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RESEARCH

The Predictive Value for Pulmonary Infection by Area Over the Neutrophil Curve (D-index) in Patients Who Underwent Reduced Intensity Hematopoietic Stem Cell Transplantation

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Abstract We evaluated the predictive value of the D-index for pulmonary infection in the early phase of reduced intensity stem cell transplantation (RIST). Out of 68 patients, ten patients developed a pulmonary infection within 100 days after RIST. Both the D-index and the cD-index were higher in the patients with pulmonary infection than in the control group (P=0.009, P=0.042, respectively). The best sensitivity and specificity, calculated with receiver operating characteristic curves, showed that the D-index was superior to the duration of neutropenia in predicting pulmonary infection. We also evaluated the utility of a cumulative D-index until 21 days after RIST (D21-index). The D21-index was higher in the patients with pulmonary infection (P=0.047). The cutoff value of the D21-index was lower than that of the D-index (8650 vs. 11000) with comparable sensitivity and specificity. Our results demonstrate that the D21-index, as well as the D-index, are useful tools for the prediction of pulmonary infection in RIST.

Keywords D-index · cD-index · D21-index · Pulmonary infection · Reduced intensity conditioning regimen

Introduction

Despite marked progress in prophylaxis strategies against microorganisms, infectious complications still remain the main cause of morbidity and mortality in hematopoietic stem cell transplantation (HSCT) [1–4]. Bloodstream and pulmonary infections are the major types of documented infections; severe, prolonged neutropenia is one of the most significant risk factors for pulmonary infection [1, 3]. However, there have been no established parameters to reflect both the intensity and duration of neutropenia. To quantify the degree of neutropenia, Portugal et al. proposed the D-index and the cD-index [5]. The D-index was based on a graph of the neutrophil count during the course of neutropenia (<500/μL) and calculated as the area over the neutrophil curve (Fig. 1). The cumulative D-index (cD-index) was calculated as a cumulative D-index until the first date of the clinical manifestation of infection. These indexes were reported to be useful predictors for invasive mold infection in patients with acute myeloid leukemia (AML). In another study (6), the D-index was also found useful for predicting infectious complications in autologous or allogeneic HSCT. These results prompted us to investigate the utility of these indexes in allogeneic HSCT only, in which prolonged and severe neutropenia has often been observed. Therefore, we decided to analyze the usefulness of the D-index for predicting pulmonary infection in the early phase of reduced intensity stem cell transplantation (RIST).

Patients and Methods

Patient Selection and Transplantation Procedure

Between February 2004 and April 2011, there were 68 patients who received RIST at our hospital. Thirteen patients were excluded from this study because of documented infection prior to the conditioning regimen. We retrospectively analyzed the charts for the remaining 55 patients. The reduced

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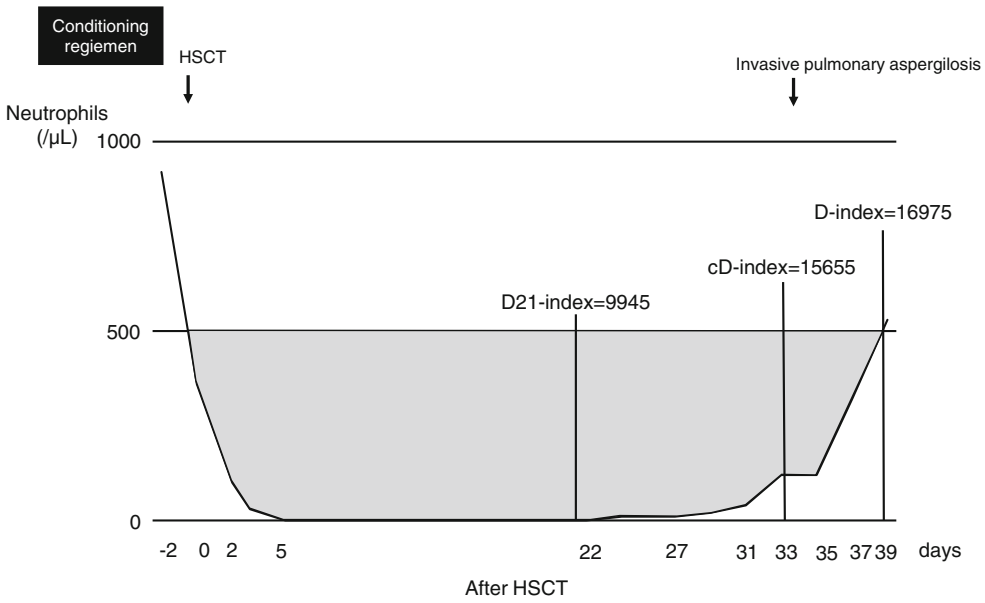

Fig. 1 Calculations of D-index, cD-index, and D21-index based on a patient's data in our cohort

intensity conditioning regimen consisted of either fludarabine (125 mg/m^2); melphalan (80 mg/m^2) and low dose total body irradiation (TBI, 4 Gy); or fludarabine (180 mg/m2), busulfan (6.4 mg/kg) and low dose TBI. Prophylaxis for acute graft-versus-host disease (GVHD) consisted of a short course of methotrexate and cyclosporin A/tacrolimus. Tacrolimus was used in cases of either unrelated or HLA-mismatched transplantation. Prophylaxis for bacterial infection consisted of new oral quinolones (tosufloxacin or levofloxacin). In 21 patients, cephalosporins or carbapenems were administered prior to the conditioning regimen because of fever of undetermined origin. Prophylaxis for fungal infection consisted mainly of fluconazole. Acyclovir was also given for prophylaxis of herpes simplex virus infection. The control group was the group of patients who did not develop pulmonary infection.

Definition of Pulmonary Infection, D-index Calculation, and the Definition of Duration of Neutropenia

Pulmonary infection was diagnosed by a new infiltrative shadow detected by chest X-ray film or computed tomography within 100 days after HSCT.

The D-index and the cD-index were calculated according to previous reports [5]. Briefly, the D-index was calculated as the difference between the observed area under the curve of neutrophils during neutropenia (<500/μL) and the expected area. (500/μL × days during neutropenia) (Fig. 1). The cD-index was calculated as the cumulative D-index from the start of neutropenia until the development of a pulmonary infection.

Cumulative duration of neutropenia was defined as the duration of neutropenia until the onset of pulmonary infection or at 21 days after HSCT (which was defined as the "cumulative duration of neutropenia on day 21"). Since neutrophil recovery was observed more than 21 days after HSCT in about half the patients (n=26) and most patients (8 of 10) developed pulmonary infection after 21 days post HSCT, we also calculated the cumulative D-index between the start of neutropenia and day 21 post HSCT or the onset of infection if the patient was diagnosed with pulmonary infection before day 21 (D21-index), for the evaluation of its predicting power. If a patient had already been neutropenic for any reason when the conditioning regimen was started, the duration of neutropenia and the D-index were calculated from the start of the conditioning regimen.

Statistical Analysis

Categorical characteristics were compared using the χ^2 or Fisher's exact test, and continuous baseline characteristics were compared using the Mann–Whitney U test. To evaluate the utility of the D-index, the cD-index, and the D21-index to predict pulmonary infection, receiver operating characteristic (ROC) curve analyses were performed, and the positive and negative predictive values were calculated. The statistical data were obtained using SPSS II software (IBM SPSS, Chicago, IL, USA). All statistical tests were two-sided, and a P value of less than 0.05 was considered statistically significant.

Results

The patients' characteristics are summarized in Table 1. Pulmonary infections were observed in 10 (18.2 %) patients. Fungal pneumonia (provable) was observed in two patients, invasive pulmonary aspergillosis (provable) in 2,

Table 1 Patient characterist

	Patients without pulmonary infection (control) (n=45)	Patients with Sulmonary infection (n=10)	P-value
Age	60 (23–73)	63 (39–66)	0.965
Sex			0.285
Male	36	8	
Female	19	2	
Underlying disease			0.517
AML	24	4	
ALL	2	2	
MDS	8	2	
SAA	2	1	
Lymphoma	3	0	
Others	6	1	
Antibiotics			0.123
TFLX or LVFX	27	7	
CFPM	6	3	
Others	12	0	
Antifungal agent			0.247
FLCZ	30	5	
ITCZ	4	3	
VRCZ	9	1	
Others	2	1	
Use of systemic steroids			
Yes	21	7	0.18
No	24	3	
Donor source			
Related BM	0	1	0.38
Related PB	8	0	
Unrelated BM	29	4	
CB	8	5	
Duration of neutropenia (<500 μ/L)	17 (3–69)	30 (14–75)	0.011
D-index	7510 (805–33595)	12952.5 (6130–30380)	0.008
Cumulative duration of neutropenia	17 (3–69)	26 (14–42)	0.084
cD-index	7510 (805–33595)	12022.5 (6130–18010)	0.042
Duration of neutropenia on day 21	16 (3–31)	23 (14–32)	0.086
D21-index	7265 (805–15160)	9412.5 (6130–13535)	0.047

Abbreviations: *AML* acute myeloid leukemia; *ALL* acute lymphoblastic leukemia; *MDS* myelodysplastic syndrome; *SAA* severe aplastic anemia; *TFLX* tosufloxacin; *LVFX* levofloxacin; *CFPM* cefepim; *FLCZ* fluconazole; *ITCZ* itraconazole; *VRCZ* voriconazole; *BM* bone marrow; *PB* peripheral blood; *CB* cord blood

pneumocystis pneumonia in 1, cytomegalovirus pneumonia in 1 and an unknown pathogen in 4. The median duration from HSCT to the onset of pulmonary infection was 29.5 (range: 16–82) days. No statistical significance was observed in terms of age, sex, underlying diseases, prophylactic antibiotics and prophylactic antifungal agents, use of systemic steroids, or donor source.

The duration of neutropenia and the D-index were longer and higher than in the control group (i.e., the patients without pulmonary infection). (P=0.011; P=0.008, respectively), whereas there was no statistical difference in the cumulative duration of neutropenia (P=0.084). The cD-index and the D21-index were also higher than in the control group (P=0.042; P=0.047, respectively). Although the duration of neutropenia on day 21 tended to be longer than in the control group, this value showed no statistical difference (P=0.086).

ROC curve analyses showed that sensitivity and specificity tended to be better in both the D-index (ROC area of 0.769 vs. the duration of neutropenia, ROC area of 0.758) and the cD-index (ROC area of 0.707, vs. the cumulative duration of neutropenia, ROC area of 0.676) (Fig. 2a, b; Table 2). The D21-index also showed better sensitivity and specificity compared with the cumulative duration of neutropenia on day 21. The ROC areas were 0.702 and 0.674 for the D21-index and the cumulative duration of neutropenia on day 21, respectively (Fig. 2c; Table 2). When the best sensitivity and specificity

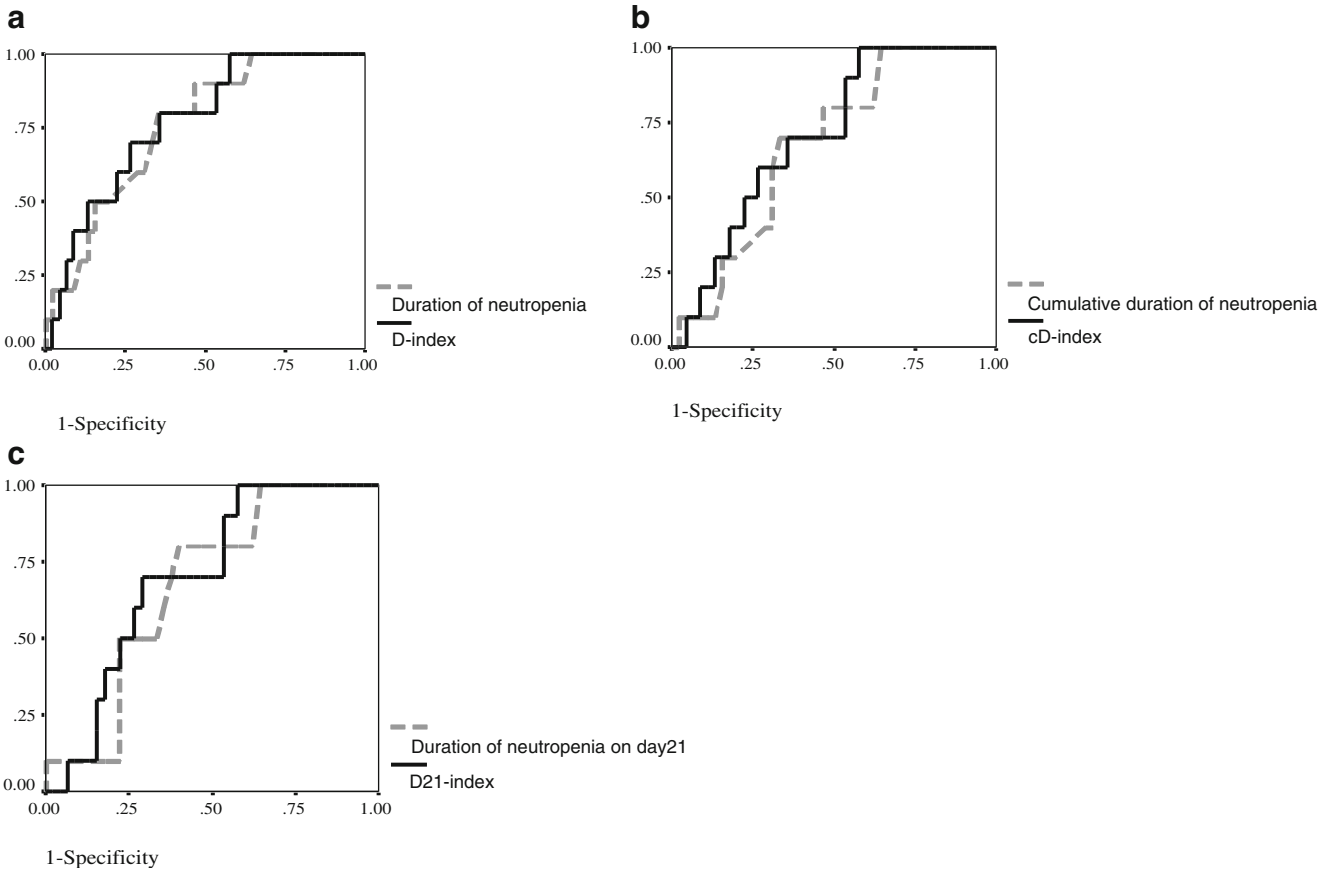

Fig. 2 Receiver operating curves comparing the D-index with the duration of neutropenia (**a**), comparing the cD-index with the cumulative duration of neutropenia (**b**), and comparing the D21-index with the cumulative duration of neutropenia on day 21 (**c**)

were calculated using the ROC curve, the sensitivity and the specificity for the D-index and duration of neutropenia; and the cD-index and cumulative duration of neutropenia were 70.0 and 73.3 % (cutoff value: 11000) and 70.0 and 66.7 % (cutoff value: 24); and 70.0 and 64.4 % (cutoff value: 8870) and 70 and 66.7 % (cutoff value: 24), respectively (Table 2). When these cutoff values were used, the positive and negative predictive values were 36.8 and 91.7 % for the D-index, 31.8 and 90.2 % for the duration of neutropenia, 30.4 and 90.6 % for the cD-index, and 31.8 and 90.9 % for the cumulative duration of neutropenia, respectively (Table 2). The sensitivity and specificity for the D21-index and the cumulative neutropenia on day 21 were 70.0 and 71.1 % (cutoff value: 8650) and 70.0 and 62.2 % (cutoff value: 19) (Table 2). When these cutoff values were used, the positive and negative predictive values were 35.0 and 91.4 % for the D21-index, and 29.2 and 90.3 % for the cumulative duration of neutropenia on day 21, respectively (Table 2).

Table 2 Predicting value for pulmonary infection

	Cutoff value	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	AUC
Duration of neutropenia	24	70	66.7	31.8	90.2	0.758
D-index	11000	70	73.3	36.8	91.7	0.769
Cumulative duration of neutropenia	24	70	66.7	31.8	90.9	0.676
cD-index	8870	70	64.4	30.4	90.6	0.707
Duration of neutropenia on day 21	19	70	62.2	29.2	90.3	0.674
D21-index	8650	70	71.1	35	91.4	0.702

Abbreviations: *PPV* positive predictive value; *NPV* negative predictive value; *AUC* area under the curve

Discussion

In the early period of HSCT, pulmonary infection is one of the most common infectious complications [2]. The D-index, as well as the cD-index, have originally been reported as useful prediction tools for invasive mold infections in AML [5]. Kimura et al. (6) subsequently reported that these indexes were also useful for predicting infectious complications in HSCT, especially for pulmonary infection. Our results demonstrated that the D-index was useful for predicting pulmonary infection in RIST with better specificity and negative predictive value than duration of neutropenia, whereas the cD-index did not show superiority in either sensitivity or specificity compared with the cumulative duration of neutropenia [5]. One of the reasons for this discrepancy may be due to the difference in patient populations. Our study was focused on the patients who underwent allogeneic HSCT with a reduced intensity conditioning regimen, in which severe and prolonged neutropenia is observed more often. Indeed, 10 of 55 patients (18.18 %) developed pulmonary infection within 100 days after HSCT, which was higher than the incidence in other reports [6, 2, 7]. Thus, the evaluation of the D-index in allogeneic HSCT has enormous significance. However, the D-index seems to be somewhat inadequate in allogeneic HSCT because four of ten patents developed pulmonary infection during neutropenia in our study. Furthermore, the cutoff value of the D-index exceeded 10000, which was higher than the cutoff values in previous reports [6, 5]. Thus, we also evaluated the efficacy of the cumulative D-index at a certain point (i.e. 21 days after HSCT), and the D21-index was also shown to be a useful predictor of pulmonary infection in our study. The cutoff value of the D-21 index was lower than that of the D-index (8650 vs. 11000), and its sensitivity and specificity were comparable with those of the D-index. Thus, the D21-index could be a useful predictive tool for pulmonary infection in the earlier stage of HSCT, having equal power to the D-index. The D-index is a unique and powerful tool for the dynamic evaluation of neutropenia. The evaluation of this index for various infections in various HSCT settings (for example, invasive mold infections in the myeloablative conditioning regimen, or with the use of cord blood transplantation) will have enormous significance. Our results showed that the cumulative D-index at a certain point, such as with the D21-index, could also be useful for predicting pulmonary infection. However, statistical difference in our result was quite moderate (P=0.047, Table 1). In addition, in view of the small number of patients in our study, further investigation with a large patient population is warranted.

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Conflict of Interest The authors declare that there are no competing financial interests.

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