target CsA concentration for GVHD prophylaxis at our center (17) might counteract the effect of delayed lymphocyte recovery on the development of acute GVHD. Alemtuzumab-containing regimen was also associated with high L-index(100) values, because alemtuzumab strongly inhibited T lymphocyte for approximately 2 months (16).

Another purpose of this study was to investigate the effect of lymphopenia indexes, including L-index, on CMV reactivation. According to Einsele et al., lymphopenia parameters of ALC < 300/μl, $CD4^+$ cell count < 100/μl, and $CD8^+$ cell count < 100/μl at day 49 after HSCT were significant risk factors for the development of CMV disease in patients with polymerase chain reaction (PCR)-proven CMV viremia (10). Kim et al. showed that cumulative incidence of CMV reactivation in patients with ALC < 350/μl at day 21 after HSCT was significantly higher than that in high ALC group (≥ 350/μl) (2). The result of this study showed that L-index(30) might be more closely associated with CMV reactivation than ALC at day 30. L-index(30) was identified as an independent factor in multivariate analysis when it was dealt as dichotomous variable with a cut-off value of 22,318 determined by ROC curve analysis. This finding suggested that not only the intensity but also the duration of lymphopenia was an important risk factor for CMV reactivation. Furthermore, the area under the lymphocyte curve in the same period did not show statistically significant association with CMV reactivation. Hence, the extent of lymphocyte deficit might be more closely

associated with CMV reactivation than the simple sum of area under the lymphocyte curve. The difference between these 2 parameters becomes important when ALC ≥ 700/μl is achieved early after HSCT. In fact, in this study, ALC exceeded 700/μl at least temporarily within 30 days after HSCT in approximately 20% of patients. On the other hand, L-index(100) was not related to CMV reactivation, probably because CMV antigenemia was detected in more than 3 cells at a median of 29 days after transplantation, and therefore, the cumulative L-index until reactivation might have a significant effect. The limitation of the L-index(30) was that it could be obtained only after 30 days from HSCT, when half of the patients in CMV-AG ≥ 3 group had already developed CMV reactivation. Therefore, the L-index(30) might be less useful as a predictor of CMV reactivation. With regard to lymphocyte subset analysis, $CD4^+$ cell count was significantly lower in CMV-AG ≥ 3 group than in CMV-AG < 3 group. According to the study by Kim et al., the incidence of CMV reactivation was not affected by $CD4^+$ cell count at 3 months after transplantation (28). However, our result suggested that $CD4^+$ cell count at the early phase after HSCT might play an important role in preventing CMV reactivation. The ability of the L-index(30) for predicting CMV reactivation was considered to be almost the same as that of $CD4^+$ cell count at day 30 because the area under the curve of these two indexes were almost equal in the ROC curve analyses. Our result also showed that $CD8^+$ cell count on day 90

after HSCT in CMV-AG ≥ 3 group was significantly higher than that in CMV-AG < 3 group, which agreed with the result reported by Heining et al. (9), because CMV reactivation enhanced immune function and significantly improved $CD8^+$ T cell recovery (9, 11).

Advanced age has been known as a risk factor of CMV disease (29, 30). The present study showed that patients' age older than 41 years old was identified as an independent significant factor on CMV reactivation, which suggested that patients' age affect not only CMV diseases but also reactivation. Other well-recognized risk factors for CMV infection include seropositivity for CMV before HSCT, unrelated donor status, development of aGVHD, and corticosteroid use (31-33). In this study, all patients in CMV-AG ≥ 3 group were donor or/and recipients who were seropositive for CMV, though there was no significant difference as compared with those in CMV < 3 group. The percentage of unrelated donor status was significantly higher in CMV-AG ≥ 3 group than in CMV < 3 group in univariate analysis, but it was not identified as an independent factor in multivariate analysis. The development of acute GVHD and corticosteroid use did not differ between the 2 groups.

In conclusion, our present study showed that both the intensity and the duration of lymphopenia in early phase after HSCT evaluated as the L-index(30) were significantly associated with CMV reactivation. However, L-index(30), which became available only after 30 days from transplantation, might be less useful as a predictor of CMV reactivation.

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310-318.

Table 1 Clinical and epidemiological characteristics of the study patients

Clinical and epidemiological factors	Total (n = 50)	CMV-AG ≥ 3 (n = 30)	CMV-AG < 3 (n = 20)	P value
Median age, years (range)	41 (15-63)	47 (15-61)	36 (15-63)	0.062
Sex male/female	27/23	14/16	13/7	0.203
Underlying disease				0.460
Acute myeloblastic leukemia	19 (38%)	10 (33%)	9 (45%)	
Acute lymphoblastic leukemia	5 (10%)	2 (7%)	3 (15%)	
Lymphoma	4 (8%)	4 (13%)	0	
Adult T cell leukemia/lymphoma	6 (12%)	4 (13%)	2 (10%)	
Myelodysplastic syndrome	5 (10%)	3 (10%)	2 (10%)	
Aplastic anemia	8 (16%)	6 (20%)	2 (10%)	
Others	3 (6%)	1 (3%)	2 (10%)	
Lymphoid malignancy	15 (30%)	10 (33%)	5 (25%)	0.529
Conditioning regimen				0.556
Myeloablative regimen	30 (60%)	17 (57%)	13 (65%)	
Reduced-intensity regimen	20 (40%)	13 (43%)	7 (35%)	
Fludarabine containing	20 (40%)	13 (43%)	7 (35%)	0.556
ATG containing	7 (14%)	5 (17%)	2 (10%)	0.506
Alemtuzumab containing	4 (8%)	4 (13%)	0	0.083
Donor Related/Unrelated	29/21	14/16	15/5	0.047
HLA Match/Mismatch	35/15	20/10	15/5	0.350
Graft				0.704
BM	30 (60%)	18 (60%)	12 (60%)	
PBSC	19 (38%)	11 (37%)	8 (40%)	
CB	1 (2%)	1 (3%)	0	
GVHD prophylaxis				0.032
CsA300 + sMTX	19 (38%)	15 (50%)	4 (20%)	

CsA500 + sMTX	30 (60%)	14 (47%)	16 (80%)	
FK506 + sMTX	1 (2%)	1 (3%)	0	
CMV serostatus				0.162
Donor and recipient seronegative	2 (4%)	0	2 (10%)	
Donor or/and recipient seropositive	48 (96%)	30 (100%)	18 (90%)	
Acute GVHD				
Grade II–IV	16 (32%)	10 (33%)	6 (30%)	0.804
Grade III–IV	7 (14%)	5 (17%)	2 (10%)	0.506
Corticosteroid use	11 (22%)	5 (17%)	6 (30%)	0.265
Chronic GVHD				
Total	18 (36%)	9 (30%)	9 (45%)	0.399
Extensive type	13 (26%)	7 (23%)	6 (30%)	0.368

CMV-AG, cytomegalovirus antigenemia; BM, bone marrow; PBSC, peripheral blood stem cell; CB, cord blood; GVHD, graft-versus-host disease; CsA, cyclosporine A; CsA300, target CsA concentration of 300 ng/ml during continuous intravenous infusion; CsA500, target CsA concentration of 500 ng/ml during continuous intravenous infusion; sMTX, short-term methotrexate.

Table 2 Factors associated with high L-index(30) and L-index(100) values

	Median value	P value
L-index(30)		
Univariate analysis		
Female sex (vs. male)	22,264 vs. 18,950	0.048
Non-lymphoid disease (vs. lymphoid malignancy)	21,684 vs. 16,552	0.009
ATG-containing regimen (yes vs. no)	22,299 vs. 19,268	0.157
Alemtuzumab-containing regimen (yes vs. no)	24,956 vs. 19,461	0.001
Unrelated donor (vs. related donor)	22,264 vs. 19,268	0.023
HLA-mismatched donor (vs. HLA-matched donor)	22,962 vs. 19,038	0.015
BMT (vs. PBSCT)	21,953 vs. 17,110	0.089
Multivariate analysis		
HLA-mismatched donor (vs. HLA-matched donor)		0.010
Female sex (vs. male)		0.019
Non-lymphoid disease (vs. lymphoid malignancy)		0.042
L-index(100)		
Univariate analysis		
Female sex (vs. male)	34,406 vs. 23,711	0.142
Non-lymphoid disease (vs. lymphoid malignancy)	34,935 vs. 16,757	0.017
ATG-containing regimen (yes vs. no)	45,394 vs. 28,455	0.069
Alemtuzumab-containing regimen (yes vs. no)	52,621 vs. 27,872	0.020
HLA-mismatch donor (vs. HLA-matched donor)	39,535 vs. 23,711	0.008
BMT (vs. PBSCT)	31,249 vs. 19,933	0.134
Grade III–IV aGVHD (yes vs. no)	45,937 vs. 28,185	0.078
Multivariate analysis		
Grade III–IV aGVHD (yes vs. no)		0.003
Alemtuzumab-containing regimen (yes vs. no)		0.002
Non-lymphoid disease (vs. lymphoid malignancy)		0.003

ATG, anti-thymoglobulin; BMT, bone marrow transplantation; PBSCT, peripheral blood stem cell transplantation; GVHD, graft-versus-host disease.

Table 3 Association between lymphopenia indexes and CMV infection

Lymphopenia indexes	Total (n = 50)	CMV-AG ≥ 3 (n = 30)	CMV-AG < 3 (n = 20)	P value
L-index(30), median (range)	21,081 (8,757-26,512)	22,030 (10,062-26,512)	19,038 (8,757-24,527)	0.050
L-index(30)				
≥ 22,318	18	15	3	0.016
< 22,318	32	15	17	
L-index(100), median (range)	29,987 (8,757-65,789)	31,453 (10,062-65,789)	29,585 (8,757-60,624)	0.476
ALC, median (range)				
Day 30	366 (21-3,774)	326 (21-1,453)	418 (58-3,774)	0.607
Day 60	598 (52-4,308)	589 (106-2,705)	630 (52-4,308)	0.843
Day 90	754 (0-5,261)	859 (0-5,261)	724 (67-2,822)	0.411
$CD4^+$ cell count, median (range)				
Day 30	97 (4-902)	77 (4-587)	174 (17-902)	0.023
Day 60	130 (7-702)	60 (7-544)	176 (91-702)	0.263
Day 90	193 (6-1,005)	189 (6-1,005)	289 (30-401)	0.739
$CD8^+$ cell count, median (range)				
Day 30	142 (5-2,264)	86 (5-1,027)	170 (41-2,264)	0.189
Day 60	295 (22-3,132)	215 (22-1,563)	300 (155-3,132)	0.441
Day 90	383 (25-2,994)	622 (25-2,994)	205 (28-383)	0.041

CMV-AG, cytomegalovirus antigenemia; ALC, absolute lymphocyte count.

Table 4 Factors associated with CMV infection

A. Univariate analyses			
Factors	CMV-AG ≥ 3 (n = 30)	CMV-AG < 3 (n = 20)	P value
Age			
< 41	10	13	0.043
≥ 41	20	7	
Donor			
Related	14	15	0.047
Unrelated	16	5	
GVHD prophylaxis			
CsA300 + sMTX	15	4	0.032
CsA500 or FK + sMTX	15	16	
L-index(30)			
≥ 22,318	15	3	0.016
< 22,318	15	17	

B. Multivariate analyses			
Factors	Odds ratio	95% CI	P value
Age	4.45	1.190 - 16.60	0.0263
L-index(30)	6.71	1.470 - 30.70	0.0141

CsA300, target CsA concentration of 300 ng/ml during continuous intravenous infusion; CsA500, target CsA concentration of 500 ng/ml during continuous intravenous infusion; sMTX, short-term methotrexate.

Figure legends

Fig.1. Area over the lymphocyte curve (L-index) in a hypothetical patient. Lymphopenia (absolute lymphocyte count; ALC < 700/μl) developed 7 days before transplantation in the patient. The L-index (Ae–Ao) was calculated as the difference between the expected lymphocyte area (shaded area, Ae) and the observed area under the curve (striped area, Ao), which was calculated by the trapezoidal method. If the area under the lymphocyte curve until day 30 is 4,550, the L-index(30) = 37 × 700 – 4,550 = 21,350. In the same way, if the area under the lymphocyte curve until day 100 is 43,928, the L-index(100) = 107 × 700 – 43,928 = 30,972.

Fig.2. Receiver operating characteristic (ROC) curves comparing L-index(30) with $CD4^+$ cell count at day 30. The area under the ROC curve was 0.68 and 0.71 for the L-index(30) and $CD4^+$ cell count at day 30, respectively. The sum of the sensitivity and specificity reached the maximum when the thresholds for the L-index(30) and CD4+ cell count at day 30 were 22,318 and 59, respectively. With the use of these cut-off values, the sensitivity and specificity for predicting CMV infection were 50.0% and 85.0%, and 47.8% and 88.9%, respectively.

Fig.1. Area over the lymphocyte curve (L-index) in a hypothetical patient

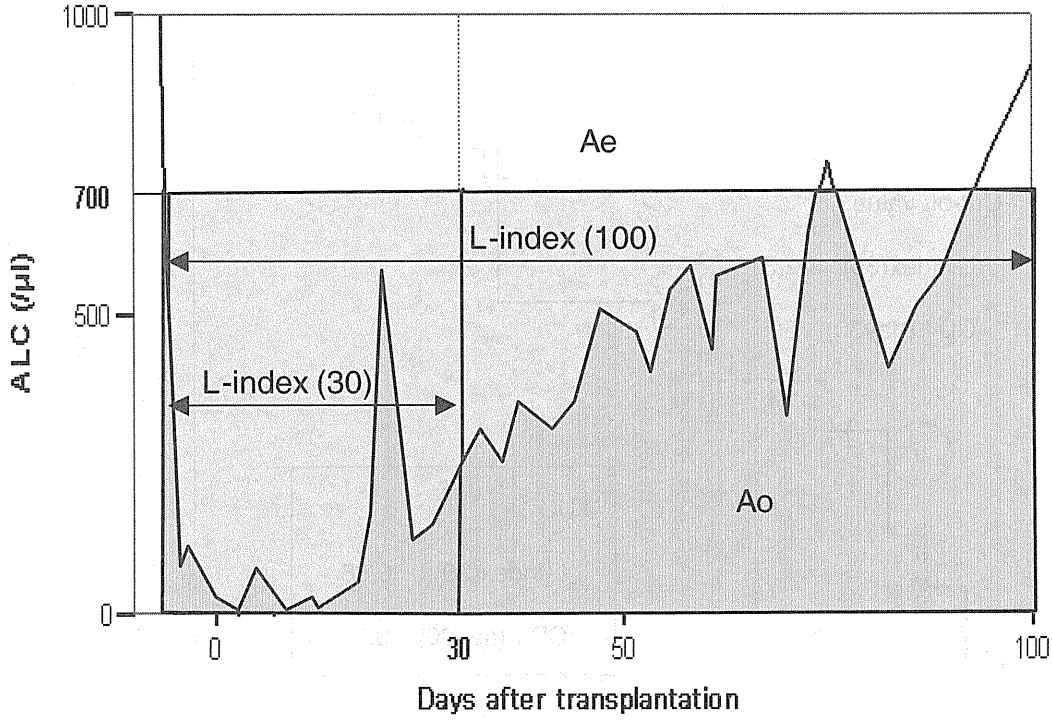
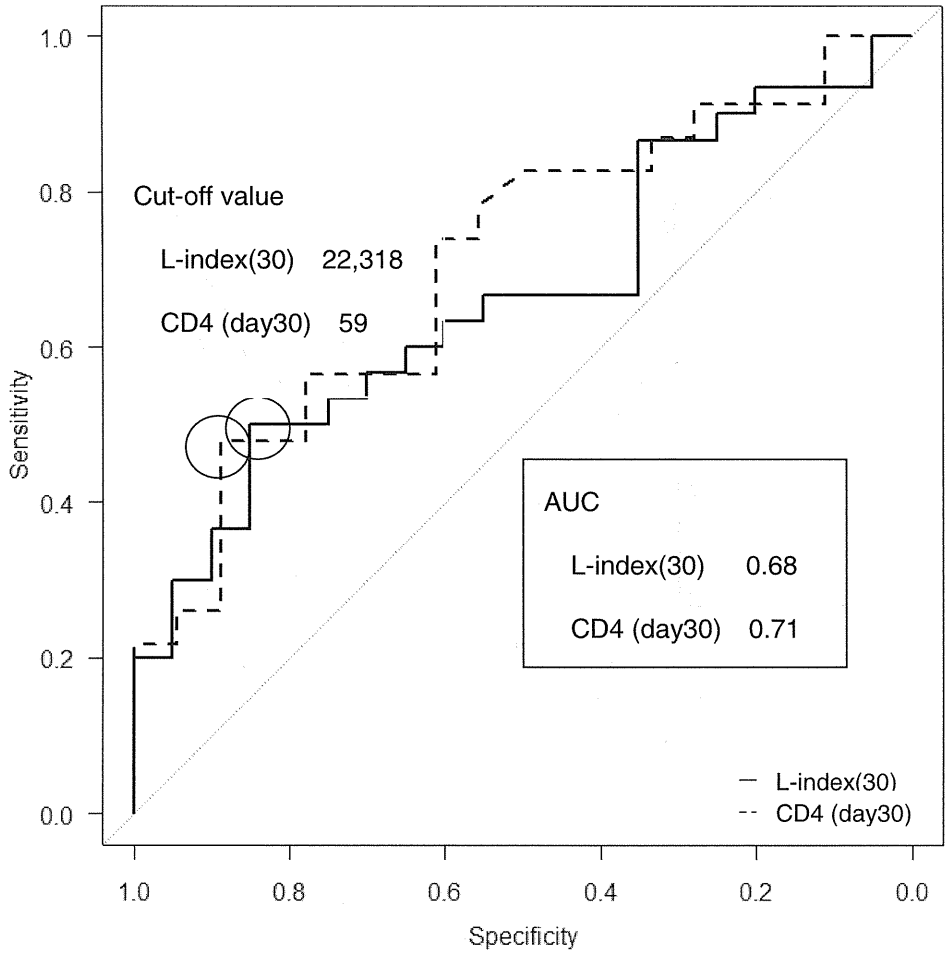


Fig.2. Receiver operating characteristic (ROC) curves comparing L-index(30) with $CD4^+$ cell count at day 30.



4. 予防的血小板輸血の適応と用量について

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key words　platelet transfusion, prophylactic transfusion, therapeutic transfusion, trigger value, inflammation, bleeding

動　向

造血器疾患およびそれに関連した治療や，化学療法に関連した血小板減少時の血小板輸血については，血液学的側面と臨床輸血医学的側面から考察する必要がある．血小板輸血の考え方には，予防的血小板輸血と治療的血小板輸血がある[1]．出血の重症度分類には，しばしばWHOの重症度分類[2]が用いられるが，微小な出血の頻度は，血小板数に逆相関するものの，重篤な出血の頻度は，少なくとも血小板数5,000～10,000/μl以上では，血小板数に関連しないことが示されている[3,4]．また，血小板以外の止血系の障害のほかにも，感染，炎症，急激な血小板数の変化，薬剤といった出血を助長する因子が提示されている[2]．一方で，不必要な血小板輸血は輸血不応状態をもたらしやすいこと[5]，TRALI (transfusion related acute lung injury)[6]やアナフィラキシーなど重篤な副作用を合併しやすいこと，随時使用が困難なことを考慮する必要がある．さらに，近年，ADAMTS13の，先天性欠乏もしくは抗体産生による後天的欠乏により血小板血栓を生じる血栓性血小板減少性紫斑病（TTP）[7]や，抗ヘパリン/PF4複合体抗体によりヘパリン投与時に生じる，ヘパリン起因性血小板減少症（HIT）[8]は，予防的血小板輸血は禁忌の病態であるとされている．このような中で，現在一般的には，化学療法や造血細胞移植による，急性期の血小板減少に対しては，血小板数10,000/μlをトリガーとする予防的輸血が，また再生不良性貧血や骨髄異形成症候群の慢性期においては，治療的血小板輸血が推奨されている[9]．わが国のガイドラインでは，予防的血小板輸血は，10,000～20,000/μlをトリガーとしており[10]，再生不良性貧血や骨髄異形成症候群では，5,000/μlがトリガーとなっている[10]．これは，わが国における，血小板製剤の供給状況を反映した数字と考えられる．

至適血小板輸注量については，至適量自体が決まっていない上に，濃厚血小板製剤の規格自体が，国によって異なるため，検討が困難である．一般的に，1回輸注量が少なくなれば，総輸注量は減少するが，輸注回数は増加する[1]．至適輸注量を出血事象で評価した場合，比較的少ない1回輸注量で問題ないことが示されつつあり，わが国での10単位製剤に相当する製剤の使用で問題ないことが示されている[11]．