietin produced in an hypothetical extrarenal site, or independent of this hormone. Therefore, it seemed of doctrinal importance to report the observation of a high erythropoietin titer in the plasma of one anephric patient.

Materials and Methods

Eleven anephric anemic patients (five female and six male) chronically submitted to hemodialysis previous to renal transplantation, have been studied, from 5 to 323 days after bilateral nephrectomy. The age range was from 19 to 42 years. Thirty-seven determinations of erythropoietin have been performed on anemic plasma samples (hematocrit below 26 per cent, excepted in one case, 27 per cent). Blood was collected with heparin and plasma samples were kept frozen (−10°C) until used. Assay of erythropoietin was carried out on polycythemic mice following the method of Degowin et al.10 Six virgin female TO mice, weighing from 20 to 25 gr., are used for each assay. Polycythemia was achieved by intraperitoneal injection of one ml. of blood (hematocrit 75 per cent) for two consecutive days. Six days after the second transfusion, one ml. of plasma to be tested is injected subcutaneously. Radioiron is injected intravenously by the tail 50 hours later, and Fe 59 incorporation into red cells is measured after 72 hours, assuming a blood volume of 7 per cent body weight. Results from animals whose hematocrit is below 55 per cent are discarded. Bone marrow smears were stained by May-Grunewald stain. Normoblasts were counted on at least 500 cells. Hematocrit was measured by micromethod, Hb concentration...
Table 1.—Hematologic data and erythropoietin plasma titer

<table>
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<tr>
<th>Days*</th>
<th>Hematocrit %</th>
<th>Hemoglobin g %</th>
<th>Reticulocytes %</th>
<th>Urea mg %</th>
<th>Marrow normoblasts %</th>
<th>Fe 59 uptake % (mean ± S.E.M.)</th>
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<td>Saline 0.42±0.139</td>
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*Before and after nephrectomy.

was determined by the oxyhemoglobin method. Blood urea was determined in an autoanalyzer. Reticulocytes were counted by the direct smear method using brilliant cresyl blue, determinations being made from 1,000 cells.

RESULTS

Erythropoietic activity has been observed in only one of the 37 determinations performed in the eleven anephric patients investigated.

The patient with an active plasma was a 39-year-old man found to have albuminuria during a routine examination in 1957. Six years later, symptoms of renal insufficiency appeared, and the diagnosis of chronic glomerulonephritis was made in 1965. Treatment with intermittent hemodialysis was started in March 1966. Blood pressure was 220/120. Funduscopic examination showed hemorrhages. The hematocrit was 23 per cent, Hb 7.4 g per cent, reticulocyte count, 5 per cent. Urea level averaged 190 mg. per cent, creatinine 191 mg. per cent. Bilateral nephrectomy previous to renal transplantation was performed in August 1966 for severe hypertension. Since then, the patient is still awaiting a suitable donor, and is kept alive by hemodialysis. Seven assays of erythropoietin have been performed in this patient after bilateral nephrectomy. An erythropoietin titer of 1.4 St. B units (Fe 59 incorporation into red cells: 19.31 per cent) has been observed after an acute decrease of the hematocrit to 14 per cent, secondary to severe bleeding from the shunt. No activity was detectable in the six other samples, despite hematocrits as low as 19 per cent. Hematologic data including erythropoietin determinations are summarized on Table 1.

DISCUSSION

The observation of a high titer of erythropoietin in one anephric patient implies that in man, at least under certain circumstances, erythropoietin can be produced outside the kidney. Extrarenal production of erythropoietin has been previously observed in anephric rabbits and rats in response to hypoxic or hemorrhagic stimulation, immediately after bilateral nephrectomy. Several authors failed to detect erythropoietin in the plasma of anephric pa-
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These negative results could be imputed to the small number of cases investigated. Furthermore, it seems that a severe anemia is required to increase the erythropoietin titer to a measurable level in these patients. It is noteworthy that the hematocrits of the six patients studied by Denny et al. are superior to 19 per cent, against 14 per cent in the only positive sample among the 37 tested in this study. Erythropoietin was not measurable in the plasma of this patient after transfusion, even for an hematocrit of 19 per cent.

Persistence of erythropoietin production in one anephric patient suggests that erythropoiesis could be dependent on the hormone after bilateral nephrectomy. The observation of some regulation of erythropoiesis related to tissular oxygenation in anephric man agrees with this hypothesis. Erythropoiesis is increased after hypoxic stimulation, and decreases after transfusion. On the other hand, as erythropoietin has been detected in the plasma of anephric man only in one single case of high grade anemia, it is questionable whether erythropoiesis is dependent on the hormone in the absence of severe tissular hypoxic stress.

SUMMARY

Thirty-seven plasma samples from eleven anemic anephric patients have been assayed for erythropoietin determination. In one case, a high titer of the hormone was observed after a severe hemorrhage. The significance of this finding is discussed.

SUMMARIO IN INTERLINGUA

Trenta-septe specimen de plasma ab dece-un anemic patientes anephric esseva essayate pro lor activitate de erythropoietina. In un caso, un alte titro del hormon esseva observate post un sever hemorrhagia. Le signification de iste constatation es commentate.

REFERENCES

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