Significance of Bone-Marrow Scintigraphy in Aplastic Anemia: Concise Communication

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Tc-99m colloid and In-111 transferrin were used in a semiquantitative scintigraphic study of bone-marrow activity in 76 patients with aplastic anemia, the majority of which were severe cases. The results are compared with other known prognostic parameters and with a predictive index formulated from a prior multiparametric analysis performed in 352 cases.

In 47 cases parallel abnormality of Tc and In uptakes was noted and was well correlated with other prognostic factors. Indium uptake is apparently a good indicator of the severity of aplasia; extension of active erythroid tissue, demonstrated with this method, is correlated with prognosis.

In nine cases, excessive In uptake is explained by dyserythropoiesis associated with granulo- and thrombocytopenia (Fanconi's anemia in most cases).

In 20 of our patients, TcSC uptake was excessive compared with that of In and with other prognostic factors. Statistically, this phenomenon carries an unfavorable prognosis but its physiological meaning remains to be defined.

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As new therapeutic approaches are proposed for severe aplastic anemias (homologous bone-marrow transplantation, immunosuppressive therapy), a reliable prognosis is required at the time of evaluation, to permit selection of these cases in which such a risk (*1, 2*) may be justifiable.

There are relatively few reports dealing with prognostic parameters in large series of aplastic anemias. Lynch et al. (*3*) and Najean and Pecking (*4*) both utilized multiparametric analyses to propose a prognostic index based on the results of clinical and hematological data. Although this type of index is well correlated with survival in very severe and relatively mild cases, it is apparently unsatisfactory as a decision-making factor in the intermediate cases (*5*), where the choice of treatment is the most difficult. Accordingly, any method capable of detecting those cases whose survival expectancy is low enough to justify the risk of bone-marrow transplant, or of immunosuppressive therapy, would be useful.

Bone-marrow scintigraphy has been proposed as a method to predict the evolution in aplastic anemia (*6*), but this suggestion was based on small series of cases and without comparison with other prognostic criteria. During the past 3 yr, we have studied 76 cases of aplastic anemia with varying degrees of severity, by the simultaneous use of Tc-99m colloids and In-111 transferrin. The present report is an analysis of the possible clinical significance of the use of these tracers in aplastic anemia, for the prediction of evolution.

MATERIALS AND METHODS

The scintigraphic data obtained were compared with other clinical and biological findings, including the severity of hemorrhage and infection at the time of the study; the severity of the anemia as estimated by the

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frequency of transfusions; granulocytopenia and thrombocytopenia; reticulocytopenia; and the proportion of nonmyeloid cells in bone-marrow smears. Most of the cases underwent a bone-marrow needle biopsy and quantitative estimation of granulocytic stem cells by bone-marrow culture in vitro on a semisolid medium.

Most of the patients were studied before any decision as to therapy had been made (initial evaluation). Apart from the patients assigned to bone-marrow graft, all have been further treated with high-dose androgen therapy. A few have been referred for possible bone-marrow transplant after the failure of previous androgen therapy, or after relapse.

All patients were followed for at least 3 mo after evaluation, or until death. Amont the total of 76 patients studied, 14 received a homologous bone-marrow transplant between 1.5 and 3 mo after initial evaluation and the beginning of androgen therapy; nine other patients received antithymocyte horse serum (*2*) and continued androgen therapy. Cases in which transfusions were stopped and the granulocyte count improved by 500 per cm^3 or more were considered to be androgen-responsive.

Two successive bone-marrow scintigrams were obtained in all cases at the time of initial evaluation. First, Tc-99m sulfur colloid (TcSC) with a mean particle diameter of 30 nm,* was injected. Images were obtained 30 to 60 min later with a scintillation camera equipped with a divergent collimator. After this study, imaging with In-111 transferrin was performed. In-111 chloride† was incubated with normal iron-unsaturated transferrin in vitro; 48 hr after injection of the labeled transferrin, the bone marrow was imaged with a camera equipped with a parallel-hole medium-energy collimator. The Tc-99m dose was 40-60 μCi/kg, and that of In-111 was 30-50 μCi/kg.

The following regions were imaged: liver and spleen areas, anterior and posterior pelvis, hips, knees, cervical and spinal column, skull, and shoulders. Technical parameters (intensity, voltage, windows, etc.) were the same in all cases, and images were obtained by integrating the radioactivity measured during 4 min. Tracer uptake was estimated semiquantitatively, using the scale proposed by Mac Neil et al. (*7*): 0 when uptake was indistinguishable from background, 1 if it were slightly higher, 2 if organs could be delineated, and 3 when the anatomical drawing was satisfactory. Two observers assigned values to each image independently, and means were calculated. Mean uptake of Tc and In in the pelvis, cervical and spinal column, and shoulders was considered as an index of total bone-marrow uptake of the tracers; the index lies between 0 and 3 in the following tables.

Quantitative scintigraphic data were compared with quantitative or qualitative clinical and biological data by using correlation coefficients or analysis of variance (Fischer's and Student's tests).

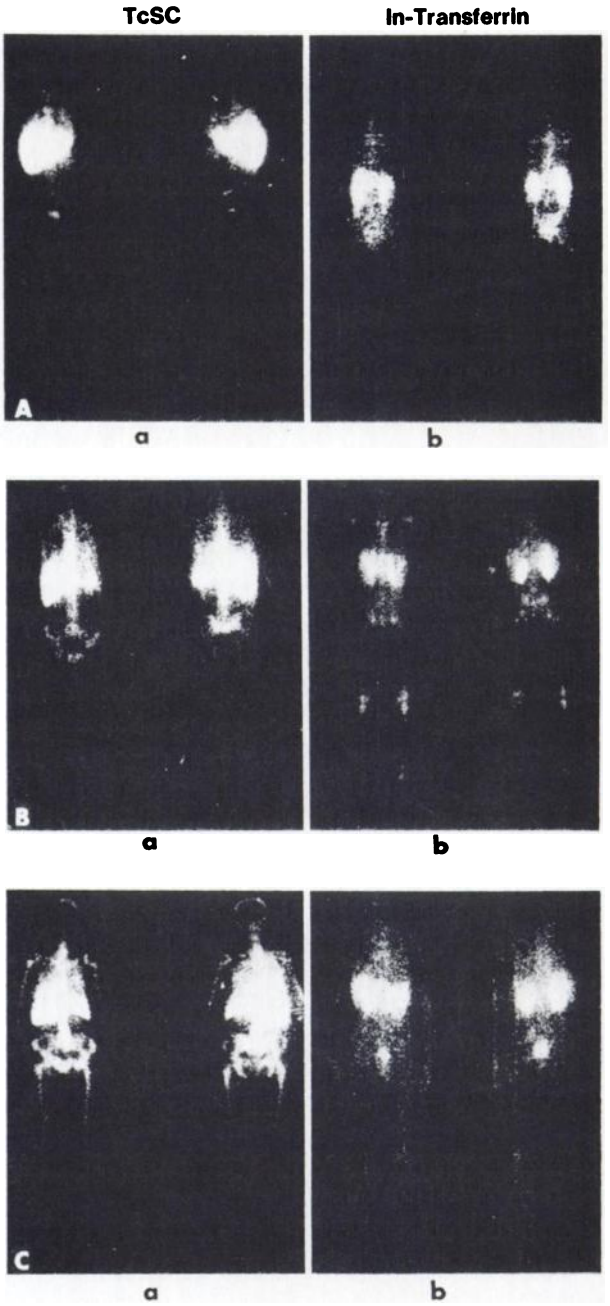

FIG. 1. (A) Similar indium and technetium uptake. (B) Indium-transferrin uptake extending outside central areas. (C) High Tc-colloid uptake associated with low In-transferrin uptake: poor prognostic index.

RESULTS

In this series of 76 cases, *Tc-colloid and In-transferring scintigrams were similar in 47 cases* (Fig. 1A). The correlation coefficient between the quantitative uptakes of the two tracers was 0.84. The results of tracer uptake in these cases were compared with the prognostic index formulated earlier from a multiparametric analysis of 352 cases. The scintigraphic data for both Tc and In

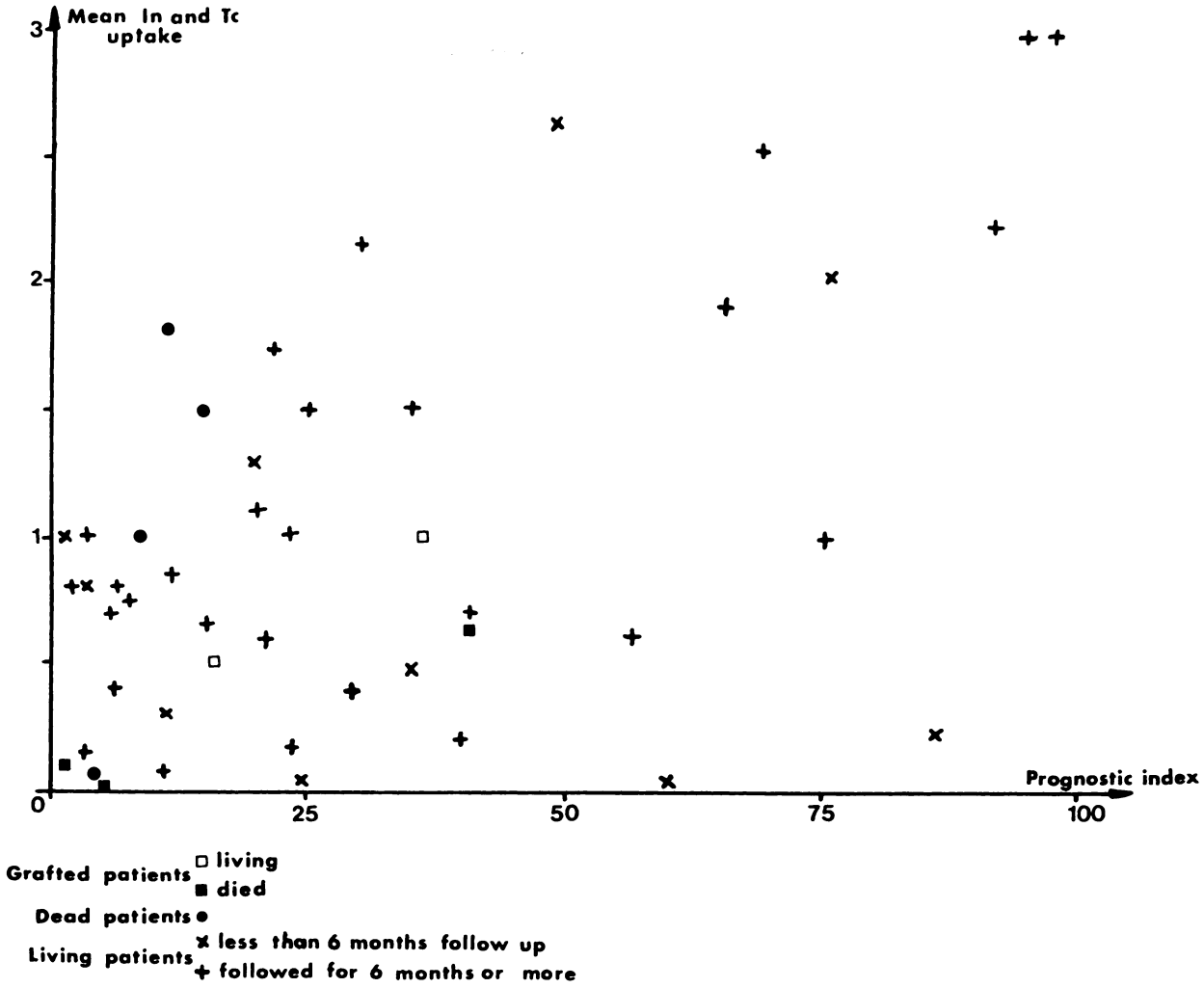

FIG. 2. Correlation between bone-marrow scintigraphy and multiparametric prognostic index in 47 aplastic anemias with similar Tc and In uptake.

uptake were shown to correlate well with this prognostic index (Fig. 2, Table 1). In this group of patients, four died within 2 mo of evaluation and five underwent bone-marrow transplant because of the clinical severity of their disease; all but two of these cases initially had very severe clinical and hematological prognostic parameters, as shown by the low value (<20) of the cumulative predictive index. The predictive index (5) is calculated as follows: − 0.554 MLY − 0.416 HEM + 0.027 RET + 0.024 GRA + 0.234 PLA + 0.053 DEL 0.209 SEX, where:

MLY = percentage of nonmyeloid cells on bone-marrow smears;

HEM = degree of hemorrhagic troubles, from 1 = absent to 3 = severe;

RET = blood concentration of reticulocytes (thousands per mm^3);

GRA = blood concentration of granulocytes (thousands per mm^3);

PLA = blood concentration of platelets (thousands per mm^3);

DEL = delay between diagnosis and the evaluation (in months), and

SEX = 1 for males, 2 for females.

In these cases, the number of granulocytic stem cells was generally very low, so that this parameter does not appear to bring any useful supplementary information (Table 2).

An extension of In and Tc uptake was observed in 19 cases in this group; none of these died or was further grafted.

In nine other cases, In uptake was excessive compared with Tc uptake (Fig. 1B). In all these cases, In uptake extended beyond the central areas.

These patients were all young (5 to 22 yr of age) and five of them presented with unambiguous criteria of Fanconi's anemia. Hematological study demonstrated a contrast between high bone-marrow erythroblastosis

TABLE 1. CORRELATION BETWEEN Tc AND In BONE-MARROW UPTAKE AND PROGNOSTIC PARAMETERS

	No. of cases	Observed value	Prognostic index	Correlation coefficient	Bone-marrow myeloid cells (%)	Correlation coefficient
Similar TcSC and In-transferrin uptake	47	Tc 1.12 ± 0.93 In 1.06 ± 0.84	30.5 ± 29.6	r = 0.51 r = 0.53	43.6 ± 26	r = 0.49 r = 0.58
Excessive TcSC uptake	20	Tc 2.27 ± 0.64 In 0.57 ± 0.42	34.3 ± 24.5	r = 0.23 r = 0.37	42.1 ± 29	r = 0.03 r = 0.22
Excessive In-transferrin uptake	9	Tc 0.83 ± 0.66 In 2.14 ± 0.75	28.5 ± 25.7	— —	56.6 ± 25	— —

TABLE 2. CORRELATION BETWEEN SCINTIGRAPHIC PARAMETERS AND NUMBERS OF GRANULOCYTIC STEM CELLS

	Low stem-cell count (less than 30/10^5 nucleated cells plated)		Stem-cell count higher than 30 per 10^5 cells plated	
	Low In uptake (<1.5)	Normal In uptake (≥1.5)	Low In uptake (<1.5)	Normal In uptake (≥1.5)
Concordant In-transferrin and TcSC uptake	27 cases	8 cases	4 cases	2 cases
Excessive TcSC uptake	7 cases	0 cases	4 cases*	0 cases

* Three of these showed a large number of clusters and a small number of colonies (two early deaths, one bone-marrow graft).

and blood reticulocytosis on the one hand, and low granulocyte and platelet counts on the other.

Bone-marrow culture of granulocytic stem cells (six cases) is in accordance with the low granulocytic-cell cellularity, and the low value of the multiparametic prognostic index.

The clinical severity of two cases (multiparametric prognostic indices of 4 and 16, contrasting with a normal or excessive In uptake) led to bone-marrow transplants. The remaining seven patients are still living at least 6 mo later, on androgen therapy (initial multiparametric prognostic index between 24 and 90).

In the remaining 20 cases, In-transferrin uptake was much lower than that of TcSC. In this group, the multiparametric index was very similar to that measured in the other groups, but, as shown in Table 1, TcSC uptake was excessively high. Twelve of these 20 cases died (or were grafted because of their very poor clinical condition) within 6 mo of initial evaluation.

DISCUSSION

The clinical value of prognostic parameters in cases of aplastic anemia remains a matter of discussion. It is generally agreed (*3, 4, 8, 9*) that the degree of bone-marrow deficiency, estimated from the severity of blood pancytopenia and from the lack of myeloid cells in bone-marrow smears, constitutes a satisfactory short-term prognostic index. It is clear, nevertheless, that a proportion of the most severe cases can improve and be cured if they survive the critical period of the first 3 mo (*5*). Thus, before taking the risks (*1*) of a bone-marrow transplant or drug-induced myelosuppression, we need to discover those cases that, in spite of obviously severe prognostic criteria, could improve on androgen and adequate maintenance therapy.

We have previously shown (*4*) that in vivo incorporation of Fe-59 by erythrocytes is an additional useful prognostic parameter, since it demonstrates the erythroid recovery that generally precedes the recovery of granulocytes—and perhaps that of platelets—in androgen-treated patients. Similarly, it has been suggested (*6*) that In-111 transferrin uptake by bone marrow could be a useful prognostic parameter. Our paper constitutes a critical analysis of this possibility.

The semiquantitative assessment of bone-marrow scintigrams is open to technical criticism, since it takes no account of the weight of marrow in the three regions studied relative to the total active bone marrow. Nev-

TABLE 3. EXTENSION OF TRACER UPTAKE AS A POSSIBLE PROGNOSTIC INDEX*

Multiparametric index of prognosis	Surviving more than 6 mo		Grafted		Dead	
	extension	none	extension	none	extension	none
<50	14 cases	19	1	10	1	9
<25	11 cases	15	1	6	—	9

* In both groups $0.02 > r > 0.01$ when compared with other cases.

ertheless, using postmortem observations on these marrow regions (*10*), we have assigned weighting coefficients to each region, and these did not consistently modify the estimates of bone-marrow radionuclide uptake.

On another hand, although In uptake by bone marrow is generally admitted to be a significant index of erythroid tissue volume (*11, 12*), the true significance of TcSC appears less clear cut (*13*). Its uptake depends not only on the functional activity of the reticuloendothelial and macrophage systems in marrow, but also on its vascularity and on the simultaneous uptake by the liver and spleen, which may vary from one patient to another. Similar In and Tc uptakes were noted, however, in 47 of our 76 cases; 17 successive studies several months apart showed that In and Tc uptakes evolved in parallel; and in ten of these patients, clinical and hematological improvement was accompanied by a simultaneous and parallel increase of In and Tc uptake by bone marrow.

In the majority of our cases, bone-marrow scintigrams closely paralleled other prognostic factors. Thus, in this patient group, the semiquantitative estimation of tracer uptake by the bone marrow does not appear to contribute any new useful prognostic information in addition to those obtained from routine hematological procedures.

The extension of active myeloid tissue, as observed with In-transferrin and TcSC scintigraphy, could, however, be a useful prognostic parameter. As shown in Table 3, such extension appears to be a useful guide to prognosis (Fig. 1B).

In a minority of our cases (nine out of 76 cases analysed), In uptake was far greater than that of TcSC, and was not representative of the clinical severity of the aplastic anemia. This apparent paradox was explained by the observation of the relative intactness of the erythroid line (on bone-marrow slides and biopsy), as is often observed in Fanconi's anemia during the initial phase. We note that, in this group, In uptake is not correlated with the prognostic index (the most important

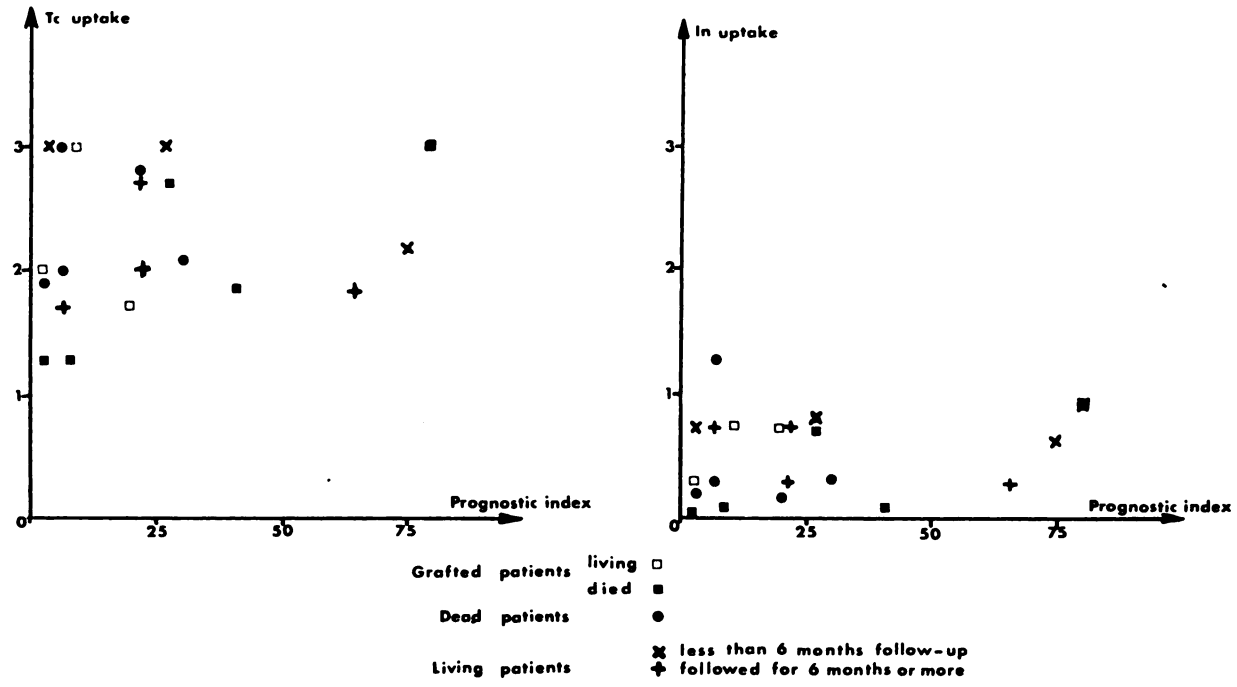

FIG. 3. Twenty patients with excessive Tc uptake in comparison with In uptake: correlation with multiparametric index, and evolution.

parameter being the granulocyte count), or with the stem-cell count.

The most interesting and amazing finding concerns the patient group in which high TcSC uptake was associated with low In uptake and a poor prognostic index. The majority of these patients died or received a transplant as a result of their poor condition (Fig. 3). On the basis of experimental and anatomical arguments, it has recently been suggested that a stromal abnormality could be responsible for the failure of recovery in some cases of aplastic anemia (*14, 15*). Review of our pathological charts indicates that abnormal marrow stroma (edema, hemorrhage, disorganization of the reticulin fibers) and/or abnormal proliferation of nonmyeloid cells ("reticulohistiocytic," or "lymphoplasmocytic" excess) were noted in nine of 12 cases with excessive TcSC uptake, compared with only three of 31 cases with concordant In and Tc uptake and similar hematological severity. It is possible that a currently undefined bone-marrow abnormality may be detected by radioactive colloid uptake, and thus have predictive significance.

The present results are interesting but must be extended, not only by larger series of cases with the same methods, but by search for a better quantitative measure for blood flow and function of the entire bone marrow.

FOOTNOTES

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† Philips-Duphar, Pettens, The Netherlands

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