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# The Importance of Age, Fludarabine, and Total Body Irradiation in the Incidence and Severity of Chronic Renal Failure after Allogeneic Hematopoietic Cell Transplantation

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## ABSTRACT

**Nonmalignant late effects, including chronic renal failure (CRF), impair the quality of life of long-term survivors after allogeneic hematopoietic cell transplantation. One of the major risk factors is the use of total body irradiation (TBI) in the preparative regimen; TBI is currently fractionated in an attempt to reduce toxicity. We analyzed 241 patients who had TBI-based preparative regimens for allogeneic hematopoietic cell transplantation. TBI was delivered as a single fraction of 7.5 Gy (7.5S group), 12 Gy in 6 fractions (12F group), or 14.4 Gy in 8 fractions (14.4F group). The cumulative incidence of CRF at 2 years was 12%. Statistical analysis revealed that older age ($P < .001$) and fludarabine administration ($P = .016$) had a significant effect on the incidence of CRF. Furthermore, single-fraction TBI was also significantly associated with CRF severity, because 7 (6.3%) of 111 patients in the 7.5S group developed severe CRF, as opposed to 1 (0.8%) of 130 patients in the 12F and 14.4F groups combined ($P = .044$). However, these conclusions should be regarded as preliminary in view of the retrospective and nonrandomized nature of this study.**


## KEY WORDS

Renal failure • Total body irradiation • Fludarabine • Hematopoietic cell transplantation

## INTRODUCTION

Allogeneic hematopoietic cell transplantation (HCT) after high-dose chemotherapy and total body irradiation (TBI) is a widely accepted therapeutic approach for several hematologic malignancies [1]. Many patients currently enjoy long-term survival after HCT, and late clinical effects are therefore of major concern. Secondary malignancies are usually a consequence of immunosuppression, high-dose chemotherapy, TBI, or a combination of these [2,3]. Nonmalignant late effects, although rarely life-threatening, significantly impair the quality of life of long-term survivors [4]. The major risk factors for nonmalignant complications after HCT are chronic graft-versus-host disease (GVHD), its treatment, and the use of TBI in the preparative regimen [5].

Although there is still some controversy regarding the mode of delivering TBI, there has been a shift toward the use of fractionated TBI (FTBI), as opposed to single-fraction TBI (STBI), in an attempt to reduce toxicity and improve relapse-free survival [6]. For instance, some long-term complications such as cataracts, avascular necrosis of the bone, and hypothyroidism seem more frequent in patients who receive STBI compared with FTBI [7-9]. Results are nevertheless contradictory in terms of antileukemic activity; some studies show better tumor control with STBI [10,11], and others reach the opposite conclusion [12,13].

Few articles exist on the renal consequences of HCT, although many warn about the potential toxicity that could eventually lead to renal failure [14-16]. Chemotherapeutic drugs, nephrotoxic antibiotics, cy-

closporin A (CSA), sepsis, hypotension, and concurrent liver disease all potentially cause acute renal failure after HCT. This early renal failure is usually reversible and does not seem to be related to the later development of radiation nephropathy [17]. This syndrome, also called *HCT nephropathy*, has been confirmed in animal models [18] and clinical studies [19,20], which have revealed a direct association between TBI dose and renal failure after HCT. However, the exact incidence of HCT nephropathy is unknown, ranging between 0.6% and 13% [21], because of a lack of well-designed analyses to provide cumulative incidence rates and descriptions of risk factors [22].

Single doses of x-ray sufficient to cause radiation nephropathy in laboratory mice do not cause nephropathy when fractionated in multiple doses [23]. We recently detected an unusually high incidence of severe chronic renal failure (CRF) in our group of transplant recipients conditioned with STBI-based regimens and, therefore, decided to assess retrospectively the risk of acute and CRF after STBI- or FTBI-based preparative regimens. Together with this information, we also analyzed the roles of other patient, disease, and posttransplantation factors in the development of renal dysfunction.

## PATIENTS AND METHODS

### Study Patients

From February 1996 to March 2004, data from 301 consecutive patients who had TBI-based preparative regimens for allogeneic HCT at the Royal Free and University College Hospitals were retrospectively collected. Because the main end point of this study was the incidence of CRF, 52 patients who died within 3 months after HCT and 8 patients without sufficient follow-up data were excluded from analysis (see below). Of these patients, 119 had their transplantation at the Royal Free Hospital (RFH) and 122 at University College London Hospitals (UCLH). All patients volunteered written informed consent in accordance with regulatory and institutional guidelines. Details regarding diagnosis, disease status at transplantation, donor type, stem cell source, preparative regimens, and immunosuppression after transplantation are listed in Table 1. Only 5 (2%) patients had had a previous autologous HCT, and 8 (3%) had multiple myeloma, a disease typically associated with renal dysfunction. None of the patients assessed in this study had a nonmyeloablative transplantation.

### Preparative Regimens

Chemotherapy mainly consisted of cyclophosphamide (120 mg/kg in 2 divided doses) alone or in combination with fludarabine (90 mg/m$^2$ in 3 divided doses), alemtuzumab (100 mg in 5 divided doses), or both. Fludarabine and alemtuzumab were administered preferentially to those patients who received T cell–depleted (TCD) transplants to ensure engraftment. According to RFH institutional guidelines, fludarabine doses were reduced by 50% in patients with a glomerular filtration rate (GFR) <60 mL/min/1.73 m$^2$, but this adjustment was not observed at UCLH. Occasionally patients received etoposide (60 mg/kg) or melphalan (140 mg/m$^2$) instead of cyclophosphamide (Table 1).

TBI was commenced after completion of chemotherapy and was delivered as a single fraction of 7.5 Gy (7.5S group) in 111 patients, 12 Gy in 6 fractions (12F group) in 84 patients, and 14.4 Gy in 8 fractions (14.4F group) in 46 patients. Most RFH patients received STBI, whereas all UCLH patients had FTBI in accordance with institutional protocols. The median dose rate was 13 cGy/min in the 7.5S group and 6 cGy/min, with a minimum 6-hour interval between fractions, in the 12F and 14.4F groups. In vivo dosimetry was performed during every treatment session on the central axis and off-axis anatomic sites. In FTBI patients, lung blocks were used to limit the lung dose to 10 Gy. The liver and kidneys were not shielded in any patient.

### Supportive Care

All patients received standard nursing and supportive care protocols. Infection prophylaxis comprised itraconazole suspension 200 mg orally twice daily; acyclovir 5 to 10 mg/kg intravenously every 8 hours; and co-trimoxazole 960 mg orally twice daily from day −7 to day −1, followed by aerosolized pentamidine 300 mg monthly until co-trimoxazole could be resumed. Patients with neutropenic fever were treated with broad-spectrum antibiotics, generally meropenem or piperacillin-tazobactam. Additional agents (teicoplanin and aminoglycosides) were added as clinically indicated. Amphotericin B (1-3 mg/kg/d) was given to patients with unexplained fever that persisted beyond 96 hours. Blood products were universally leukodepleted and irradiated. Cytomegalovirus-seronegative recipients received cytomegalovirus-seronegative blood products.

### GVHD Prophylaxis

GVHD prophylaxis was with a combination of CSA and methotrexate in unmanipulated transplants or with CSA alone or nothing in case of TCD transplants. Methotrexate was infused at a dose of 15 mg/m$^2$ on day +1 and 10 mg/m$^2$ on days +3, +6, and +11. Beginning on day −1, CSA was administered at 5 mg/kg for unmanipulated transplants or 3 mg/kg for TCD transplants. The CSA dose was adjusted to maintain trough blood levels of 300 to 400 ng/mL in unmanipulated transplants or 200 to 300 ng/mL in

**Table 1.** *Patient and Transplantation Details According to TBI Dose*

| Patient Characteristic | Group 1 (7.5 Gy; n = 111) | Group 2 (12 Gy; n = 84) | Group 3 (14.4 Gy; n = 46) | *P* Value ($\chi^2$ or Kruskal-Wallis tests) |
|---|---|---|---|---|
| Sex, male/female (%) | 64/36 | 70/30 | 52/48 | .126 (NS) |
| Median age, y (range) | 27 (5-55) | 30 (9-53) | 38.5 (13-57) | <.001 |
| Median follow-up, mo (range) | 14.7 (3.2-65) | 29.5 (3.3-102) | 12.15 (3.3-81) | .002 |
| Disease, n (%) | | | | <.001 |
| Acute myeloid leukemia | 41 (37) | 4 (5) | 37 (81) | |
| Acute lymphoid leukemia | 36 (33) | 36 (43) | 1 (2) | |
| Chronic myeloid leukemia | 18 (16) | 12 (14) | 1 (2) | |
| Non-Hodgkin lymphoma | 5 (4) | 16 (19) | 1 (2) | |
| Myelodysplastic syndrome | 7 (6) | 2 (3) | 6 (13) | |
| Multiple myeloma | 2 (2) | 6 (6) | 0 (0) | |
| Hodgkin disease | 1 (1) | 0 (0) | 0 (0) | |
| Myeloproliferative disorder | 1 (1) | 4 (5) | 0 (0) | |
| Chronic lymphocytic leukemia | 0 (0) | 4 (5) | 0 (0) | |
| Median pre-HCT GFR, mL/min (range) | 115 (40-289) | 104 (39-286) | 103 (51-177) | .027 |
| Prior autologous transplantation, n (%) | 4 (4) | 1 (1) | 0 (0) | .276 (NS) |
| Donor, n (%) | | | | .003 |
| Matched related | 58 (52) | 50 (60) | 20 (44) | |
| Matched unrelated | 33 (30) | 30 (36) | 13 (28) | |
| Mismatched | 20 (18) | 3 (4) | 13 (28) | |
| Stem cell source, n (%) | | | | <.001 |
| Peripheral blood stem cells | 58 (52) | 9 (12) | 26 (82) | |
| Bone marrow | 53 (48) | 63 (88) | 8 (18) | |
| Grafts, n (%) | | | | <.001 |
| Unmanipulated | 39 (35) | 83 (99) | 1 (2) | |
| T-cell depleted | 72 (65) | 1 (1) | 45 (98) | |
| Conditioning, n (%) | | | | <.001 |
| TBI + Cy ± Alem | 45 (40) | 70 (83) | 20 (44) | |
| TBI + Cy + fludarabine ± Alem | 65 (59) | 0 (0) | 26 (56) | |
| TBI + etoposide | 0 (0) | 8 (10) | 0 (0) | |
| TBI + melphalan | 1 (1) | 6 (7) | 0 (0) | |
| GVHD prophylaxis, n (%) | | | | <.001 |
| Cyclosporin A + methotrexate | 39 (35) | 84 (100) | 1 (2) | |
| Cyclosporin A | 32 (29) | 0 (0) | 45 (98) | |
| Nothing (alemtuzumab in vivo) | 40 (36) | 0 (0) | 0 (0) | |

HCT indicates hematopoietic cell transplantation; GFR, glomerular filtration rate; TBI, total body irradiation; Cy, cyclophosphamide; Alem, alemtuzumab; GVHD, graft-versus-host disease; NS, not significant.

TCD transplants. In the absence of GVHD, the dose of CSA was tapered from day +100. Tapering schedules were modified at the discretion of the attending physicians by disease status and activity of GVHD. In the event of renal dysfunction or other signs of CSA-induced toxicity, its dose was reduced accordingly. In case of posttransplantation hemolytic-uremic syndrome/thrombocytopenic thrombotic purpura (HUS/TTP), CSA was generally substituted by mycophenolate mofetil 15 mg/kg twice daily. TCD was mostly accomplished by adding 20 mg of alemtuzumab to either bone marrow or peripheral blood stem cells, although $CD34^+$ selection was also used in a few patients.

## Histopathology

Renal biopsy tissue was available for review in 5 patients and was processed for light, fluorescence, and electron microscopy. In each case, paraffin-embedded sections were stained with hematoxylin and eosin, Masson trichrome, periodic acid–Schiff, and methenamine silver/periodic acid–Schiff (Jones stain).

## Measurements, Definitions, and Statistical Analysis

Renal function was assessed by serum creatinine (SCr) concentration and estimated GFR, calculated by the modification of diet in renal disease equation [24]:

$$\mathrm{GFR\,(mL/min/1.73\ m^2)} = 186.3 \times \mathrm{SCr\ (mg/dL)}^{-1.154} \times \mathrm{Age\ (y)}^{-0.203} \times 1.212 \text{ if black}, \times 0.742 \text{ if female}$$

According to National Kidney Foundation guidelines, CRF was defined as an estimated GFR of <60 mL/min/1.73 $m^2$ that persisted for at least 3 months [25]. Because most patients received CSA or other calcineurin inhibitors for at least 3 months after transplantation, we decided to extend this period to 6 months. However, patients who died within 6 months after transplantation and had an abnormally reduced GFR for at least 3 months were also considered to

**Table 2.** *Definitions of Chronic Renal Failure Adapted from the National Kidney Foundation Criteria [25]*

| Stage | Description | GFR (mL/min/1.73 $m^2$) |
|---|---|---|
| 1 | Kidney damage with normal GFR | ≥90 |
| 2 | Mild GFR reduction | 60-89 |
| 3 | Moderate GFR reduction | 30-59 |
| 4 | Severe GFR reduction | 15-29 |
| 5 | ESRF | <15 or dialysis |

Chronic renal failure was defined as GFR <60 mL/min/1.73 $m^2$ for ≥6 months. Patients who died within 6 months after transplantation but had an abnormally reduced GFR for at least 3 months were also considered to have CRF.

GFR indicates glomerular filtration rate; ESRF, end-stage renal failure.

have CRF. Severe CRF was defined as a GFR of <30 mL/min/1.73 $m^2$, and end-stage renal failure (ESRF) was defined as a GFR of <15 mL/min/1.73 $m^2$, the need for dialysis, or both (Table 2).

Cumulative incidence curves of CRF, severe CRF, and ESRF were estimated by considering non–renal failure–associated deaths as a competing risk and were compared by using the Lunn-McNeil approach. We analyzed several factors for their association with these outcomes, including age (≤30 versus >30 years), pretransplantation GFR (≤100 versus >100 mL/min/1.73 $m^2$), disease status (first chronic phase/first complete remission versus advanced), preparative regimen (fludarabine versus no fludarabine; alemtuzumab versus no alemtuzumab), TBI (7.5S versus 12F versus 14.4F; S versus F), donor type (related versus unrelated; matched versus mismatched), GVHD prophylaxis (CSA versus no CSA; TCD versus no TCD), and GVHD incidence (no versus yes; grades 0-II versus III-IV). No attempt was made to evaluate the effect of other nephrotoxic drugs, such as aminoglycosides or amphotericin B. Baseline characteristics were compared by using the Kruskal-Wallis or Mann-Whitney tests for continuous variables and $\chi^2$ or Fisher exact tests for categorical factors. In all statistical calculations, unadjusted $P$ values <.05 were considered significant.

## RESULTS

### Chronic Renal Failure

The cumulative incidence of CRF was 12% at 2 years. The median time to renal impairment was 3 months (range, 0.3-54.5 months), and the median peak SCr level was 205 μmol/L (range, 141-745 μmol/L). The median estimated GFR was 40 mL/min/1.73 $m^2$ (range, 10-56 mL/min/1.73 $m^2$) at last follow-up or death. In 98% of CRF patients, ultrasound and/or computed tomographic scans were performed to rule out an obstructive condition, and results were abnormal in 2 of them (6.9%). The first patient was a 45-year-old acute myeloid leukemia (AML) patient who had a nephrectomy as a teenager and developed severe acute renal failure during AML induction chemotherapy. A renal biopsy performed at that time revealed focal segmental glomerulosclerosis by leukemic infiltration. The second patient was a 51-year-old AML patient who had mild bilateral hydronephrosis of no obvious cause. This last patient fulfilled criteria for severe CRF but did not require dialysis (see below).

In terms of risk factors, age at transplantation strongly predicted the risk of CRF. The cumulative incidence of CRF within 2 years of transplantation was 4% for patients <30 years of age compared with 20% for patients who were ≥30 years (hazard ratio [HR], 6.58; 95% confidence interval [CI], 2.29-18.90; $P < .001$; Figure 1). In addition, patients who received fludarabine as part of their preparative regimen had a significantly increased risk of CRF compared with patients who did not receive fludarabine immediately before TBI (HR, 2.57; 95% CI, 1.19-5.52; $P = .016$; Figure 1). Because fludarabine doses were reduced according to renal function at RFH only, a hospital-stratified analysis was performed, which revealed a very similar result. Finally, because CSA has been widely reported to cause reversible renal failure [26] and because 34% of our CRF patients were receiving CSA at the time of the last SCr measurement, we decided to study only those patients not taking CSA or any other calcineurin inhibitor. Not surprisingly, the results were equivalent, and the same variables were identified: age at transplantation and fludarabine administration. None of these variables had a significant effect on non–renal failure–associated mortality by competing risk analysis.

### Severe and End-Stage CRF

The cumulative incidence of severe CRF and ESRF in our cohort was 3.6% at 2 years (8 of 241 patients). Two factors seemed to be significantly associated with the incidence of severe CRF and ESRF: STBI and fludarabine administration (Figure 2). Indeed, 7 (6.3%) of 111 patients who received STBI qualified for severe CRF and ESRF compared with 1 (0.8%) of 130 patients who received FTBI (HR, 8.59; 95% CI, 1.06-69.84; $P = .044$). Also, 7 (7.5%) of 93 patients who received fludarabine in their preparative regimens fulfilled criteria for severe CRF and ESRF, compared with 1 (0.7%) of 148 patients who did not receive it (HR, 11.93; 95% CI, 1.47-97.02; $P = .020$). It is interesting to note that the 2-year non–renal-associated mortality in the STBI group was significantly higher than in the FTBI group (31% versus 9%; HR, 3.844; 95% CI, 1.87-7.90; $P < .001$; Figure 2).

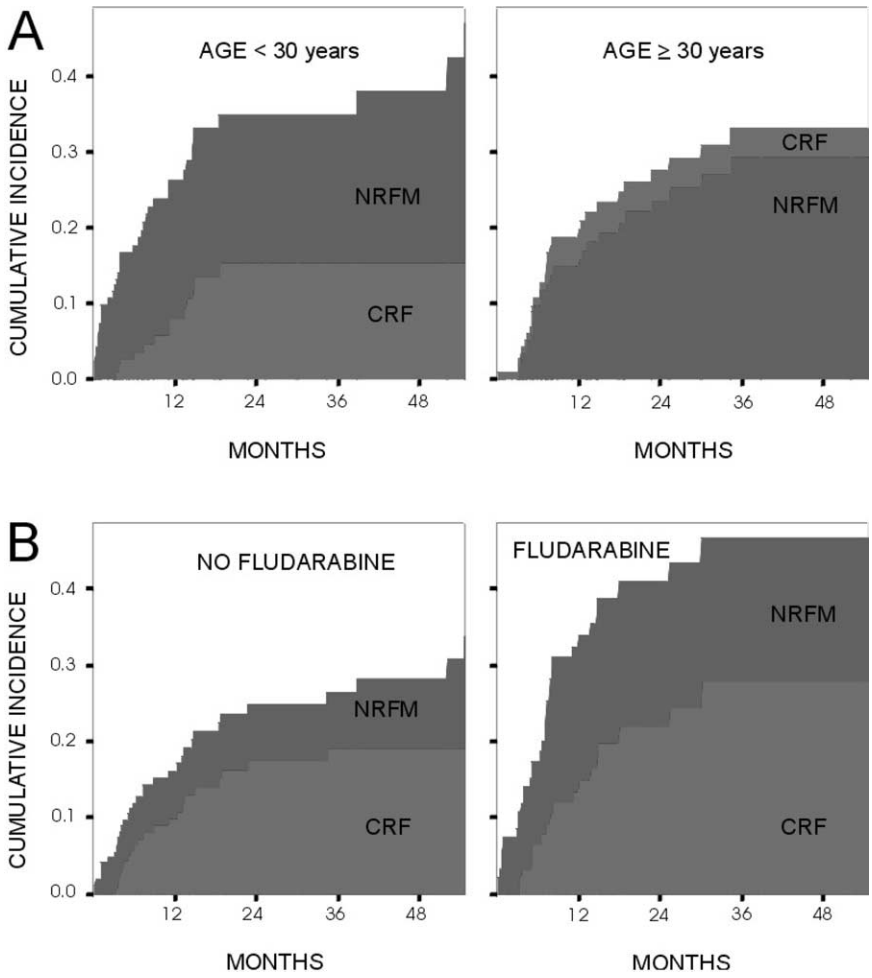


**Figure 1.** Cumulative incidence plots of chronic renal failure (CRF) and non–renal failure–associated mortality (NRFM) according to (A) age at transplantation and (B) fludarabine administration. CRF was defined as an estimated GFR of <60 mL/min/1.73 $m^2$ that persisted for at least 6 months [25].

The characteristics of the 8 patients with severe CRF and ESRF are shown in Table 3. It is interesting to note that the only patient with severe CRF who received FTBI as part of his preparative regimen had bilateral hydronephrosis, which makes it difficult to assess the contribution of TBI or other factors in his CRF. The remaining 7 patients all received STBI, and 6 (86%) of 7 received fludarabine as part of their preparative regimens. Five of these patients presented with clinical and laboratory features suggestive of HUS/TTP at a median of 6.6 months after transplantation (range, 2.3-8.2 months). The hematologic abnormalities (ie, anemia, thrombocytopenia, and an increased number of fragments in the peripheral blood) generally responded to CSA withdrawal with or without plasma exchanges, but the renal failure slowly progressed to severe CRF or ESRF. Renal biopsies performed in 4 of these 5 patients were suggestive of thrombotic microangiopathy/radiation nephropathy. Light microscopy showed irregularity of capillary loop outlines, mesangiolysis, and mesangial hypercellularity, and when silver stains were used, the typical double contours became apparent. Ultrastructural examination revealed subendothelial widening, new basement membrane formation, and foot process fusion [27,28].

The other 2 patients with severe CRF had a different clinical evolution. The first was a 48-year-old man with a low pretransplantation GFR (55 mL/min/1.73 $m^2$) who received a transplant from an HLA-mismatched unrelated donor and developed acute GVHD. He then had CSA-induced renal failure but never recovered after CSA withdrawal. The second patient was a 40-year-old man who developed progressive renal failure 3 months after transplantation of no obvious cause. A renal biopsy sample was consistent with severe tubulopathy, showing marked flattening and disruption of tubular epithelial cells and lymphocytic infiltration with clusters of neutrophils. It is interesting to note that no glomerular abnormalities were suggestive of thrombotic microangiopathy/radiation nephropathy.

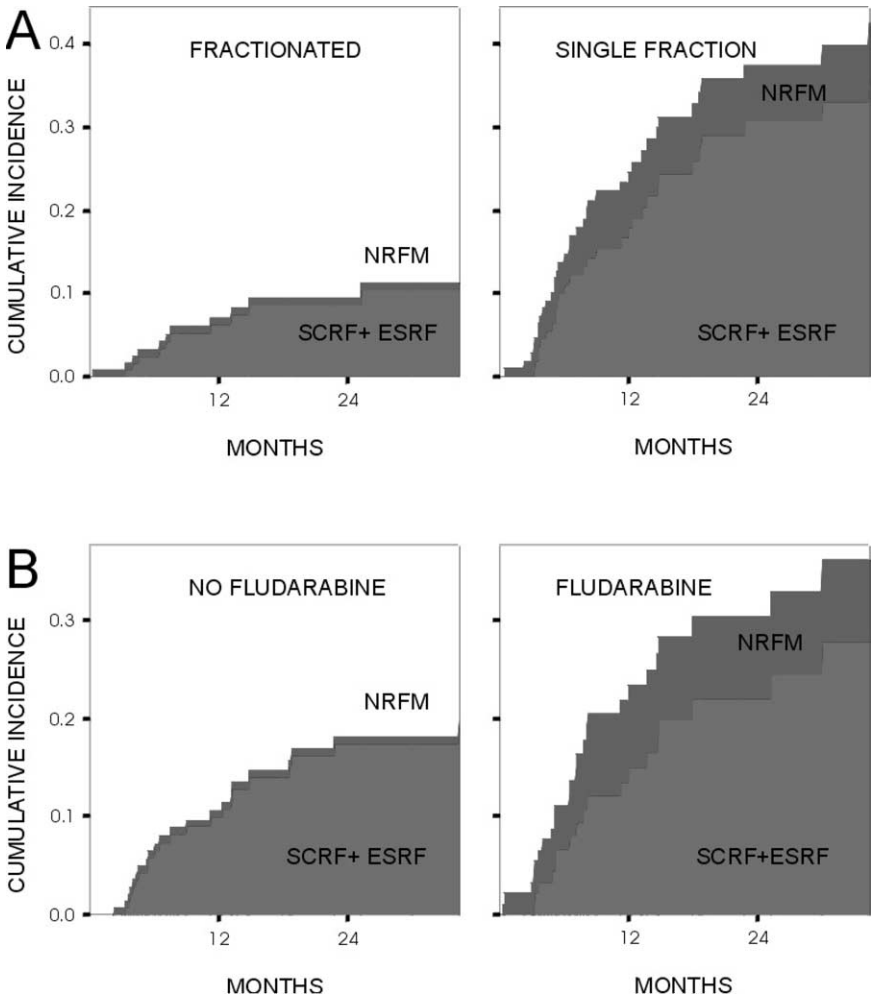


**Figure 2.** Cumulative incidence plots of severe chronic renal failure (SCRF), end-stage renal failure (ESRF), and non–renal failure–associated mortality (NRFM) according to (A) total body irradiation fractionation and (B) fludarabine administration. Severe CRF and ESRF were defined as an estimated GFR of <30 mL/min/1.73 $m^2$ that persisted for at least 6 months [25].

## DISCUSSION

CRF after HCT has been long recognized as related to the TBI used in the preparative regimen [29]. TBI is usually preceded by cytotoxic chemotherapy, which potentiates the effects of radiation [30]. Clinically, when CSA and other nephrotoxic drugs have been ruled out as the cause of CRF, the disorder may be labeled as HCT nephropathy, a form of radiation nephropathy [21]. This condition typically presents 8 to 12 months after HCT as hypertension, progressive azotemia, and dispropor-

**Table 3.** *Characteristics of Patients with Severe CRF or ESRF*

| Age (y) | Donor | GFR (mL/min/1.73 $m^2$) | FLUD | TBI | Median TTRF (mo) | GVHD | TTP | US/CT | Biopsy Results | Dialysis |
|---|---|---|---|---|---|---|---|---|---|---|
| 51 | mMUD | 74 | Yes | F | 0.3 | Yes | No | Hydronephrosis | — | No |
| 37 | MUD | 77 | Yes | S | 7.2 | No | Yes | Normal | TMA/RN | No |
| 36 | MRD | 94 | Yes | S | 6.6 | No | Yes | Normal | TMA/RN | Yes |
| 37 | MRD | 101 | Yes | S | 8.2 | Yes | Yes | Normal | TMA/RN | Yes |
| 30 | mMUD | 170 | Yes | S | 4 | No | Yes | Normal | — | Yes |
| 53 | MRD | 88 | No | S | 2.3 | No | Yes | Normal | TMA/RN | Yes |
| 48 | mMUD | 55 | Yes | S | 0.5 | Yes | No | Normal | — | No |
| 40 | MRD | 104 | Yes | S | 3 | No | No | Normal | Tubulopathy | Yes |

mMUD indicates mismatched unrelated donor; MUD, matched unrelated donor; MRD, matched related donor; GFR, glomerular filtration rate before transplantation; FLUD, fludarabine; TBI, total body irradiation; F, fractionated; S, single fraction; TTRF, time to renal failure; GVHD, graft-versus-host disease; TTP, thrombotic thrombocytopenic purpura; US/CT, ultrasound/computed tomography; TMA/RN, thrombotic microangiopathy/radiation nephropathy.

tionately severe anemia, thus resembling the radiation nephritis described 50 years ago as a complication of abdominal radiotherapy for seminoma [31]. HCT nephropathy may result in a substantial comorbidity due to progressive renal function loss, which frequently requires dialysis or kidney transplantation [32]. Because CRF affects only a few patients who receive TBI, additional factors may influence the development of HCT nephropathy. TBI dose, GVHD development [19], the absence of a renal shielding during TBI, and angiotensin-converting enzyme gene polymorphisms [33] have been suggested as possible risk factors for renal dysfunction after HCT. In this study, CRF was defined as a persistent GFR $<60$ mL/min/1.73 $m^2$ because those patients are at an increased risk of further renal function loss, cardiovascular disease, hospitalization, and death [25,34]. However, most CRF patients are asymptomatic, and therefore other clinically relevant outcomes, such as severe CRF and ESRF, were also assessed. In this analysis, age and the administration of fludarabine as part of the preparative regimen were significant risk factors for CRF, whereas TBI fractionation also determined the incidence of severe CRF and ESRF.

FTBI was originally devised to reduce acute and long-term toxicity after HCT [6,10,12]. Indeed, most ocular, endocrine, and skeletal long-term complications seem to be more frequent in patients who receive STBI as opposed to FTBI [5,7-9]. Also, large retrospective studies suggest that the incidence of solid tumors after transplantation depends on the total cumulative dose administered and its fractionation [2,3]. However, FTBI has never proved to prevent CRF, mainly because very few institutions currently use STBI. In murine models, single doses of radiation able to cause radiation nephropathy are less toxic when fractionated into multiple doses [23]. This increased "tolerance" of radiation with increasing fractionation is probably due to repair of sublethal radiation damage between fractions and to the reduced dose rates used in FTBI (6 cGy/min as opposed to 13 cGy/min). In this study, TBI fractionation had a significant effect on the incidence of severe CRF and ESRF, which confirmed our hypothesis that STBI may have a deleterious effect on renal function. Finally, we suspect that STBI is associated with a higher incidence of posttransplantation HUS/TTP, as was the case in our study (data not shown). This is intriguing because the target cells for both HUS/TTP and HCT nephropathy are the endothelium of arterioles and capillaries, and the resulting histopathologic picture is identical. Indeed, these conditions seem to be associated, because the more severe cases of HCT nephropathy present much like HUS/TTP [19]. However, this hypothesis could not be formally tested because the diagnostic criteria for HUS/TTP were not homogeneous across the entire cohort of patients.

The effect of age on the incidence of renal failure is more controversial. Currently, 5% to 20% of adults who undergo HCT develop CRF [21], whereas several studies have reported higher incidences of CRF in children after allogeneic HCT, ranging from 28% to 45% [35-37]. In children, CRF seems to be less frequent with reduced TBI doses (5-8 Gy), which also decrease the incidence of HUS/TTP [37], but no formal comparison has ever been made. In addition, a different study performed in children with acute lymphoid leukemia reported a CRF rate of only 3%, concluding that pretransplantation chemotherapy rather than young age was the major contributing factor to the lower tolerance of children to TBI [38]. Our study revealed that age at transplantation $>30$ years was significantly associated with a higher risk of CRF. It may be argued that older age at transplantation is usually associated with a lower pretransplantation GFR, as was the case ($P < .001$; Fisher exact test), but age remained significant after adjustment for GFR ($P = .002$), which confirmed its independent effect on the incidence of CRF.

Probably the most interesting finding of this study is the unexpected effect of fludarabine on the incidence of CRF. It has been previously reported that fludarabine enhances radiation-induced damage in both murine [39] and human [40] tumors. Upon cell entry and phosphorylation, the active metabolite fludarabine triphosphate (F-ara-ATP) inhibits several key enzymes involved in DNA synthesis and repair. Potential mechanisms for fludarabine-induced radiosensitization include repair inhibition of radiation-induced chromosome breaks, cell synchronization to a more radiosensitive cell-cycle phase, S-phase cell loss by apoptosis, and inhibition of cell repopulation [41]. Conversely, the effectiveness of this combination depends on the fludarabine dose and the time interval between fludarabine administration and irradiation; the combination is particularly effective when irradiation is delivered as a single fraction at least 24 hours after drug treatment [39,42]. On the basis of the similarities between these tumor models and our TBI-based preparative regimens, we hypothesize that fludarabine may have a radiosensitizing effect on the glomerular endothelium. However, this possible radiosensitizing effect must be closely related to the TBI dose, because the Seattle group recently used fludarabine 90 mg/$m^2$ followed by STBI (2 Gy) as a preparative regimen for allogeneic HCT and did not notice a high incidence of renal dysfunction [43].

In conclusion, this analysis provides data on the incidence and severity of CRF after transplantation in an unselected population of HCT recipients. The incidence of CRF was associated with age and fludarabine administration. Furthermore, STBI seemed to be particularly deleterious because it significantly increased the incidence of severe CRF and ESRF. Therefore, a technique that delivers a maximum dose of 12 Gy in 6

fractions, preferably without fludarabine, could be considered "renally safe" for most patients and could decrease the development of CRF. However, these conclusions should be regarded as preliminary in view of the retrospective and nonrandomized nature of this analysis. Indeed, the potential biases inherent to these studies should be kept in mind when the results are assessed.

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